

Growth Performance, Feed Utilization and Sensory Characteristics of Nile Tilapia, *Oreochromis niloticus* Fed Diets with High Inclusion Levels of Copra Meal

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Abstract

Background: The low cost and wide-availability of copra meal in many tropical countries where aquaculture is practiced have generated much interest in its potential inclusion in fish diet formulations. The present study was designed to investigate the effect of very high inclusions of autoclaved copra meal on the growth and feed utilization parameters as well as the sensory qualities of the Nile tilapia, *Oreochromis niloticus*.

Methods: Fish were fed three experimental diets, a control (CTRL) diet which had fishmeal as the main protein source at an inclusion of 365 g kg⁻¹ and two test diets which contained copra meal at 680 g kg⁻¹ inclusions for a 6-week period. One of the copra meal diets contained sesame meal supplementation (CM+S) as a natural high source of methionine which is the first limiting essential amino acid in copra meal.

Results: The dietary treatments had no differential effects on growth, feed intake, or feed utilization parameters in *O. niloticus*. With the exception of the significantly higher lipid content of the CM+S group, the whole body compositions of the different fish groups were not significantly affected by the different dietary treatments. Remarkably, the high dietary inclusions of copra meal did not have any significant effects on fillet sensory attributes.

Conclusions: The results of this study demonstrate that it is possible to include autoclaved copra meal up to 680 g kg⁻¹ in Nile tilapia diets without any deleterious effects on fish growth or on flesh sensory characteristics.

Keywords: Copra meal; Feed utilisation; Growth; High inclusion; Nile tilapia; Organoleptic properties

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Introduction

The Nile tilapia (*Oreochromis niloticus*) is one of the most favoured fish species in aquaculture, due to its high tolerance of a relatively wide range of environmental conditions, which is presumably also a key factor in the marked increase in global production [1]. Global tilapia production has increased considerably during the last few decades with total outputs increasing more than four-fold, from 830,000 T in 1990 to 5.3 million T in 2014 [2]. The low trophic level and the omnivorous food habits of this species make them a relatively inexpensive fish to feed, unlike other strictly

carnivorous finfish, such as salmon, which rely on high protein and lipid diets which are largely based on fish sources [1]. Tilapia is able to grow well on less refined protein sources, thus making them attractive as sources of animal protein for humans [3]. Over the years, numerous studies have evaluated the nutritional value of low-cost plant ingredients in tilapia diets, using digestibility, growth performance and feed intake as measurement end-points. These plant resources include palm kernel meal [4,5], groundnut (peanut) meal [6], Cottonseed meal [7], sesame seed meal [8] and copra meal [9].

Copra meal, often classified as a medium-grade feed ingredient in terms of its nutritional composition is particularly common in ruminant nutrition but less common in monogastrics like fish [10]. However, its low cost and wide-availability in many tropical countries where aquaculture is practiced have generated much interest in its potential inclusion in fish diet formulations. The high fiber content and anti-nutritional factors (ANFs) limit its inclusion in aqua feeds to very low levels [11] recommend maximum dietary inclusion levels of 150 g kg⁻¹ for omnivorous fish and 100 g kg⁻¹ for carnivorous fish species. Some earlier studies have, however, shown that unprocessed copra meal can replace 300 g kg⁻¹ of fishmeal in Nile tilapia diets without adversely affecting feed intake [12], protein digestibility and short-term growth and feed utilization parameters [13]. Interestingly, simple processing techniques, such as soaking in water [14], and wet heating/autoclaving [15] can significantly reduce the levels of anti-nutritional factors in copra meal and facilitate higher levels of inclusions in fish feeds. Supplementing for the deficient amino acids in copra meal, particularly lysine and methionine can also improve fish growth and feed utilization [11].

Therefore, as part of the larger investigation to reduce dependency of fishmeal in the steadily increasing aquaculture sector and to provide affordable feeds for farmers, the present study investigated the effects of high dietary inclusion level (680 g kg⁻¹) of autoclaved expeller-pressed copra meal on the growth performance, feed utilisation and sensory characteristics of Nile tilapia, *O. niloticus*. It also assessed the effects of supplementing the high copra-based diet with sesame seed meal which is rich in methionine (the richest among oilcakes and meals), arginine and leucine on general growth and feed utilization parameters of the Nile tilapia.

Materials and Methods

Experimental diets

For this trial, three diets were formulated to satisfy the nutritional requirements of adult Nile tilapia [3]. The formulations of the experimental diets as well as their respective proximate composition are presented in **Table 1**. The control diet (CTRL) had fishmeal as the main protein source while the test diets contained copra meal at 680 g kg⁻¹ inclusions with (CM+S) and without (CM) sesame seed meal supplementation. The copra meal was autoclaved at 120°C for 30 min before dietary inclusion which significantly reduced tannin levels from 2.5% to 1.4%. All the basal ingredients used in the feed formulation were finely-ground and sieved to obtain a homogenous mixture and weighed out according to their respective formulations. Mineral and vitamin premixes were added to each diet. Pre-gelatinised tapioca flour was added as the binder and the diets pelleted using a meat grinder fitted with a 2 mm die plate. The resulting pellets were oven-dried (Hereaus Instruments K1252, Hanau, Germany) at 40°C for 48 h.

Experimental fish and rearing conditions

The Nile tilapia (mixed sex) used for this study were obtained from a commercial farm (Ekofisk, Sjöbo, Sweden) and transported to

experimental facilities of the Technical University of Denmark at the North Sea Research Centre in Hirtshals for the trials. Following a 2-week quarantine and acclimatization period, fish were transferred to a recirculation system consisting of twelve 120 L, cylindro-conical PETG thermoplastic tanks at a stocking density of 15 individuals (66.68 ± 0.10 g mean weight) per tank, for a 6-week growth trial. Water supply to each tank was maintained at a flow rate of 100 L h⁻¹. Water temperature in the experimental system was maintained at 25°C throughout the experiment by means of a relay-controlled 1.5 kW electric porcelain heater (Jevi, Vejle, Denmark) submerged in the reservoir tank. Dissolved oxygen concentration (>7 mg L⁻¹), pH and levels of nitrogenous excretions in the tanks were monitored daily. Fish were kept under a 12 h light: 12 h dark photoperiod during the trial period.

Each diet was randomly assigned to quadruplicate groups of tanks and fish were hand-fed at the same fixed rate (3% of total bulk weight), divided into two equal portions and administered twice daily at 9:00 and 16:00 h. And all uneaten feed during the trial period was accounted for. All fish were individually weighed at the beginning of the trial to ensure uniformity in the initial weights. For the intermediate (after 3 weeks) and final weight measurements (after 6 weeks), fish were bulked together, weighed and counted. An additional 30 fish from the initial stock were randomly sampled and euthanized using ethylene glycol monophenyl ether (Merck KGaA, Darmstadt, Germany). The length and weight measurements of fish selected at the end of the trial were determined using a measuring board and a scale (Ohaus Scout SPX, New Jersey, USA) respectively. After determination of condition factor, hepatosomatic and visceromatic indices, fish were stored at -20°C for subsequent whole-body proximate composition. At the end of the trial, 7 fish from each of the tanks were randomly sampled and similarly processed.

Proximate analysis of the feed and fish samples

Dry matter, crude protein, crude lipid and ash of the diets and carcasses were determined following the procedures of the Association of Official Analytical Chemists [16]. The seven (7) randomly selected fish samples from the different experimental tanks were each homogenized prior to the proximate analysis. Dry matter was determined after oven drying for 24 h at 105°C (Mettler UN110, Büchenbach, Germany). Ash contents were calculated from the weight loss after incineration of the samples for 6 h at 550°C in a muffle furnace (Hereaus Instruments K1252, Hanau, Germany). Crude protein levels were determined from the Kjeldahl method (Foss Kjeltac 2200, Hillerød, Denmark) and crude lipid by the method of [17].

Organoleptic tests

At the trial termination, five individuals per treatment were randomly caught and fillets cut from both sides of each fish. The fillets were vacuum-packed and kept frozen prior to the organoleptic tests. Prior to the sensory session, the fillets (with skin) were thawed in a refrigerator overnight and cooked according to the methods described by [18]. Fillets were presented in 10 g samples in coded bowls to 8 people randomly selected from the staff and students of the Technical University

of Denmark at the North Sea Research Centre in Hirtshals. The tests were conducted under normal white lighting in order not to affect the flesh colour assessment. A category scale of 1-5 generally ranging from slight to moderate to extreme was employed for each descriptor (**Table 2**).

Economic analysis

A simple economic analysis using incidence cost and profit index calculations was conducted at the end of the study to assess the cost effectiveness of diets used in the feed trial. The value of fish produced during the trial was calculated using the net bulk weight gain (g) over the trial period and not final bulk weight (g). Feed costs (per kg) for the CTRL, CM and CM+S diets taking into consideration the local market prices of the basal feed ingredients as of February 2014, were US\$ 0.71, US\$ 0.52 and US\$ 0.44, respectively. The prevailing local Ghanaian market value of 1 kg of *O. niloticus* during the study period was US\$ 3.9 (February, 2014). The incidence cost and profit index were then calculated as follows;

Incidence Cost=Cost of feeding fish/Weight of fish produced

Profit Index=Value of fish/Cost of feeding

Statistical Analysis

Table 1: Formulation (g kg⁻¹ as fed) and proximate composition (g kg⁻¹ dwb) of the experimental diets.

	Control	CM	CM+S
Ingredients			
Fishmeal ^a	365	217	185
Copra meal ^b	-	680	680
Sesame meal ^c	-	-	80
Wheat bran ^d	140	-	-
Rice bran ^e	410	10	-
Palm oil	40	48	10
Vitamin and mineral premix ^f	5	5	5
Diphosphate	20	20	20
Cassava starch (binder)	20	20	20
Proximate composition			
Dry matter	968	941	952
Crude protein	323	311	318
Crude lipid	148	151	160
Ash	125	109	104

^aDanish fishmeal: 900 g kg⁻¹ dry matter, 720 g kg⁻¹ crude protein, 120 g kg⁻¹ crude lipid, FF Skagen A/S, Skagen, Denmark
^bExpeller-pressed (autoclaved at 120°C for 30 min): 930 g kg⁻¹ dry matter, 200 g kg⁻¹ crude protein, 80 g kg⁻¹ crude lipid, Imported from Kumasi, Ghana
^cMilled Danish sesame seeds: 940 g kg⁻¹ dry matter, 270 g kg⁻¹ crude protein, 500 g kg⁻¹ crude lipid
^dDanish wheat bran: 940 g kg⁻¹ dry matter, 160 g kg⁻¹ crude protein, 10 g kg⁻¹ crude lipid, Valsemøllen DK, Denmark
^eMilled rice bran: 900 g kg⁻¹ dry matter, 90 g kg⁻¹ crude protein, 74 g kg⁻¹ crude lipid, Imported from Ghana
^fThe following was supplied (mg kg⁻¹ except as noted): vitamin A 3750 IU; cholecalciferol 750 IU; α-tocopherol, 131.3; thiamine, 7.5; riboflavin, 15; pyridoxine, 7.5; vitamin B12, 0.002; vitamin K3, 7.5; zinc, 75; iodine, 0.9; copper, 3.75; manganese, 22.5; cobalt, 0.75; selenium, 0.19.

Table 2: The sensory descriptors used for the organoleptic testing [18].

Descriptors	Definitions	Range (1-5)
Colour	The colour strength of cooked muscle	White-reddish
Sweet taste	The taste on the tongue associated with sugar	Light-strong
Salty taste	The taste on the tongue associated with sodium ions	Light-strong
Total flavour	The total oral impression as perceived by taste, smell, feeling in the mouth.	Light-strong
Vegetal flavour	General flavour impression (smell and taste) associated with plants	Absent-intense
Cooked meat flavour	General flavour impression (smell and taste) associated with cooked meat (e.g., chicken)	Absent-intense
Fatty flavour	Impression of fatness perceived as aromatic notes and mouth coating film	Light-strong
Firmness	Degree of firmness/connection of the fibres during chewing	Tender-firm

All data are presented as means ± standard deviations (SD). Statistical analysis was performed using one-way analysis of variance (ANOVA) and differences between the treatment means compared by the Turkey multiple comparison test. Differences were considered significant at p<0.05. All graphs and statistical analyses were executed using SigmaPlot ver. 12.0 (Systat Software, Inc).

Results

Growth performance and biometric parameters

No mortalities were recorded during the trial period. The high dietary inclusions of copra meal did not impair general growth performance as well as feed utilization parameters over the experimental period (**Table 3**). There were no significant differences between any of the calculated growth performance parameters for the treatment groups. Specific growth rates varied between 1.31 and 1.34 % d⁻¹. Similarly, feed conversion ratio (FCR) as well as thermal growth coefficients (TGC) varied over a narrow range among the 3 dietary treatments. The sesame-supplemented copra (CM+S) diet resulted in slight improvements in general growth performance and feed efficiency but these were not significantly different from the control or copra meal diets. Hepatosomatic indices (HSI) and condition factors (CF) were not significantly affected by the diets. The fish groups fed the diets containing copra meal (CM and CM+S); however, had significantly higher VSIs due to depositions of intraperitoneal fat.

Body composition and nutrient utilization

The whole body nutrient compositions of Nile tilapia groups fed different diets for the trial duration are presented in **Table 4**. With the exception of lipid, the whole body compositions of the different fish groups, expressed on a wet weight basis, were not significantly affected by the different dietary treatments. Whole body lipid content was significantly higher in the fish

Table 3: Growth, feed utilization and biometric parameters of the Nile tilapia groups after 6 weeks.

	Control	CM	CM + S
Mean initial bulk weight (kg)	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00
Mean final bulk weight (kg)	1.75 ± 0.06	1.73 ± 0.06	1.76 ± 0.04
Weight gain (%)	74.87 ± 6.50	73.35 ± 6.22	75.62 ± 4.40
Feed intake (gfish ⁻¹ 42 days ⁻¹)	62.79 ± 0.28	62.39 ± 0.58	62.84 ± 0.11
SGR ^a (%day ⁻¹)	1.32 ± 0.09	1.31 ± 0.08	1.34 ± 0.06
TGC ^b	1.95 ± 0.14	1.92 ± 0.14	1.97 ± 0.10
FCR ^c	1.27 ± 0.11	1.28 ± 0.12	1.24 ± 0.08
HSI ^d (%)	1.77 ± 0.60	1.75 ± 0.32	1.81 ± 0.32
VSI ^e (%)	8.65 ± 1.37a	9.31 ± 1.01b	9.33 ± 0.82b
CF ^f	3.41 ± 0.27	3.56 ± 0.22	3.62 ± 0.24

Each value is the mean±SD of data from quadruplicate groups.
 Weight Gain=100 x (final body weight – initial body weight)/ initial body weight
 Feed Intake per fish=(Feed offered (g)/average total weight (g))
^aSpecific growth rate=100 x ln(final body weight-initial body weight)/ Experimental days
^bThermal growth coefficient = $\left[\frac{(\sqrt[3]{W_t} - \sqrt[3]{W_0})}{\text{Experimental days} \times \text{Experimental Temperature}} \right] \times 1000$
^cFeed conversion ratio=Dry feed consumed/wet weight gain
^dHepatosomatic index=(Liver weight/whole body weight) x 100
^eVisceromatic index=(Viscera weight/whole body weight) x 100
^fCondition factor=(Whole body weight/standard length³) x 100

Table 4: Whole body proximate compositions and nutrient utilisation of the Nile tilapia groups after 6 weeks.

Body Proximate Composition (g kg ⁻¹ wet weight)	Control	CM	CM+S
Moisture	72.00 ± 6.00	73.17 ± 0.76	71.55 ± 1.35
Crude protein	153.90 ± 2.60	153.80 ± 0.21	154.10 ± 0.14
Crude lipid	78.70 ± 4.10 ^a	80.70 ± 8.50 ^a	90.90 ± 13.10 ^b
Ash	48.20 ± 0.80	45.1 ± 2.10	44.70 ± 2.60

Nutrient utilisation

Protein efficiency ratio	2.46 ± 0.20	2.52 ± 0.23	2.45 ± 0.18
Protein retention efficiency (%)	41.45 ± 2.90	42.66 ± 4.49	42.66 ± 3.69
Lipid retention efficiency (%)	49.23 ± 7.11 ^a	48.94 ± 6.41 ^a	55.97 ± 11.87 ^b

Each value is the mean ± SD of data from quadruplicate groups Initial carcass proximate composition (g kg⁻¹ wet weight basis): (Moisture content: 755.70; Crude protein: 145.90; Crude lipid: 49.20; Ash: 4.82)
 Protein efficiency ratio=Fish wet weight gain/protein intake Protein retention efficiency=(Nitrogen gain/nitrogen intake) x 100
 Lipid retention=(Lipid gain/lipid intake) x 100

fed the CM+S diet than in the other dietary groups. In terms of nutrient utilization, protein efficiency ratio (PER) as well as protein retentions were not significantly affected by the dietary treatments. Lipid retention efficiencies expressed in terms of ingested dietary lipid over the trial period were significantly different among the 3 dietary treatments with the CM+S group recording a significantly higher retention compared to the other two dietary groups.

Sensory characteristics

There were no significant differences in the scores for the sensory descriptors among the 3 dietary treatments indicating little or no effects of the high dietary inclusion of copra meal on the flesh colour, flavour and firmness. The effects of the dietary treatments on sensory characteristics are shown in **Table 5**.

Simple economic analyses were conducted to assess the cost effectiveness of the diets used in the growth trial. The 680 g kg⁻¹ dietary inclusion of the copra meal resulted in significantly lower feed cost compared to the control diet; the CM and CM+S diets reduced feed cost by 27 and 38%, respectively relative to the cost of the control diet. The monetary values of the fish produced at the end of the trial were similar among all the dietary treatments and did not vary significantly. At the end of the trial, the incidence cost (Cost of feed (In monetary terms)/weight of fish produced (kg) was significantly higher for the control diet compared to the diets containing the copra meal, indicating that it is comparatively less profitable using the fishmeal-based control diet. Calculations of profit indices showed that the CM+S diet offers the best economic viability taking into consideration both the monetary values of fish produced and feed given over the trial period.

Discussion

Growth performance

In this study, it was possible to replace fishmeal with copra meal in Nile tilapia diets to a very high extent (680 g kg⁻¹ inclusion level, corresponding to more than 40% of the absolute protein content), without observing negative effects on fish growth or feed utilization efficiencies. The study also demonstrated that autoclaving copra meal can facilitate their inclusions at very high levels (~700 g kg⁻¹) in tilapia diets without significantly affecting growth and feed utilization parameters. Earlier studies on the inclusions of untreated copra meal in fish diets even at the comparatively lower levels have generally reported significantly depressed growth as well as feed utilization parameters [9,14,19,20]. The high tannin content of unprocessed copra meal is known to negatively affect feed palatability and intake as well as nutrient absorption and consequently depress fish growth [14,15]. Condensed tannins present in copra meal at a level of approximately 24 g kg⁻¹ caused growth depression in Rohu fingerlings even at lower inclusion levels of 200 g kg⁻¹ [14]. In the

Table 5: Sensory characteristics of the tilapia fed the different experimental diets (Mean±SD) Economic Analysis.

Descriptors	Dietary treatments		
	Control	CM	CM+S
Colour	1.38 ± 0.52	1.50 ± 0.53	1.38 ± 0.52
Sweet taste	1.50 ± 0.71	1.63 ± 0.92	1.88 ± 0.83
Salty taste	1.38 ± 0.74	1.25 ± 0.71	1.63 ± 0.74
Total flavour	2.38 ± 0.92	2.75 ± 0.89	3.00 ± 0.93
Vegetal flavour	1.75 ± 0.89	1.88 ± 0.99	2.13 ± 1.36
Cooked meat flavour	2.63 ± 0.92	3.00 ± 1.20	3.25 ± 0.89
Fatty flavour	2.50 ± 0.53	2.50 ± 0.53	2.38 ± 0.52
Firmness	2.63 ± 1.06	2.50 ± 0.76	2.75 ± 1.03

Scores are on a scale of 1 to 5 (increasing intensity).
n=8 for each descriptor

present study, feed intake was not impaired by the additions of copra meal indicating the possible elimination of the negative effects of tannin on feed palatability by the autoclaving process after tannin level was reduced by 44%.

Although not significantly different from the other dietary treatments, the better growth performance and feed efficiency of the CM+S dietary groups is likely to be as a result of the inclusion of sesame in the diet. Sesame seed meal unlike most oilseed meals is rich in methionine (the richest among oilcakes and meals), arginine and leucine [3,21]. It is thus likely that the sesame-supplementation augmented the amino acid profile, particularly the methionine content of the diet leading to the better growth indices and feed efficiencies. An inclusion level of more than 80 g kg⁻¹ might, however, be required for more significant effects of sesame-supplemented Nile tilapia diets on growth and feed utilisation.

Nutrient utilization and whole body proximate composition

Aside the significantly higher whole body lipid content of the CM+S tilapia group, this study recorded no significant differences in the whole body proximate compositions of the different treatment groups. The significantly higher whole body lipid content of the CM+S fish group is possibly reflective of excessively high depositions of dietary lipid as visceral fat rather than in the flesh (**Figure 1**). This is more evident in the inability of the sensory panel to differentiate between the organoleptic parameters of the flesh of the tilapia fed the different diets.

Sensory evaluation

The sensory evaluation of the cooked tilapia fillets indicated that the high dietary inclusion of copra meal did not have significant impacts on the descriptors tested although dietary lipid sources and levels are often associated with changes in fillet characteristics. Some trends were, however, apparent despite the similarities in the sensory descriptors indicated by the evaluators. Taste (sweet and salty tastes) as well as flavour (total, vegetal and cooked meat flavours) were perceived to rank highest in the flesh of the tilapia fed the CM+S diet. Fatty acid composition of feed ingredients is an important factor that strongly influences the organoleptic properties of meat [22]. Variations in flesh fatty

acid compositions significantly affect organoleptic properties with unsaturated fatty acids, particularly those with more than two double bonds having high propensities to produce volatile, odourous, lipid oxidation products during cooking [22]. About 90% copra meal fatty acids are saturated [23]. Saturated fatty acids have strong resistance to the generation of lipid oxidation products and off-flavours which possibly explain why the copra meal diets did not induce discernable changes in the sensory responses of the cooked fillets of the fish groups fed these diets. The reductions in lipid oxidation rates in the fish muscles as a result of feeding diets containing copra meal can represent an inexpensive way of improving the shelf life of tilapia without negatively affecting fish growth and consumer perceptions of quality.

The possible high hepatic oxidation of the medium chain fatty acids (MCFAs) in copra meal coupled with the high deposition of excess dietary lipid as visceral fat rather than muscle fat could thus have accounted for the inability of the sensory panel to differentiate between the sensory qualities of the different dietary groups. There have been similar sensory studies in different fish species in which replacements of marine ingredients with plant proteins and vegetable oils resulted in panellists not being able to distinguish between the sensory characteristics of dietary treatments [18,24].

Economic analysis

The high replacement of fishmeal with copra meal resulted in significant reductions in feed costs. The 27 and 38% respective reductions in feed costs relative to the cost of the control diet by the CM and CM+S diets are very good indicators of significant reductions in investment costs over long-term production cycles using these two diets. With fishmeal being an expensive component of aquafeeds it is predictable that its replacement by plant protein sources often results in feeds with better comparative economical indices such as feed cost. Several studies [7,25,26,27] on the economic evaluation of plant-based aqua feeds vis-à-vis fishmeal-based aquafeeds have in fact indicated lower feed costs of plant-based diets relative to fishmeal-based control diets. However, in most of these studies the monetary advantage of using these low-cost diets is usually offset by lower growth and feed utilization parameters in target species, consequently reducing profitability in the long term. In this study, however, growth and feed utilization parameters of the Nile tilapia fed the copra meal diets were comparable to the control diet group, demonstrating the potential use of high levels of autoclaved copra meal in tilapia diets without compromising growth rates and extending culture periods.

Conclusion

In this study, it was possible to include autoclaved copra meal up to 680 g kg⁻¹ in Nile tilapia diets with no deleterious effects on fish growth. The remarkable acceptability of diets containing very high levels of copra meal coupled with the identical growth performance and feed efficiencies among the tilapia groups indicate the possibility of substantially reducing dietary fishmeal levels in Nile tilapia diets without compromising somatic growth



Figure 1 Formation and deposition of intraperitoneal fat in fish fed the control, copra meal and copra+ sesame meal diets (from L to R).

and feed utilization. The results of the sensory evaluation of the cooked tilapia fillets indicated that the high dietary inclusion of copra meal does generate flesh fatty acid compositions that significantly affect the sensory descriptors tested. From an economic perspective, the possibility of high inclusions of copra meal in tilapia diets represents high prospects of reducing feed cost in the face of the rising fishmeal prices.

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