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Research Article

Influence of Anthocyanins from Katiola Purple Corn on Growth, Body Composition and Health of Nile Tilapia (*Oreochromis Niloticus* Linnaeus 1758) in Côte d'Ivoire

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Abstract

The intensification of fish farming activity has led to the proliferation of pathogens, consequently the development of epidemic diseases. Thus, the use of antibiotics has become widespread but this practice has had effects on the environment, health of fish and consumers. Faced with this problem, the technique consisted of strengthening the immunity and health of fish with feed additives containing pigments, known for their antioxidant properties. This study aims to determine the influence of anthocyanin-rich purple Katiola corn on the growth, health, and body composition of tilapia. Five isoprotein feeds (30%) were formulated based on purple corn flour with different quantities of anthocyanins (0, 230, 345, 460, 690mg/100g) and submitted to juveniles (25.39 ±5.95g) in tanks for 49 days. Daily weight gain varied from 0.79 to 0.85 g/day without significant difference. In terms of nutrient retention coefficients, lipid retention increases proportionally with the increase in the quantities of anthocyanins. On the other land, protein retention no longer increases at the threshold of 460mg/100g. This was reflected in the body composition of fish with a significant difference ($p < 0.05$) in lipid content. There is an accumulation of fat in the body of fish when using purple corn. For health of fish, the hepato-somatic index and condition factor were better with diets enriched with more anthocyanins. In conclusion, it can be estimated that the use of anthocyanins from purple corn had an influence on the quality of body composition and the health of the fish.

Keywords

Pigments, Cereals, Hepato-Somatic Index, Isoprotein, Cichlidae

1. Introduction

Global aquaculture production in 2024 has reached a record level of 223.2 million tons, including 185.4 million tons of aquatic animals and 37.8 million tons of algae [1]. This growth demonstrates the potential of aquaculture to meet the growing demand for aquatic feed. However, future expansion of aquaculture must focus on sustainability and benefit the

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regions and populations most in need.

Currently, aquaculture is dominated by ten countries (China, Indonesia, India, Vietnam, Bangladesh, the Philippines, South Korea, Norway, Egypt and Chile) which produce more than 89.8% of the total [2]. Targeted policies, technology transfer, capacity building and responsible investments are essential to promote sustainable aquaculture where the needs are greatest, particularly in Africa.

This significant demand in Africa has resulted in a change in fish farming strategy from the traditional extensive to modern intensive system [3]. This high-intensity fish farming increases a farm's annual yield and a yield per unit of space, but has some drawbacks. Such as physiological stress due to overpopulation and deterioration of rearing water caused by increased fish wastes, constituting a crucial threat to fish farming activity [4]. These various factors lead to a slowdown in fish growth and low resistance to disease. Pathogens can cause epidemics and economic loss. Thus, the use of chemical drugs and antibiotics has been widely used to control epidemic diseases in fish farming, but environmental problems and the increase in resistant pathogens have prompted global authorities to restrict the use of chemical drugs and antibiotics [5]. Faced with this problem, the technique consisted of strengthening the immunity and health of fish with dietary supplementation, in particular feed additives, based on plants [6]. Herbal additives are known for their immunostimulatory, antioxidant, and antimicrobial, growth-promoting properties [7, 8]. Although various plants have been used as feed additives in aquaculture [7, 8]. There are still other agricultural products that deserve to be studied in this area. In this context, a variety of corn called Katiola purple corn, due to the color of its grains, is the subject of our study.

This variety is of great importance to the populations of this central region (Katiola) of Côte d'Ivoire thanks to its medicinal virtues [9, 10]. Indeed, rich in anthocyanins [11] to which it owes its sublime purple color. Purple corn contains many properties and has interesting health benefits. It is a natural source of antioxidants. Research indicates that it promotes good heart health, improves blood circulation, and helps slow aging due to its richness in antioxidants [11].

Recent studies have shown that incorporating natural sources of anthocyanins into fish diets can stimulate growth, the immune system, improve their resistance to diseases and promote more intense pigmentation, an essential criterion for the market value of ornamental and consumable species [12, 13].

This study aims to evaluate the influence of anthocyanins from Katiola purple corn on the growth, health and quality of the flesh of tilapia *Oreochromis niloticus*, thus contributing to a better understanding of the potential benefits of this natural resource in sustainable aquaculture.

2. Material and Methods

2.1. Experimental Design and Fish

This study was carried out in the hatchery of the Oceanological Research Center. The device used were constituted of 15 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.

The fingerlings male of tilapia *O. niloticus* (25.39 ± 5.95 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.

2.2. Feed Ingredients and Formulation

The ingredients used for the preparation of the diets during this experiment were chosen due to their availability and nutrient content (Table 1). Purple corn comes from the National Center for Agronomic Research (CNRA). It contains an amount of 1300 mg/100g of anthocyanins. Other ingredients such as fish meal, soybean meal and white corn flour were purchased from the local market in Abidjan, Côte d'Ivoire.

Table 1. *Biochemical composition of ingredients (% dry weight).*

Ingredients	Proteins	Lipids	Ashs	Fibers	Carbohydrates	Gross energy (kJ/g)
Fish meal	45.23	8.75	20.35	-	8.6	14.94
Soybean meal	45.25	4.3	5.95	5.22	44.5	18.73
Purple corn meal	10.8	3.2	1.08	6.29	76.55	15.39
White corn meal	10.8	3.2	1.08	6.29	76.55	15.39
Cassava starch					100	17.02

An automatic calculation system using the linear programming method [14] on excel was used for diets formulation. Five isoprotein (30%) and isolipid (10%) feed were formulated with different concentrations of anthocyanins from purple corn meal (Table 2).

The diets production was carried out in the Aquaculture Nutrition laboratory of Oceanological Research Center. The feed was made into 3.5 mm diameter pellets using a meat grinder. All diets were sealed in plastic bags and stored at 20 ℃ throughout the experiment.

Table 2. *Quantity of ingredients and biochemical composition of diets.*

Ingredients			**Quantity of anthocyanins (mg/100g)**		
(g/ 100 g)	**0**	**230**	**345**	**460**	**690**
Fish meal	41	39	39	39	39
Soybean meal	20	20	20	20	20
White corn meal	30	20	15	10	0
Purple corn meal	0	10	15	20	30
Cassava starch	3	3	3	3	3
Palm oil	4	4	4	4	4
Vitamin mixture	1	1	1	1	1
Mineral mixture	1	1	1	1	1
Total	100	100	100	100	100
Proximate analysis					
Proteins	30.83	30.83	30.83	30.83	30.83
Lipids	10	10	10	10	10
Ash	10	10	10	10	10
Fiber	2.02	2.02	2.02	2.02	2.02
Nitrogen free extract	38.66	38.66	38.66	38.66	38.66
Digestible energy (Kj/g)	13.93	13.93	13.93	13.77	13.77
Gross energy (Kj/g)	17.49	17.49	17.49	17.49	17.49
P/E (mg protéines/Kj)	22.13	22.13	22.13	22.13	22.13

P/E: Proteins energy ratio.
0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

2.3. Experimental Protocol

After a acclimation period, 225 juvenile *O. niloticus* (25.39 ± 5.95 g) were distributed among the 15 glass tanks, 10 per tank. Then, the fish were fed with the experimental diets twice a day at a ration of 4% of the biomass (8 a.m. and 4 p.m.) for 49 days. The first distribution of feed is preceded by the measurement of temperature (27.7 to 28.80 ℃), pH (7.35 to 7.8) and dissolved oxygen (5.88 to 6.86 mg/L). Once a week, the fish were randomly sampled in each glass tank to measure weight and length. At the beginning the experiment, an initial sample of ten fishes were taken and frozen (-20 ℃) for subsequent whole body proximate analysis. At the end of the experiment, the weight, length, gonad and liver of each fish was recorded. Ten fish from each diet were stored at – 20 ℃ for whole-body composition analysis.

2.4. Biochemical Analysis

The analyses were carried out on the ingredients, experimental diets and body of fish. The dry matter was determined by oven drying at 105 ℃ for 24 hours. Crude proteins (% N x 6.25) were determined by the Kjeldahl method, lipids by ether extraction method (Soxtherm, Gerhardt, Germany), fibers by acid and alkaline hydrolysis. Ash content was determined by combustion in a muffle furnace at 550 ℃ [15]. The extraction of phenolic compounds was carried out according to the method of [16] and the determination of anthocyanin content was carried out according to the method described by [17].

2.5. Biological Evaluation

Survival Rate (SR), Daily Weight Gain (DWG), Specific Growth Rate (SGR), Feed Conversion Ratio (FCR), Protein Efficiency Ratio (PER), Nutrient Retention Ratio (NR), Hepato-Somatic Index (HIS), Gonado-Somatic Index (GSI) and Condition factor (K) were calculated as follows:

- SR (%) = Nf / Ni; Ni: initial number, Nf: final number of fish;
- DWG (g/d) = (Pf – Pi) / t,
- SGR (%/d) = 100 (LnPf – LnPi) /t; Pi: initial weight, Pf: final weight of fish, t: duration of the experiment:
- FCR = Dry feed consumed (g) / weight gain (g);
- PER = weight gain (g) / Protein consumed;
- NR (%) = [(Pf x Ncf) - (Pi x Nci) / Nti] x 100 With, Pf: final weight, Ncf: final body nutrient, Pi: initial weight, Nci: initial body nutrient, Nti: total nutrient consumed.
- Hepato-somatic index by the [18] method, HSI (%) = (Liver weight / fish weight) x 100;
- Gonado-somatic index according to [19], GSI = (Gonad weight / fish weight) x 100;
- Condition factor [20], K = (Total fish mass / Length^3) x 100.

2.6. Statistical Analysis

Results were analyzed with analysis of variance using Statistical Version 7.1 software package. Tukey HSD post hoc tests were used to compare difference between treatments means when significant F-values were observed. All percentage and ratio data were arc-sin transformed before analysis. The treatment effects were considered to be significant at $p < 0.05$.

3. Results

3.1. Zootechnical Parameters

Figure 1 illustrates evolution of the weight and length of juvenile *O. niloticus* during the experiment. For all diets, a progressive increase of the weight and length is observed over time. Evolution of the weight is almost similar for all diets, but it is different for length at the end of experiment.

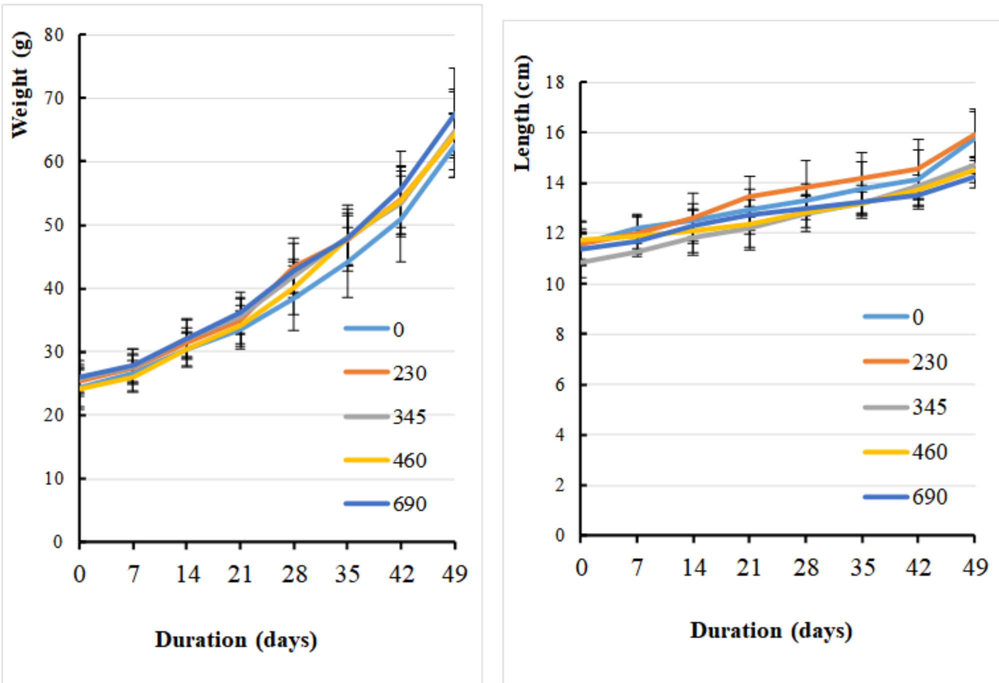

Figure 1. *Evolution of the weight and length of juvenile O. niloticus during the experiment.*

0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

Table 3 shows the survival rates and zootechnical parameters of the fish according to the diets. No significant difference is observed for the determined parameters except for the length gain ($p< 0.05$).

Table 3. *Survival rates and zootechnical parameters of fish.*

	Quantity of anthocyanins (mg/100g)				
Parameters	**0**	**230**	**345**	**460**	**690**
W_i (g)	24.34±2.96	25.31±2.20	25.8±2.11	24.12±3.13	25.97±2.58
W_f (g)	62.55 ±5.03^a	64.23 ±3.14^a	64.82 ±6.13^a	64.45 ±6.91^a	67.66 ±7.14^a
L_i (cm)	11.6 ±0.86	11.57 ±0.61	10.85 ±0.60	11.71 ±0.77	11.39 ±0.66
L_f (cm)	15.78 ±1.04^b	15.9 ±1.01^b	14.69 ±0.34^a	14.5 ±0.51^a	14.22 ±0.43^a
SR (%)	86.66 ±23.33^a	90 ±7.07^a	83.33 ±14.14^a	93.33 ±7.07^a	90 ±0^a
WG (g)	38.21 ±2.84^a	38.92 ±2.53^a	39.02 ±3.57^a	40.33 ±3.38^a	41.69 ±4.01^a
LG (cm)	4.18 ±1.21^b	4.33 ±1.01^b	3.84 ±0.1^{ab}	2.79 ±0.78^b	2.83 ±0.62^b
DWG (g/d)	0.79 ±0.18^a	0.79 ±0.10^a	0.80 ±0.11^a	0.82 ±0.20^a	0.85 ±0.15^a
SGR (%/d)	1.93 ±0.17^a	1.90 ±0.13^a	1.88 ±0.17^a	2.01 ±0.21^a	1.95 ±0.15^a

W_i: initial weight; W_f: final weight; Li: initial length; Lf: final lenhgt; SR: survival; WG: gain weight gain; LG; length gain; DWG: Daily weight gain; SGR: specific growth rate. 0: Food containing 0 mg/100g of anthocyanin; 230: Food containing 230 mg/100g of anthocyanin; 345: Food containing 345 mg/100g of anthocyanin; 460: Food containing 460 mg/100g of anthocyanin; 690: Food containing 690 mg/100g of anthocyanin. Means in a row with different superscripts significantly ($p < 0.05$).

0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

Weight gains (Figure 2) increase with the progressive incorporation of anthocyanins even if no significant difference is observed between the different diets.

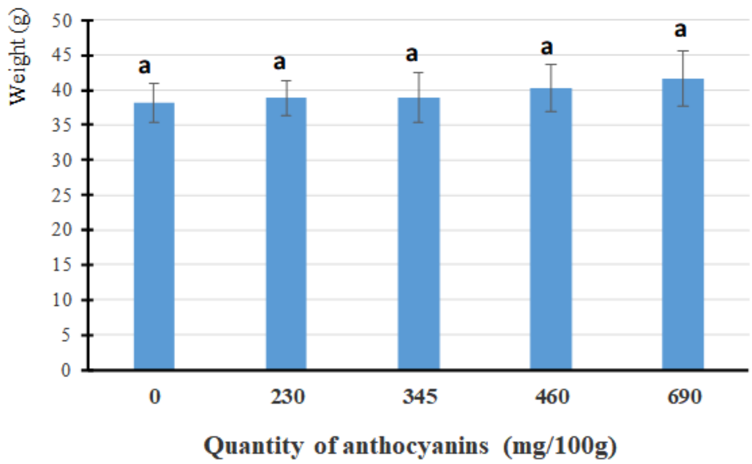

Figure 2. *Weight gain as a function of anthocyanin quantity in diets.*

0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

3.2. Feed and Nutrient Utilization

Feed Conversion Ratio (Figure 3) ranges from 2.63 to 2.93, with the lowest value observed in the diet containing 690 mg/100g of anthocyanins. Concerning, Protein Efficiency Coefficient, the values are between 1.10 and 1.22. No significant difference is observed for the two parameters ($p > 0.05$).

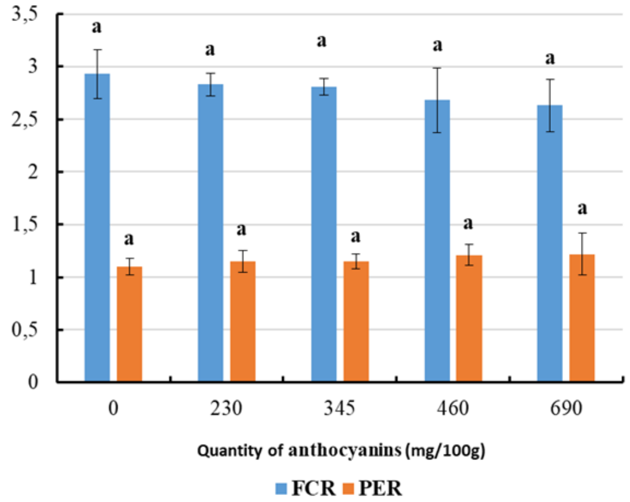

Figure 3. Feed Conversion Ratio and Protein Efficiency Ratio as a function of the quantities of anthocyanins in the diets.

0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

Evolution of nutrient retention in the fish organism is observed in Figure 4. Protein retention curve remains stable after 460 mg/100g of anthocyanins, whereas the lipid retention curve shows an increasing trend after 690 mg/100g.

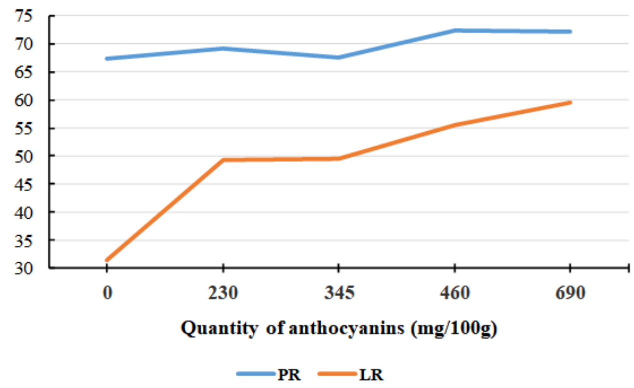

Figure 4. The evolution of nutrient retention in the fish organism.

0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

3.3. Whole Body Proximate Composition

Whole body proximate composition at the end of this experiment is shown in Table 4. The parameters determined do not present any significant difference as a function of diets consumed, except the lipid content. Lipid content ranges from 9.22 to 13.22% of dry matter, with the highest content for the diet containing 690mg/100g of anthocyanins ($p < 0.05$).

Table 4. Whole body proximate composition of juvenile O. niloticus.

	Quantity of anthocyanins (mg/100g)				
Parameters	**0**	**230**	**345**	**460**	**690**
Proteins (% DM)	60.45 ±0.7^a	60.12 ±0.11^a	61.02 ±0.69^a	59.84 ±0.28^a	59.23 ±0.71^a
Lipids (% DM)	9.22 ±0.12^a	12.08 ±0.32^b	12.45 ±0,24^b	12.75 ±0.41^b	13.22 ±0.74^b
Ash (% DM)	18.69 ±1.21^a	18.87 ±2.01^a	17.13 ±2.75^a	18.98 ±3.01^a	18.15 ±2.45^a
Energy (Kj/g)	17.66 ±0.56^a	18.37 ±0.62^a	18.77 ±1.03^a	18.45 ±1^a	18.72 ±1.01^a

DM: Dry Matter. Means in a row with different superscripts significantly ($p < 0.05$).
0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

3.4. Health Parameters

Data presented in Table 5 concern fish health parameters. Hepato-somatic index and condition factor constant were high, respectively, with anthocyanin quantities of 460 mg/100g (2.94) and 690 mg/100g (2.35). There was no significant difference between the diets in the gonado-somatic index ($p > 0.05$).

Table 5. Mean values of hepato-somatic index, gonad-somatic index and condition factor of juvenile O. niloticus after 49 days of rearing.

	Quantity of anthocyanins (mg/100g)				
Parameters	**0**	**230**	**345**	**460**	**690**
HSI	1,64 ±0,45[a]	1,86 ±0,31[a]	2,04 ±0,15a[b]	2,94 ±0,31[b]	2,34 ±0,22[b]
GSI	1,08 ±0,11[a]	0,93 ±0,31[a]	1,27 ±0,54[a]	1,22 ±0,42[a]	1,12 ±0,38[a]
K	1,59 ±0,10[a]	1,57 ±0,05[a]	1,98 ±0,21[b]	2,11 ±0,18[b]	2,35 ±0,25[b]

HIS: hepato-somatic index; GSI: gonado-somatic index; K: Condition factor. Means in a row with different superscripts significantly ($p< 0.05$). 0: diet containing 0 mg/100g of anthocyanin; 230: Diet containing 230 mg/100g of anthocyanin; 345: Diet containing 345 mg/100g of anthocyanin; 460: Diet containing 460 mg/100g of anthocyanin; 690: Diet containing 690 mg/100g of anthocyanin.

4. Discussion

The quality of the rearing water represented by the different parameters such as temperature (27.7 to 28.8 ℃), pH (7.35 to 7.8) and dissolved oxygen (5.89 to 6.86 mg/L) are within the range of values recommended for tilapia rearing [21, 22]. Concerning ammonium (0.38 to 0.55 mg/L) and phosphate (0.11 to 0.38 mg/L), the values are below the critical values for tilapia [23] which is 1 mg/L for ammonium. These values show the good quality of the breeding environment throughout our experience. Therefore, the breeding environment cannot be a limiting factor in the analysis of the parameters determined in the rest of the study.

Fish survival rates ranged from 83.33 to 93.33% and showed no significant difference between the different diets. These values are comparable to those of [24] which are between 89.5 and 93.5% in ponds and of [25] in tanks with rates of 90 to 100%. These high values may be linked to the good quality of the breeding water recorded throughout the experiment. They also reflect the good quality of the experimental feed given to the fish.

Evolution of weight of the fish fed with the different diets show a similar trend during the first week. This phase could correspond to a period of habituation essential for the fish to adapt to the diets imposed on them. Beyond this period, the evolution is different with a more significant progression in the fish receiving diets rich in anthocyanins. In terms of length, evolution is significantly different. Concerning weight gain, it increases with the quantity of anthocyanins provided even if no significant difference is observed, on the other hand a significant difference appeared with length gain. This reflects a low rate of relationship between weight and length of fish [26]. In this study, weight gain ranged from 38.21 g to 41.69 g depending on the quantities of anthocyanins. An improvement was observed with the intake of anthocyanins. This progression could be attributed to the beneficial effect of anthocyanins, which would improve digestion, intestinal flora and the bioavailability of nutrients [27]. Moreover, phenolic compounds are known for their antioxidant activity, helping to limit oxidative stress and stimulate growth [28]. The diet containing 690 mg/100g presents the highest value of DWG (0.85 g/d), which indicates optimal daily growth at this level of incorporation. Diets with 460 and 690 mg/100g of anthocyanins maintained a good level of DWG, but not significantly different from the diet without anthocyanins. This slight weight gain may be related to the quantities of anthocyanins incorporated into the experimental diets. It can be assumed that a greater quantity of anthocyanins significantly improved growth. Although SGR did not differ significantly, the trend towards increased SGR with the addition of anthocyanins for diets with 460 and 690 mg/100g was observed.

Feed conversion ratio and protein efficiency ratio are two key parameters for assessing the feed performance of diets. In this study, the FCR values ranged from 2.63 to 2.93. The diet with 690 mg/100g of anthocyanins shows the lowest value indicating optimal feed efficiency, while the diet without anthocyanins shows the highest FCR, reflecting poorer feed conversion. This improvement with enriched diets, particularly at 690 mg/100g, could be related to better palatability, digestibility and assimilation of nutrients promoted by anthocyanins in purple corn. These bioactive compounds are known to promote intestinal health, reduce oxidative stress and improve zootechnical performance [28, 29]. Protein efficiency ratio represents the quantities of weight gained per gram of protein consumed. the PER varies from 1.1 to 1.22 with the best value also obtained for the diet containing anthocyanins. This indicates that the protein is used more efficiently at this level of anthocyanin incorporation. These results confirm the observations of [30], who showed the importance of a balance between protein quality and metabolic efficiency in tilapia. The presence of natural antioxidants such as anthocyanins may have contributed to reducing nitrogen losses, increasing the bioavailability of amino acids and stimulating digestive enzymes [31, 32].

Figure 4 shows the evolution of nutrient retention in relation to the quantity of anthocyanins. Protein retention remains constant at 460 mg/100 g of anthocyanins, which may explain the protein content in the body composition. This content does not vary significantly between diets. On the other hand, lipid retention evolves progressively with the quantity of anthocyanins. This correlation can explain the significant difference in lipid contents in the carcass. This content increases with the progressive incorporation of purple corn meal, which suggests that anthocyanin causes the accumulation of fat in the fish carcass. This relationship can be explained by the close link between pigment metabolism and lipid metabolism [33].

In this study, condition factor ranged from 1.57 to 2.25, the results show that K was greater than 1 ($p > 0.05$). This result means that the fish subjected to the different experimental diets were overweight during the experiment [34]. The highest factors are observed in fish fed diets containing 345 to 690 mg/100g of anthocyanins, this suggests that fish in this group have a good general body shape. These results corroborate those of [35], who showed that well-balanced diets lead to an improvement in the condition index in tilapia. The richness in anthocyanins would thus have made to improve the general body condition of the fish by promoting muscle storage and digestive health. No significant differences were observed in gonado somatic index, suggesting that the anthocyanins do not compromise reproductive function. Hepato-somatic index is high with diets containing 345 to 690 mg/100g of anthocyanins, reflecting a moderate stimulation of hepatic activity, probably linked to the transformation of bioactive compounds and increased energy mobilization due to faster growth [27].

5. Conclusion

Objective of this study was to verify the influence of anthocyanins from purple katiola corn in fish feed. The results show that there is no significant difference in fish growth. There is a progressive lipid retention with the increase in anthocyanin leading to an accumulation of fat in the body of the fish. In terms of health parameters, the highest values are observed in fish fed diets enriched with anthocyanins. This result suggests a general overweight of the fish and a moderate stimulation of hepatic activity. This result can help fight epidemics in fish farms by improving the immune system of fish. It would be important to carry out an experiment over a long period in order to verify the influence of lipid retention on growth and determine the influence of those anthocyanins on the pigmentation of the flesh.

Abbreviations

SR	Survval Rate
DWG	Daily Weight Gain
SGR	Specific Growth Rate
FCR	Feed Conversion Ratio
PER	Protein Efficiency Ratio
HIS	Hepato-Somatic Index
GSI	Gonado-Somatic Index

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Author Contributions

Yéo Gopéyué Maurice: Conceptualization, Data curation, Funding acquisition, Formal Analysis, Methodology, Software, Writing – original draft.

Djeke Paul Simplice: Conceptualization, Data curation, Methodology, Software, Writing – review & editing.

N'Cho Achi Laurent: Formal Analysis, Methodology, Writing – review & editing.

Etchian Assoi Olivier: Software, Supervision, Validation, Visualization.

Blé Mélécony Célestin: Supervision, Validation, Visualization.

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Conflicts of Interest

The authors declare no conflicts of interest.

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