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Performance of Diets for *Oreochromis niloticus* and *Tilapia zillii* by Inclusion of Cassava Leaf Protein Concentrate as Partial Replacement for Solvent-Extracted Soybean Meal

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ABSTRACT

Cassava (cultivar TME419) leaves were processed into leaf protein concentrate and evaluated in diets (320 g/kg protein, 80 g/kg lipid, 19.5 MJ/kg gross energy) fed to Oreochromis niloticus (3.2±0.3 g) and Tilapia zillii (3.3±0.4 g). A control diet (TD1) contained solvent-extracted soybean meal (SBM), which was substituted at 20%, 40%, 60% or 80% with the cassava leaf protein concentrate (CLPC) in test diets TD2, TD3, TD4 or TD5, respectively. Fishes were assigned in triplicate diet treatments (60 fish/treatment) in a complete randomized design (60 fish/treatment) and fed to apparent satiation twice daily for 70 days. Data were analysed statistically using one-way analysis of variance test. No fish mortality occurred in all treatments. Values obtained for growth response indices (mean weight gain, percentage weight gain) were statistically similar ($p > 0.05$) as CLPC substituted up to 60% and 80% of SBM in diets for O. niloticus and T. zillii, respectively, and there were good growth response and diet utilization. Fish growth declined and diet was poorly utilized beyond 60% or 80% these inclusion levels, caused by reduction in protein and energy digestibility. O. niloticus fed with diet TD5 showed slight histological alterations in livers; caused by residual anti-nutrients while T. zillii showed no alterations or abnormalities. Erythrocyte sedimentation rate was significantly higher ($p < 0.05$) in O. niloticus fed with diet TD5, while haematocrit, haemoglobin concentration, erythrocyte and leucocyte counts were statistically similar ($p > 0.05$). The suitability of CLPC as dietary protein for O. niloticus will depend on further reduction/removal of inherent anti-nutrients as well as improving digestibility of nutrients.

Key words: Cassava leaf protein concentrate, protein source, soybean replacer, tilapia diets

INTRODUCTION

Over 54.83 million tonnes of cassava (*Manihot esculenta* Crantz) was produced in Nigeria in 2014. Considerable amounts of cassava leaves waste away on farmlands after harvesting the roots, which is the main commercial product. Cassava leaf meal (CLM) is produced from whole leaves of the plant by dehydration to moisture content of 15-20%. The dried cake is then milled to produce cassava green meal which contains about 24% protein. Cassava leaf protein concentrate (CLPC) is produced by chopping the leaves and pressing the juice out. The extracted juice is coagulated with injection of steam and the soluble fraction is separated from the green curd, and the curd is dried to produce CLPC (Müller, 1977; Telek and Martin, 1983).

Cassava leaf has high protein content (average 21%, Ravindran, 1993) and contains higher levels of many of the essential amino acids than the levels found in soybean meal (Gómez and Valdivieso, 1984). The high contents of essential amino acids in cassava leaves is the reason for suggesting that it represents a potential protein source in aquafeeds (Fagbenro, 2013). However, the inherent anti-nutrients and toxic substances in cassava leaves may limit their use in aquafeeds, as they interfere with nutrient digestibility and uptake (Francis *et al*., 2001). According to Ravindran (1993), the major anti-nutrients associated with cassava leaves are the cyanogenic glycosides, linamarin and lotaustralin which on hydrolysis give hydrocyanic acid (HCN).

If leaves are not processed properly, the cyanide content remains (Ngudi *et al*., 2003).

Cassava leaf meal (CLM) when evaluated as protein source in diets for tilapia (Ng and Wee, 1989; Madalla, 2008) gave varied performance results. Poor growth performance and diet utilization of the CLM-based diets were due to poor diet binding ability due to high fibre content resulting in leaching of nutrients, poor palatability and hence reduced feed intake, methionine deficiency, presence of residual anti-nutrients, and poor nutrient digestibility. Supplementation of the 100% CLM diet with 0.1% methionine improved growth response slightly (Ng and Wee, 1989). Hence, it is imperative to evolve locally adaptable and environmentally friendly mode of utilization of cassava leaves; which will enhance the nutrient value, lead to the use in aquafeeds and reduce feed costs. Processing of cassava leaves into cassava leaf protein concentrate (CLPC) reduced the crude fibre content and increased protein quality (Oresegun *et al*., 2016).

Plant protein concentrates have an advantage over whole meals as their non-digestible fibre contents are eliminated which allows the use of high levels in aquafeeds. Higgs *et al*. (1982) included rapeseed protein concentrate in salmon diets, while Olvera-Novoa *et al*. (1990) showed that 35% substitution of fish meal with alfalfa leaf protein concentrates had no adverse effect on *O. mossambicus*. Olvera-Novoa *et al*. (1997) evaluated the effect of cowpea protein concentrate as partial substitute for fish meal protein in *O. niloticus* diets.

The use of soybean products (meal, protein concentrate) in aquafeeds is increasingly becoming unjustifiable in economic terms; as it is increasingly being used in human foods and for feeds by poultry and livestock (Tacon *et al*., 1998). Therefore, there is need to exploit cheaper plant protein sources to substitute soybean meal for sustainable aquaculture production, thus stimulating the use of alternative plant protein feedstuff sources that are locally and commonly available. Cassava leaves are appropriate for this purpose when processed into CLM or CLPC, as they are non-competitive feedstuff that can be developed as protein source in aquafeeds.

CLPC contains 45-50 g/kg crude protein and low anti-nutrient contents (Müller, 1977; Ravindran, 1993; Aletor 2010). In previous studies, CLPC produced from cassava cultivar (TME 419) was similar to SBM in terms of nutrient composition (Oresegun *et al*., 2016). CLPC has been evaluated in poultry diets (Aletor and Fasuyi, 2005) but studies on the use of CLPC in tilapia diets are limited (Bohnenberger *et al*., 2008, 2010). This study investigates the effects of graded levels of CLPC substituting SBM as dietary protein source on mortality, growth response, diet utilization, nutrient digestibility, liver histology, and haematological profile of two tilapias of aquaculture importance, *Oreochromis niloticus* and *Tilapia zillii*.

MATERIALS AND METHODS

Preparation and analyses of CLPC and experimental diets

Fresh leaves of cassava cultivar TME419 were collected from the International Institute for Tropical Agriculture (IITA), Ibadan, Nigeria. The leaves were weighed, washed, and pulped with a leaf pulping machine. This process was followed by crushing in water (1:3 w/v at a pH 8.5) and filtered as described by Modesti *et al*. (2007). The separated leaf juice was boiled at 90-100ºC for 10 minutes to coagulate leaf protein fraction, which was separated by a method of filtration and subsequently dried at atmospheric temperature. Proximate composition of CLPC was determined according to AOAC (2000) methods. Hydrogen cyanide, tannic acid and phytic acid contents were determined using Francis *et al*. (2001) methods. Gross energy content was determined using direct combustion in an adiabatic bomb calorimeter (Gallenkamp, UK).

Five isoproteic, isolipidic and isocaloric tilapia diets (320 g/kg crude protein, 80 g/kg crude lipid, 19.5 MJ/kg gross energy) were formulated and prepared. A control diet (TD1) contained SBM providing 60% of total protein. The SBM component was substituted at 20%, 40%, 60% or 80% with CPLC in test diets and labelled TD2, TD3, TD4, TD5, respectively. Lipid content of diets was adjusted with 1:1 mixture of corn oil and cod liver oil, while gelatinized cassava starch was added to adjust gross energy content of diets. All the ingredients were milled to <250µm using a laboratory mill, mixed thoroughly in a Hobart A120 steam pelleter and extruder (Hobart, Troy, Ohio, USA) to obtain a homogeneous mixture and passed through a die size of 2mm to obtain strands, which were oven-dried at 60ºC for 24 h. Dried strands were broken into 2mm sizes and stored at -20ºC until analysed. Proximate composition of diets was determined according to AOAC (2000) methods while gross energy content was determined using bomb calorimetry. Lysine and methionine contents were determined using an LKB 4151 Alpha plus amino acid analyzer after treating the hydrolysate with 6 mol. L^{-1} HCl under reflux for 24 h at 110 ºC.

Feeding of *O. niloticus* and *T. zillii* fingerlings

Groups of 20 apparently healthy, full sibling *O. niloticus* (3.2±0.3 g) or *T. zillii* (3.3±0.4 g) were stocked into 60-litre capacity glass tanks filled with aerated water. Each diet was fed to fish in triplicate tanks to apparent satiation twice daily (09.00h, 16.00h) for 70 days. Fish mortality was monitored daily, total fish weight in each tank was determined at two-week intervals, while the amount of diet fed was adjusted according to new fish weights. Water

temperature and dissolved oxygen concentration were measured daily while pH was monitored weekly. Alkalinity, hardness, phosphate, nitrite, and nitrate concentrations were determined according to the Stirling *et al*. (1985) methods. Growth response and diet utilization indices were determined as (1 – 3):

Weight gain = Final weight of fish - Initial weight of fish ……. (1)

Percentage weight gain = Mean weight gain/Mean initial weight x 100 ……. (2)

Feed gain ratio (FGR) = Total feed consumed by fish/Weight gain by fish … (3)

Apparent digestibility of nutrients by *O. niloticus* and *T. zillii*

Ten *O. niloticus* or *T. zillii* were stocked in 20-litre cylindrical plastic tanks filled with aerated water. Each diet was assigned to duplicate tanks and fishes were fed to apparent satiation twice daily (8.00-9.00h, 16.0-17.00h) for 14 days. On day 15, faeces were collected from the hind gut of each anaesthetized fish (2.5 ml quinaldine/litre) when they were dissected eight hours after feeding. Dry matter and crude protein contents were determined according to AOAC (2000) methods while gross energy content was determined using bomb calorimetry, in triplicate samples of diets and faeces Apparent nutrient digestibility was determined using the acid insoluble ash method (Atkinson *et al*., 1984) as: 100 -100 (AIA in feed x nutrient in faeces)/(AIA in faeces x nutrient in feed)

Haematological and histological examinations of *O. niloticus* and *T. zillii*

Fishes were anaesthetized with 200 mg/L MS222, their caudal peduncles severed and the blood collected. Thereafter, 2 ml blood was transferred into a tube containing ethylene-diamine-tetra-acetic acid (EDTA) as anticoagulant. The supernatant plasma was collected and stored at 4^{o}C prior to determination of erythrocyte count, leucocyte count, haematocrit, haemoglobin concentration and erythrocyte sedimentation rate were determined according to the methods of Svobodova *et al*. (1991). Livers of ten fish sample were removed, weighed and fixed in 1:10 buffered formalin solution, dehydrated in graded levels of ethanol, cleared with xylene, blocked in paraffin, sectioned at 5µ thickness, placed on glass slides, stained with haematoxylin and eosin, examined under a light microscope, and interpreted using the atlas of Morrison *et al*. (2006).

Statistical analyses

Data for growth response, diet utilization, nutrient digestibility indices, and haematological parameters from the fish feeding trials were subjected to one-way analysis of variance (ANOVA) test. Duncan's multiple-range test was applied to characterize and quantify the differences between treatments using Statgraphics 5+ package (Manugistics Inc. and Statistical Graphics Corp, MD, US.).

RESULTS AND DISCUSSION

The nutrient composition of CLPC produced from cultivar TME 419 is similar to that of SBM while its anti-nutrient composition is much lower than in the corresponding leaf meal (Oresegun *et al*., 2016). HCN levels of <5.3-80 g/kg are regarded as safe for animal feeds (Ravindran, 1993). Cassava leaves contain cyanogenic glycosides (linamarin and lotaustralin which on hydrolysis give HCN and appreciable levels of other anti-nutrients notably phytin and tannin. These anti-nutrients were degraded to non-deleterious levels and crude fibre content was reduced in the CLPC (Table 1), thus the TME 419 cultivar is classified as non-toxic. Processing cassava leaves into CLPC reduced the crude fibre content and increased protein quality (Oresegun *et al*., 2016).

Table 1: Nutrient and anti-nutrient compositions (g/kg dry matter) of soybean meal (SBM) and cassava leaf protein concentrate (CLPC)

Parameters	SBM	CLPC
Dry matter	889	989
Crude protein	485	489
Crude lipid	19	133
Crude fibre	37	67
Ash	62	39
Gross energy (MJ/kg)	18.92	21.35
Lysine (g/kg protein)	32	68
Methionine (g/kg protein)	8	25
Cyanide (g/kg)	ND	0.98
Phytates (%)	ND	1.63
Tannin (%)	ND	1.07

ND- Not determined

Proximate composition and gross energy content of the experimental diets (Table 2) showed that no differences occurred in protein, lipid, fibre and ash contents of diets for the two tilapia species. Crude protein and gross energy contents of the experimental diets ranged from 325.0 to 326.3 g/kg and 19.7 to 19.84 MJ/kg, respectively. They met the nutrient requirements for both tilapias recommended by Jauncey (2000) and El-Sayed (2006).

Water quality and survival of experimental fishes

Values of water quality parameters monitored during the feeding trials were 25.0-27.2ºC, 5.0-7.6 mg/litre, 7.5-9.1, 122-135 mg/litre, 290-320 mg/litre, 0.2-0.5 mg/litre, 0.12-0.15 mg/litre and 2-4 mg/litre for water temperature, dissolved oxygen, pH, alkalinity, hardness, phosphate, nitrite, and nitrate concentrations, respectively. The values were within the acceptable limits/range for tilapias culture (El-Sayed, 2006). No abnormal fish behaviour or signs of stress were observed in fishes in all treatments during feeding trials. Fish survival in all treatments during the feeding trials was 100% and was due mainly to good fish handling.

Feeding trials

During the feeding trials, acceptance of the diets was good as both *O. niloticus* and *T. zillii* became accustomed to the test diets within five days, indicating that the incorporation of CLPC had no adverse effects on palatability of the test diets by the experimental fishes. According to Pereira-da-Silva and Pezzato (2000), acceptance of diet by fish as a result of poor palatability is a major problem when plant protein sources are used in aquafeeds. In this study, the test diets were accepted by both tilapia species indicating that CLPC did not adversely affect the palatability of the diets.

Final weight and mean weight gain of *O. niloticus* and *T. zillii* fed with CLPC-based diets were not significantly different ($p>0.05$) to those fed with the control diet; but feed intake declined when CLPC progressively substituted SBM as dietary protein. Average daiiy gain ADG, feed intake and FGR values of *O. niloticus* and *T. zillii* fed diets containing CLPC up to 60% substitution of SBM were statistically similar ($p>0.05$) to those fed with the control diet, but at 80% substitution, growth and diet utilization indices were significantly different ($p<0.05$) (Table 3). These indicate that CLPC inclusion at the expense of SBM was beneficial up to 60% substitution for both *O. niloticus* and *T. zillii*.

Contrastingly, Ng and Wee (1989) and Chhay *et al*. (2010) reported poor growth in *O. niloticus* fed with diets containing cassava leaf meal as substitute for fish meal and ascribed the reduced growth to high fibre content and endogenous anti-nutrients of diets. Similarly, Wee and Wang (1987) recorded poor tilapia growth after feeding with diets containing *Leucaena* leaf meal and ascribed it to residual mimosine (toxic amino acid) content. Johansson *et al*. (l991) opined that inferior quality of protein concentrates is due to limited availability of lysine and/or methionine.

Olvera-Novoa *et al.* (1990) reported that lysine and methionine were limiting amino acids in alfalfa protein concentrates fed to *O. mossambicus,* particularly reducing growth at >35% fish meal (FM) substitution level. Table 2 shows that lysine and methionine contents in CLPC and SBM were complementary in diets TD2, TD3 and TD4 and produced good growth in both *O. niloticus* and *T. zillii*. Hence, poor growth response and diet utilization in *O. niloticus* fed with diet TD5 (Table 3) were not due to lysine or methionine deficiencies.

Table 2: Ingredient and proximate compositions (g/kg) of the experimental diets

	Control diet	Test diets			
	TD1	TD2	TD3	TD4	TD5
Fish (herring) meal	180	180	180	180	180
Soybean meal (solvent-extracted)	400	320	240	160	80
Cassava leaf protein concentrate	0	80	160	240	320
Cassava starch (gelatinized)	327	336	345	354	363
Corn oil: Cod liver oil (1:1)	53	44	35	26	17
Vitamin-Mineral premix[1]	20	20	20	20	20
Carboxymethyl cellulose (binder)	20	20	20	20	20
Crude protein	325	325.4	325.7	326	326.3
Crude lipid	80.4	80.5	80.6	80.8	80.9
Crude fibre	23.8	26.2	28.6	31	33.4
Ash	41.9	40.1	38.2	36.4	34.5
Gross energy (MJ/kg)	19.7	19.74	19.77	19.81	19.84
Lysine (g/kg protein)	25	27.8	30.5	33.2	35.9
Methionine (g/kg protein)	7.3	8.7	10	11.4	12.7

[1]Vitamin-Mineral mix (g/kg): Manufactured by DSM Nutritional Products Limited, Basle, Switzerland - Vitamin A, 1600 IU; vitamin D, 2400 IU; vitamin E, 160 mg; vitamin K, 16 mg; thiamin, 36 mg; riboflavin, 48 mg; pyridoxine, 24 mg; niacin 288 mg; panthotenic acid, 96 mg; folic acid, 8 mg; biotin, 1.3 mg; cyanocobalamin, 48 mg; ascorbic acid, 720 mg; choline chloride, 320 mg; calcium 5.2 g; cobalt, 3.2 mg; iodine, 4.8 mg; copper, 8 mg; iron, 32 mg; manganese, 76 mg; zinc, 160 mg; Endox (antioxidant) 200 mg.

Table 3: Growth response and diet utilization of *Oreochromis niloticus* and *Tilapia zillii* fed with the experimental diets

	TD1	TD2	TD3	TD4	TD5
O. niloticus					
Initial wt. (g)	3.2±0.3	3.2±0.3	3.2±0.3	3.2±0.3	3.2±0.3
Final wt. (g)	39.1±0.5^{a}	38.9±0.4^{a}	38.9±0.4^{a}	38.8±0.3^{a}	37.5±0.5^{a}
Weight gain (g)	35.9±0.2^{a}	35.7±0.4^{a}	35.7±0.5^{a}	35.6±0.3^{a}	34.3±0.2^{a}
% wt. gain	1121.9^{a}	1115.6^{a}	1115.6^{a}	1112.5^{a}	1071.9^{b}
FGR	1.55^{a}	1.57^{a}	1.59^{a}	1.67^{b}	1.69^{b}
FI (g/fish/day)	1.6^{a}	1.6^{a}	1.5^{a}	1.3^{b}	1.3^{b}
T. zillii					
Initial wt. (g)	3.3±0.4	3.3±0.4	3.3±0.4	3.3±0.4	3.3±0.4
Final wt. (g)	33.9±3.0	33.8±3.2	34.0±3.2	33.4±3.5	33.1±3.5
Weight gain (g)	30.6±2.5	30.5±2.3	30.7±2.3	30.1±2.3	29.8±2.3
% wt. gain	927.3^{a}	924.2^{a}	930.3^{a}	912.1^{ab}	903.0^{b}
FGR	1.55	1.55	1.55	1.58	1.6
FI (g/fish/day)	1.3	1.3	1.2	1.2	1.2

Mean values in a row with different superscripts are significantly different ($p < 0.05$).

FGR - feed gain ratio, FI - feed intake

Table 4: Apparent nutrient digestibility coefficient (%) of *Oreochromis niloticus* and *Tilapia zillii* fed with the experimental diets

	TD1	TD2	TD3	TD4	TD5
O. niloticus					
Dry matter	89.3±2.0^{a}	88.1±2.0^{a}	88.3±2.3^{a}	88.9±1.8^{a}	83.3±2.1^{b}
Crude protein	85.8±1.3^{a}	85.3±2.4^{a}	84.9±2.4^{a}	83.6±1.3^{a}	71.5±1.2^{b}
Gross energy	74.3±0.8^{a}	72.2±2.0^{a}	71.6±2.0^{a}	71.3±0.8^{a}	65.5±0.6^{b}
T.zillii					
Dry matter	84.5±2.5^{a}	83.3±3.6^{a}	80.6±3.1^{a}	81.1±3.0^{a}	75.8±2.5^{b}
Crude protein	82.2±1.1^{a}	80.9±1.4^{a}	80.6±1.6^{a}	80.2±1.4^{a}	70.8±2.0^{b}
Gross energy	72.8±3.4^{a}	71.6±1.2^{a}	71.3±1.2^{a}	70.6±3.0^{a}	65.1±1.2^{b}

Mean values in a row with different superscripts are significantly different ($p < 0.05$).

Table 5: Haematological profile of *Oreochromis niloticus* and *Tilapia zillii* fed with the experimental diets

O. niloticus	Hct (g)	Hb ($g.dl^{-1}$)	Ec ($x10^{12}.l^{-1}$)	Lc ($x10^{9}.l^{-1}$)	ESR (%)
TD1	28.2±1.5	2.1±0.2	2.2±0.1	53±5	46±2^{a}
TD2	27.7±1.1	2.4±0.3	2.3±0.1	50±4	45±3^{a}
TD3	27.3±1.5	2.3±0.3	2.0±0.1	52±2	48±2^{a}
TD4	27.6±1.0	2.4±0.2	2.1±0.1	53±4	46±1^{a}
TD5	27.4±1.2	2.3±0.1	2.1±0.1	52±4	57±4^{b}
T. zillii					
TD1	27.3±1.5	2.6±0.3	2.8±0.2	58±3	47±4^{a}
TD2	27.3±1.6	2.6±0.4	2.7±0.2	57±5	46±5^{a}
TD3	27.4±2.0	2.5±0.3	2.3±0.3	57±6	48±6^{a}
TD4	27.5±1.8	2.7±0.4	2.1±0.1	56±2	47±5^{a}
TD5	27.3±1.0	2.5±0.2	2.2±0.1	54±1	48±2^{a}

Mean values in a column with different superscripts are significantly different ($p<0.05$).

Hct - haematocrit, Hb - haemoglobin concentration, Ec - erythrocyte count, Lc - leucocyte count, ESR - erythrocyte sedimentation rate

Ogino *et al*. (1978) and Nour *et al*. (1985) showed that leaf protein concentrates produced from rye grass and water hyacinth partially substituted casein and FM in rainbow trout and mirror carp diets, respectively. Soliman (2000) reported that water hyacinth protein concentrate meal could safely substitute 30% of fish meal protein in tilapia diets. Chavez *et al*. (2016) included water hyacinth leaf protein concentrate, substituting up to 75% of dietary SBM for white shrimp, and recorded beneficial effects at 25% on growth compared to a control diet. In this study, the conversion to CLPC apparently degraded antinutrients and reduced fibre contents of diets thereby increasing nutrient availability.

Apparent nutrient digestibility by experimental fishes

In all experimental diet treatments, both *O. niloticus* and *T. zillii* had high apparent crude protein digestibility (ACPD) values (85-90%) which were not significantly different ($p>0.05$) (Table 4). High ACPD values were similarly reported by Olvera-Novoa *et al.* (1990) for *O. mossambicus* fed with diets containing alfalfa leaf protein concentrates. ACPD of CLPC-based diets for *O. niloticus* (Table 4) are similar to ACPD values for full-fat soybean meal (88.5%) recorded for *O. niloticus* by Fagbenro (1998). Bohnenberger (2008) obtained a lower ACPD of 66.57% for CLPC-based diet in *O. niloticus* juveniles (86.9±3.7g). Apparent gross energy digestibility (AGED) values were significantly reduced ($p<0.05$) when CLPC substituted 80% of SBM. The AGED of CLPC for tilapias (Table 4) are similar to AGED values for full-fat soybean meal (76.4%) reported for *O. niloticus* by Fagbenro (1998).

Haematological properties of experimental fishes

The haematological parameters obtained for both *O. niloticus* and *T. zillii* were not significantly different ($p>0.05$) (Table 5), and were within the normal range for tilapias (Basiao and Arago, 1988; Ezzat, *et al*., 1974). However, Oresegun and Alegbeleye (2001, 2002) reported variations in blood parameters in *O. niloticus* fed with dry practical diets containing cassava peels, the variations were attributed to the presence of residual anti-nutrients; which were ameliorated when the diets were supplemented with DL-methionine.

Histological properties of livers in experimental fishes

There were no disruptions to liver tissues of *T. zillii* but slight vascular and fatty changes occurred in livers of *O. niloticus* fed with test diet TD5. These changes are presumably due to the presence of residual anti-nutrients in the diet which may have caused disturbance in metabolism and/or mobilization of fat as suggested for carps fed high dietary levels of *Leucaena* leaf meal or mustard seed cake (Hossain and Jauncey, 1988; Hasan *et al*., 1991, 1994). Fatty changes in the liver of fishes fed with diets containing rapeseed meal or mustard oil cake were also reported for Coho salmon (Higgs *et al*., 1979) and common carp (Hossain and Jauncey 1989). Lack of alterations in hepatic structure of *T. zillii* suggests that it is relatively more tolerant of residual anti-nutrients in CLPC; since it is naturally a herbivore in the wild (Bowen, 1982: Arrignon, 1998).

CONCLUSIONS

Cassava leaf protein concentrate has similar nutritional value with solvent-extracted soybean meal and it supported good growth and diet utilization in *O. niloticus* and *T. zillii* without histological disorders in their livers, up to 60% and 80% substitution of SBM, respectively. When the CLPC was substituted at 80% of SBM in *O. niloticus* diet, growth response and diet utilization were compromised, which was caused by reduced feed intake, protein and energy digestibility. It also elicited alterations in the liver histology. Residual anti-nutrients were mainly responsible for these effects. The suitability of CLPC as dietary protein for tilapias will depend on further reduction/removal of inherent anti-nutrients as well as improving digestibility.

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