



EFFECT OF DIETARY INCLUSION OF PAPAYA (*CARICA PAPAYA*) SEED AND PULP MEAL ON THE GROWTH OF NILE TILAPIA (*OREOCHROMIS NILOTICUS*) REARED IN CONCRETE TANK IN TOGO

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ABSTRACT: A study on the growth performance of *Oreochromis niloticus* was carried out, from June to August 2020, with diets incorporating papaya seeds and pulp meal (*Carica papaya*). Therefore, 600 mono-sex fry male of Nile tilapia (average weight ≈ 2 g) were distributed in 12 concrete tanks (0.6 m^3) at a density of 50 fishes per tank. They were fed four isoprotein diets (32% PB) in triplicate including a commercial feed (CF), a local feed containing 30% palm kernel meal cooked in water for one hour (LF) to which 5g of seeds powder (SF) was added or 4g of pulp powder (PF) of papaya/kg of diet, for 70 days. Then, after a 2-day fasting period, 5 fish from each batch were dissected in order to assess growth through the study of digestive transit. At the end of this trial, encouraging results were obtained. Survival rate was generally in the order of 90% for all batches. The best zootechnical performances were observed in the fish fed with SF and PF (FCR = 1.78 and 1.99; PER = 1.72 and 1.51; SGR = 3.48 and 3.56 %/d). In addition, feed transit was slower in all intestinal compartments of fish of these batches. Incorporating papaya products is a promising option for the growth of *O. niloticus*.

KEYWORDS: Nile tilapia, Papaya, Feed, Growth, Digestive transit.



INTRODUCTION

Global aquaculture production stood at 82.1 million tons in 2018. Despite its aquaculture potential, Africa's aquaculture contribution is insignificant and estimated at around 3% of this production. It should be noted that the constraints to the development of aquaculture in Africa result from several interconnected factors, such as a population which grows at a rate higher than the supply of fish, the stagnation of the production of fish due to the pressure exerted on capture fisheries resources and an underdeveloped aquaculture sector with very high cost and difficulties in obtaining good quality feed (FAO, 2018).

In Togo, the annual production of fish, both from fishing and aquaculture, is around 25,500 tons while the needs are estimated at over 95,000 tons (DPA, 2017), which means that 75% comes from imports. It is with a view to reducing the significant imports from certain countries in Africa and Asia that Togo has opted for the promotion of fish farming (DPA, 2017). The increase in production will go through an optimization in the improvement of production systems based on a feed cost adapted to the main species raised as well as the Nile tilapia, *Oreochromis niloticus*. However, in aquaculture, feed represents about 60 to 70% of the production cost (FAO, 2015). It then becomes necessary to carry out studies on the development and production of high-performance foods at low cost, which involves the promotion of local by-products. Many trials of incorporating agro-industrial raw materials such as palm kernel cake (Ng, 2004; Adjanké, 2018), dried moringa leaves (Richter et al., 2003) or papaya seeds (Nuushona, 2019) in the feed were conducted in order to improve the zootechnical performance of tilapia. It is with this goal we have chosen to work on papaya products which production abounds nationally. In addition, previous works have shown the effects of papaya seed powder, even at low doses, on growth in this species (Farrag et al., 2013; Ugonna et al., 2018). But by what phenomenon this effect can be possible on the growth of tilapia? Two studies are possible such as digestibility and digestive transit tests. The study of digestive transit makes it possible to assess the contact time of nutrients with the intestinal wall of fish (Moreau, 2009) and constitutes the starting point of the nutritional process because it is the food digested and absorbed, which is made available for cell metabolism (Bergot & Brecque, 1983). This study was initiated to improve Nile tilapia production because the results of digestive transit studies are an essential for aquaculture feed manufacturers. This will involve evaluating the effect of the incorporation of papaya seed or pulp meal on the production parameters in tilapia and then analysing the effect of this dietary inclusion on the digestive transit of tilapia.

MATERIALS AND METHODS

Experimental Procedure

The study was carried out from June to August 2020 at the Aquaculture Research and Development unit (REDAQ) in the agricultural experiment station of the University of Lome in Togo. This experiment, which lasted 70 days, involved 600 monosex male tilapia fry distributed at a density of 50 fries per tank containing 250 litres of water in triplicate. So 12 concrete tanks of 600L, operating in open circuit with periodic renewal of the water were used. Each tank is individually supplied with water by gravity from a water tower and air using a brand air compressor. The water from the Togolese Water Company (TdE) is used by the SEAL. At the unit level, this water is stored in a reservoir and then transported through a



submersible pump into the breeding circuit. This system can support a load capacity of 40 kg/m³ of fish according to the work of Melard (1986) and Akinwole et al. (2014).

Six ingredients such as corn, palm kernel cake cooked in water for an hour, fishmeal, roasted soybeans, vitamin mineral concentrate and palm oil were used for the present work to make the local feed. Note that the palm kernel cake was cooked in water for 1 hour at a temperature of 100-102°C and then dried before incorporated in feed made in pelleted form according to Adjanke (2018). The granules formed were dried in the sun. They were then recovered, ground and then stored for use during the test (Ble et al., 2008).

The monosex male tilapia fry were fed with 4 isoprotein diets with 32% crude protein, at the rate of 12% of their biomass during the first month of the test then at the rate of 8% until the end of the test (Toguyeni, 1996), 5 times per day. The diets are a commercial feed Ranaan (CF), a simple local feed (LF), containing 30% of palm kernel meal cooked in water for 1 hour, and to which is incorporated 5g of papaya seed meal (SF) or 4g of papaya pulp meal (PF) per kilogram of food (Table 1). The papaya seeds and pulp were pre-dried in a solar dryer for 12 hours at 42°C and were then dried in an oven at 50°C for at least 24 hours for the seeds and 48 hours for the pulps. The dried products were finely ground and the resulting meal was mixed with other ingredients to obtain tested diets.

Table 1: Feed formula of local feed for tilapia fry (% dry weight).

Ingredients	LF*	PF	SF
Corn	14	14	14
Palm kernel meal cooked in water	30	30	30
Fish meal 50 (%)	42	42	42
Roasted soybeans	10	10	10
Vitamin-mineral complex	2	2	2
Palm oil	2	2	2
Papaya pulp	0	0,4	0
Papaya seed	0	0	0,5
TOTAL	100	100	100

* Source: Adjanke (2018). Its composition is 31.88% crude protein, 11.72% fat and 8.96% fibre.

LF: local feed; PF: local feed incorporating 4g of papaya pulp meal; SF: local feed incorporating 5g of papaya seed meal. The composition of the standard food according to the manufacturer's data sheet is 32% crude protein, 5% fat and 4% fibre. The physicochemical parameters of the water (temperature, dissolved oxygen, pH) were regularly monitored. At the end of this period, the fish were deprived of food for 2 days. On the 3rd day, some were refilled once again (08 H) before depriving them of food again. A sample of 5 fish per lot was euthanized in a solution of clove (6g/10L of water) and dissected at the start and after 1, 4, 8,



12 and 24 hours of fasting to measure the intestinal parameters according to the method of Hocking et al. (2004), adapted to fish.

Determination of the Digestive Transit

After the dissection, the digestive tract was removed and measured. The digestive transit was evaluated from the contents weight of the different digestive tract compartments (stomach, proximal and distal guts). The weight of the digestive tract contents was obtained by the difference between the compartments of the fed fish and those of the unfed fish after the period of fasting.

Production Parameters Calculated

To estimate the growth of the fish during the test and to characterise the efficiency of use of the tested feed, various production parameters and the following indices were calculated (Table 2).

Table 2: Production parameters and formulas used for the study.

Production parameters	Formulas
(DWG) : Daily Weight Gain (g/d)	$DWG = (W_f - W_i) / \text{test duration (days)}$
(SGR) : Specific Growth Rate (%/d)	$SGR = 100 * (\ln W_f - \ln W_i) / \text{test duration (days)}$
(FCR) : Food Conversion Ratio	$FCR = Q / [(B_f + B_d) - B_i]$
(VI) Voluntary ingested (%/d)	$VI = : 100 * Q / ((B_i + B_f) / 2) / \text{test duration (days)}$
(PER) : Protein Efficiency Ratio	$PER = [(B_f + B_d) - B_i] / (Q \times \text{diet protein})$
Survival (%)	$\text{Survival} = 100 \times (N_f / N_i)$

Wi and Wf: initial and final weight (g); Bi, Bd and Bf: initial, dead and final biomass (g); Q: cumulative quantity of food distributed (g); Ni and Nf: initial and final fish number

Statistical Analysis

Data were analysed by one-way analysis of variance (ANOVA I). The LSD Fischer test allowed homogeneous groups means discrimination. Differences were considered significant at 5% level. Statistical analyses were performed using STATISTICA 5.1 program (Stat Soft, Inc.).

RESULTS

Physicochemical Parameters

The average values of the physicochemical parameters of the water in the tanks were respectively 26.76 ± 1.09 °C, 7.27 ± 0.17 and 5.52 ± 1.08 mg/L for the temperature, pH and dissolved oxygen. Water ammonia (0 mg/L), nitrite (3 mg/L) and nitrate (22.50 ± 3.54 mg/L) content were checked once a week. Throughout the experiment, the average values of the various physicochemical parameters of the water (temperature, pH and oxygen) recorded remained within the tolerance limits of the species.



Effect of dietary inclusion of papaya seed and pulp meal on the zootechnical performance of fish

The growth response of *O. niloticus* fed different experimental regimes is presented in Table 3.

Table 3: Effect of dietary inclusion of papaya seed or pulp meal on zootechnical performance in juvenile *O. niloticus* reared after 70 days in tanks

Parameters	Treatments			
	CF	LF	SF	PF
IBW (g)	2,02 ± 0,07a	2,08 ± 0,07a	2,13 ± 0,04a	1,92 ± 0,06a
FBW (g)	21,72 ± 0,76bc	20,47 ± 0,20b	24,43 ± 0,34a	23,27 ± 0,42ac
DWG (g)	19,70 ± 1,50b	18,39 ± 0,29b	22,30 ± 1,58a	21,34 ± 1,34a
SGR (%/j)	3,39 ± 0,13ab	3,27 ± 0,07b	3,48 ± 0,04a	3,56 ± 0,11a
VI (%/j)	4,93 ± 0,33a	5,19 ± 0,10a	4,31 ± 0,05b	4,89 ± 0,08a
FCR	2,04 ± 0,16b	2,22 ± 0,06a	1,78 ± 0,02c	1,99 ± 0,05b
PER	1,49 ± 0,12b	1,39 ± 0,03b	1,72 ± 0,03a	1,51 ± 0,03ab
S (%)	94,00 ± 3,46a	94,00 ± 2,00a	92,67 ± 1,15a	88,67 ± 8,08a

CF: commercial food; LF: local food containing 30% palm kernel cake cooked in water for one hour; SF: local food containing 5 g of papaya seed meal; PF: local food containing 4 g of papaya pulp meal. IBW: initial body weight; FBW: final body weight; DWG: weight gain; SGR: specific growth rate; VI: voluntary ingestion; FCR: feed conversion ratio; PER: protein efficiency ratio; S: survival. The values represent the mean ± the standard deviation. Values in the same row with different letters are significantly different ($p < 0.05$).

The average final weights after 70 days of rearing varied from 20.47 ± 0.20 g to 24.43 ± 0.34 g as illustrated in Figure 1. The average final weights of the fish fed with the diets containing papaya seed or pulp meal were significantly higher ($P < 0.05$) than those of the other batches; the two batches being statistically similar ($P > 0.05$).

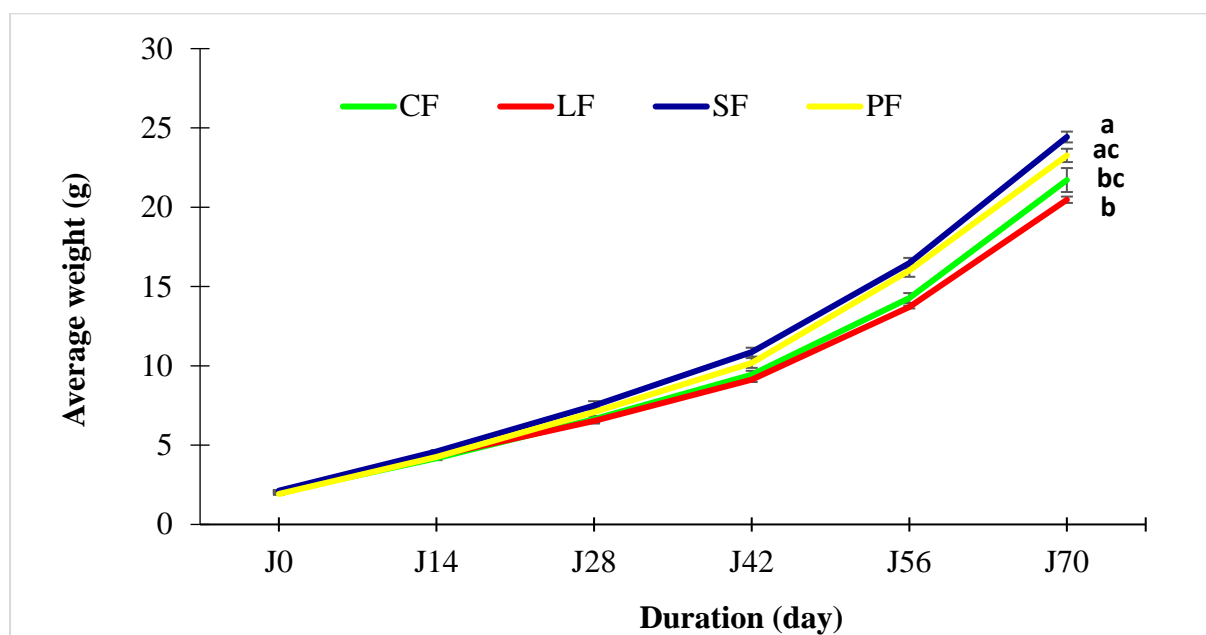


Figure 1: Mean growth of fingerlings fed with experimental diets

CF: commercial food; LF: local feed; SF: local feed containing 5 g of papaya seed meal; PF: local feed containing 4 g of papaya pulp meal. Curves with the same letters are not significantly different ($p > 0.05$).

The average weight gained at the end of the experiment varied between 18.39 ± 0.29 g and 22.30 ± 1.58 g and the statistical analysis revealed a significant difference ($p < 0.05$) between the different treatments (Table 3). The average weights of the fish from the AS0 and AT0 batches are identical to each other and significantly lower than those from the SF and PF batches which exhibited the best growth performance ($P < 0.05$). The average values of the specific growth rates (TCS) according to the different treatments are given in Table 5. The rates varied between $3.27 \pm 0.07\%$ / d and $3.56 \pm 0.11\%$ / d. No significant difference ($P > 0.05$) was observed between the fish from the batches fed with the local feed containing 5g of papaya seed meal (SF) and those of the local feed containing 4g of papaya pulp meal (PF). On the other hand, a significant difference is observed between these batches and the batch fed with the local food containing 30% of palm kernel meal cooked in water for one hour (LF). The main results obtained on the use of the feed were the voluntary intake (IV), the feed conversion rate (FCR) and the protein efficiency ratio (PER).

The voluntary intake (VI) values varied from $4.31 \pm 0.05\%$ / d to $5.19 \pm 0.10\%$ / d (Table 3). The lowest value is observed in the fish of the SF batch which is statistically different from the values of the other batches ($p < 0.05$). Regarding the feed conversion rate (FCR) of the different batches, the highest value (2.22 ± 0.06) was recorded in the fish of the LF batch and the lowest ($1.78 \pm 0, 02$) for the fish of the SF batch ($p < 0.05$). No significant difference was observed between the PF and CF lots ($p > 0.05$). The mean values of the protein efficiency ratio (PER)



according to the treatments carried out were significantly higher in the fish from the SF and PF lots compared to the fish from the CF and LF ($p < 0.05$).

The average survival rates at the end of the experiment varied between $88.67 \pm 8.08\%$ and $94 \pm 3.46\%$ (Table 3) with no significant difference between the survival rates of the different lots ($P > 0.05$).

Effect of the incorporation of papaya seed powder and pulp in food on food transit

Weight of stomach contents after fasting

The change in the average weight of the stomach contents after the fasting period in the fish is shown in Figure 2. A significant decrease in the average weight of the stomach contents is observed in all the batches during the 24 hours ($p < 0.05$). One hour after the fasting phase, the weight of stomach contents was similar in all batches, but from the 4th to 12th hour, significant differences appeared between batches. Thus, the stomach contents of the CF and SF batches were significantly greater than those of the LF and PF batches ($p < 0.05$). On the other hand, beyond the 12th hour, the weight of the stomach contents no longer showed a significant difference between the four batches ($P > 0.05$).

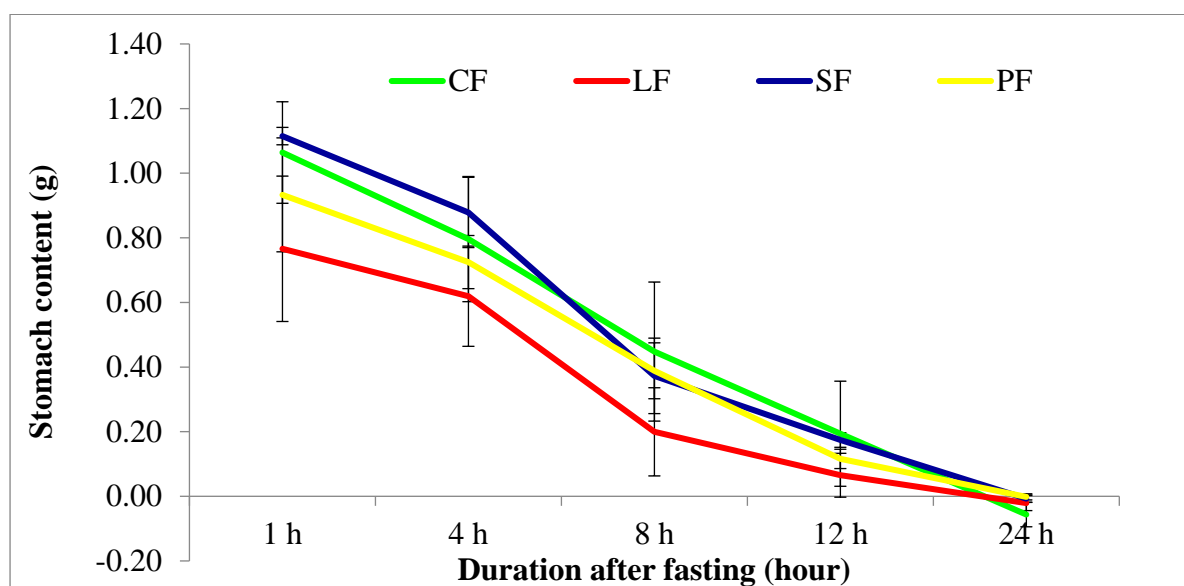


Figure 2: Evolution of the average weight (g) of the contents of the stomach of fish lots fed after fasting

CF: commercial food; LF: local feed; SF: local feed containing 5 g of papaya seed meal; PF: local feed containing 4 g of papaya pulp meal.



Weight of the contents of the proximal intestine after fasting

During the first eight hours after the fast, the content of the proximal intestine gradually increased in all batches (Figure 3). This increase is similar for all batches ($p > 0.05$). On the other hand, a decrease in the content was observed beyond the 8th hour until the end and this was more important in the fish of the LF and PF batches compared to the fish of the CF and SF batches ($p < 0.05$).

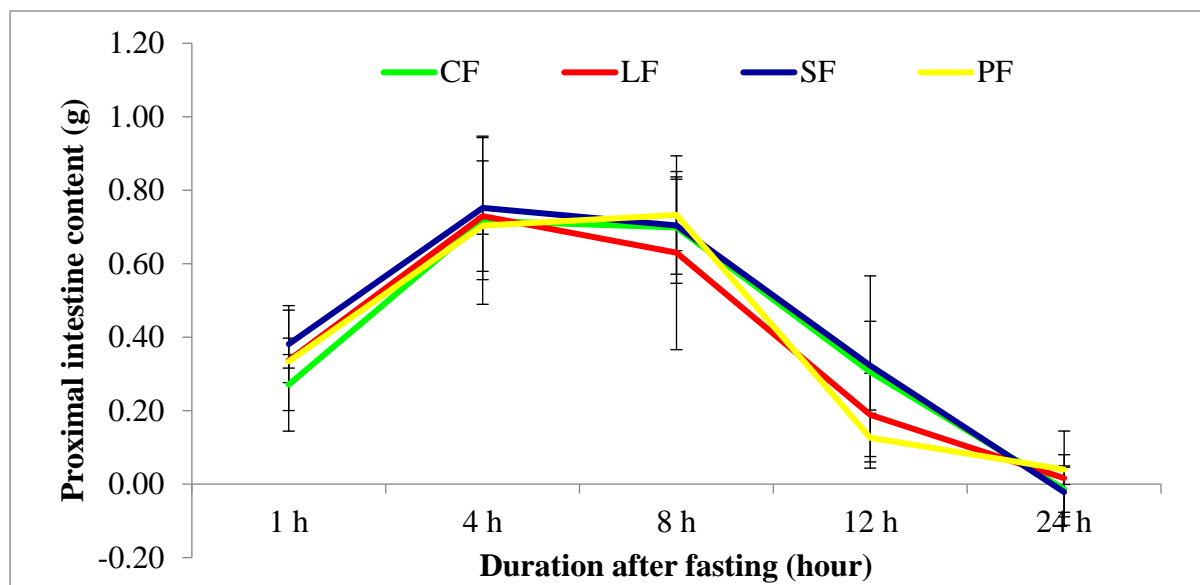


Figure 3: Evolution of the average weight (g) of the contents of the proximal gut of fish lots fed after fasting

CF: commercial food; LF: local feed; SF: local feed containing 5 g of papaya seed meal; PF: local feed containing 4 g of papaya pulp meal.

Weight of the contents of the distal intestine after fasting

The change in the mean weight of the contents of the distal intestine after the fast is shown in Figure 4. This weight significantly increased in a similar way for all batches of fish up to the 8th hour ($p > 0.05$). On the other hand, beyond this hour, a similar decrease in content is noted in all the batches ($p > 0.05$).

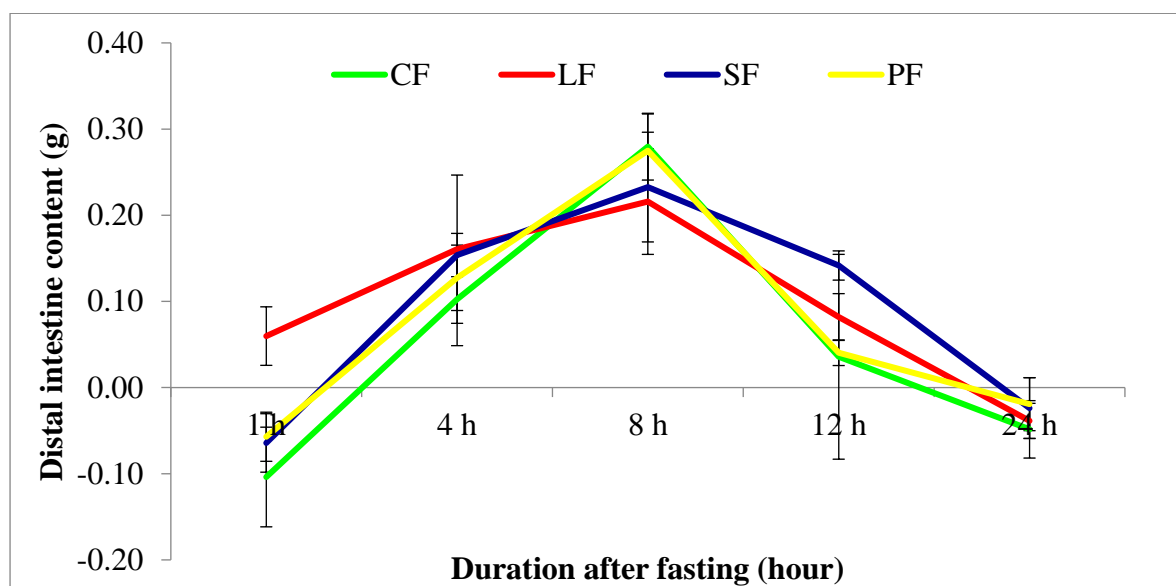


Figure 4: Evolution of the average weight (g) of the contents of the distal gut of fish lots fed after fasting

CF: commercial food; LF: local feed; SF: local feed containing 5 g of papaya seed meal; PF: local feed containing 4 g of papaya pulp meal.

DISCUSSION

Impacts of adding papaya seed and pulp meal on zootechnical parameters

The analysis of the results of the experiment shows that the addition of papaya seed or pulp meal did not have a negative influence on the fish. In fact, the survival rates recorded during the experiment are very high and do not show any difference between the batches that were fed with a food containing papaya seed or pulp meal and the control batches. The results show that these additives did not negatively modify the physicochemical parameters of the water (temperature, pH, and dissolved oxygen). Indeed, no significant difference was observed between the basins of the different batches, which were in the ranges of the optimal values of tilapia rearing conditions (Melard, 1986; Melard et al., 1989; Baroiller & Toguyeni, 1996), thus confirming the good breeding conditions.

Regarding the impact of treatments on weight growth, the results show better growth of batches supplemented with papaya seed or pulp meal from the 28th day of breeding. These differences in growth observed between the different batches could be linked either to a difference in the voluntary intake or to the efficiency of feed use. However, observation of fish during feed distribution indicated identical apparent consumption of feed distributed, although fish from lots fed the CF and LF diet appeared to consume more. In view of the results obtained, the voluntary intake would therefore be influenced by the type of food. Although in appearance, none of the foods tested seemed to have a repellent character. However, analysis of the data showed that food intake would decrease with an increase of dietary inclusion rate of papaya



seed and pulp meal. Similar results have already been shown in the African catfish, *Clarias gariepinus*, with a diet based on papaya leave, the voluntary ingestion of fish fed with these foods decreasing considerably with the level of inclusion (Olanyi & Salau, 2013).

However, it should be noted that in the same species, seed meal was used as a substitute for fishmeal and gave good results up to 80% substitution rate (Irabor et al., 2016). It has also been shown that the food intake of fish depends on their sensory capacity to find the food, to capture and to ingest it, but also on their physiological state which determines the way they digest and process the food (Kestemont & Baras, 2001). This better growth of the SF and PF lots could be linked to a better feed conversion (FCR). The FCR is a very important parameter in aquaculture which allows fish farmers to estimate the quantity of feed necessary in order to maximise profitability (Gourene et al., 2002). In the present study, the values of the feed conversion rate obtained are within the ranges of values reported by several authors (1.7 to 3) but they are higher than those reported by a recent study with FCR varying between 1.26 at 1.60 (Nuushona, 2019). However, it should be emphasised that the FCR can be influenced by several factors, such as temperature, stocking density, feeding methods, among others. It was by taking these data into account that the feeding method was set and could therefore partly explain the differences observed. In fact, the daily ration was distributed in 5 meals, which would have enabled them to better value the diet, particularly those made with better use of those containing papaya seed or pulp, in a well-stocked controlled system (Tan et al., 2018; Zea Biue et al., 2019).

Impacts of papaya seed and pulp meal on food digestibility

The analysis of the results of the food transit showed that it was slower in all the intestinal compartments of fish fed diets containing papaya seed or pulp meal (SF and PF) compared to the fish fed diets CF and LF. This slowness will be linked to the presence of papaya seed or pulp meal in these foods. Medale and Burel (2014) have shown that certain food compounds, in particular fibres, increase the speed of intestinal transit by stimulating peristalsis, thus reducing the contact duration of food components with digestive enzymes and cells of the intestinal mucosa. Richter et al. (2003) have also shown that compounds rich in fibre can act by modifying the activity of digestive enzymes through absorption and immobilisation phenomena. The incorporation of products from papaya, which contains a proteolytic enzyme namely papain, would have favoured the bioavailability of the nutrients contained in the SF and PF foods, which would have led to an improvement in their assimilation by a relative reduction in their transit speed as shown by Farrag et al. (2013) during their work.

In addition, the duration of food evacuation in the digestive tract of fish was longer and this could be explained by the adaptation of the fish to fasting on the one hand, and to the studying transit method used on the other hand, proposed by Moreau in 1988 and Riche et al. in 2004. The result of this transit is reflected in the feed conversion level (FCR), the relatively interesting values of which seem to agree with the weight growth performance of the fish. It should also be noted that ingredients of plants may contain anti-nutritional factors (ANFs) which inhibit the activity of digestive enzymes and increase the speed of intestinal transit (Medale & Burel, 2014).

In addition, the data on the protein efficiency coefficient show a better valuation of foods containing papaya seeds or pulp meal compared to the control food LF. The low CEP of LF could be attributed to a lower digestibility of its nutrients due to the high fibre content and the



absence of enzymes such as papain from papaya as suggested by the work of Kaushik et al. (1993). Thus, the data suggest that the dietary inclusion of papaya seed or pulp meal in tilapia feeds would improve nutrient utilisation and reduce the adverse effects of anti-nutritional factors present in high levels in the isolated ingredients (Farrag et al., 2013). The good growth observed in fish from batches fed with diets containing papaya seed or pulp meal could also come from the induced capacity of use of complex polysaccharides and cellulose by this species to cover its energy needs (Viola & Arieli, 1983).

The products from papaya, incorporated in the food, slowed the transit of this food in the digestive tract of the tilapia, which allowed the micronutrients to stay longer in contact with the cells of the intestinal epithelium and thus favoured their absorption. This resulted in good growth of the fish fed with the feed containing these products. All of these results confirm our hypotheses that the incorporation of papaya seed powder and pulp in the food improves digestive transit and growth in Nile tilapia.

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