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RESEARCH ARTICLE

Effects of pure and crude papain on the utilization and digestibility of diets containing hydrolysed feather meal by Nile tilapia (Oreochromis niloticus L.)

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Abstract

This study was conducted to evaluate the effects of pure and crude papain on the digestibility and utilization of diets containing hydrolysed feather meal (HFM) by Nile tilapia (Oreochromis niloticus L.) under indoor and outdoor conditions. O.niloticus fingerlings with a mean weight between 22 and 30g were stocked in aquaria and cages respectively. Recirculating water was used in the aquaria while cages were installed in an 800m^2 earthen pond. The pond was fertilized with 20 kg N and 8 kg P ha^{-1} respectively. Liming was done once with 2500 kg ha^{-1} of $CaCO_3$ at the beginning of the experiment. Five isonitrogenous (250g CP kg^{-1}) and isocaloric (12.3 kJ g^{-1}) diets designated as 1, 2 3, 4 and 5 were formulated. Diet 1 contained 6 % freshwater shrimp meal (FSM) and 4.5 % HFM and served as control, Two other diets were formulated from the control by adding pure papain (Diet 2) and 4.5 % PLM (crude papain) (Diet 3). The other two diets were formulated by completely replacing FSM with HFM plus pure papain (Diet 4) and crude papain (Diet 5). All fish were fed at 10% body weight day^{-1} in three replicates for 58 days. Results indicated that dietary levels of HFM and PLM above 4.5% led to significant ($P < 0.05$) growth reductions in aquaria. However, complete replacement of FSM with HFM did not significantly ($P > 0.05$) affect growth of fish in the cages. In both experiments, survival was similar among treatments, but protein digestibility decreased with increasing levels of HFM in the diet. In conclusion, a combination of the protein sources FSM, HFM and PLM gave the highest growth performance in both aquaria and cages. The growth depression observed for treatments 4 and 5 in aquaria was not observed in the cages, where the natural food may have provided an important nutrient supplement.

INTRODUCTION

Aquaculture production in Africa is comparatively low and represents about 1.1 percent of the total world production (FAO, 2006). Most of the African aquaculture production is concentrated in the Mediterranean region, representing over 65% of the total African production (Pedini & Shehadeh, 1997). A review conducted by FAO (2005) reported that sub-Saharan countries are lagging behind in aquaculture development; mainly due to lack of a well developed aquaculture fish feeds. Furthermore, Shang (1992) and Craig & Helfrich (2002) emphasized the importance of inexpensive and efficient feeds in fish farming, because fish feeds represent over 50 % of the total variable production costs.
Tilapia culture is one of the fastest growing enterprises worldwide with more than 1.3 million tons produced in 2000 (FAO, 2002). Among the tilapias, Nile tilapia (Oreochromis niloticus) is the principle culture species and represents 44 % of global tilapia production (Rana, 1997). O. niloticus is an excellent candidate for semi-intensive aquaculture production because of its rapid growth, tolerance to poor water quality, high reproductive rates and relatively disease resistant (Stickney 1986). Furthermore, O.niloticus feeds at the lowest trophic level and is therefore accustomed to covering much of their nutritional demand from ponds' natural food (Fitzsimmons, 1997). These attributes makes O.niloticus a suitable choice for fish farmers within sub-Saharan Africa where semi-intensive culture in fertilized earthen ponds is the common aquaculture practice.

In Kenya the main source of animal protein in fish feeds remains fishmeal (Rastrionaebola argentea) and freshwater shrimps (Carridina nilotica). The supplies of both resources are limited due to periodical closure of Lake Victoria during the fish breeding season and competition from other feed manufactures and the cost is in the upward trend (Munguti et. al, 2006; Liti et. al., 2005). So far, researchers have focused on alternative protein sources

Fishmeal remains a key protein component in diets for O. niloticus (Abdelghany, 2003; Tacon, 1993) due to complete composition of amino acid profiles. However the declining levels and high cost makes fishmeal un-attractive protein source. Plant protein sources are plenty and locally available in most aquaculture regions but have imbalance of amino acid profile (Hardy, 1996). To overcome imbalance in essential amino acids and antinutritional factors, research on feed substitutes in O.niloticus has focused on non-conventional animal protein as well as mixtures of plant and animal feedstuffs (Robinette and Dearing, 1978; Borgeson et al., 2006; Bishop et al., 1995; El-Sayed, 1998; Fowler, 1982; Liti et al., 2005; El-Saidy and Gaber, 2003). In Kenya, however, freshwater shrimp which is mainly from Lake Victoria fishery and is increasingly becoming scarce in the market partly due to competition from livestock feed manufactures and the frequent closures of the lake's fishery during breeding season. Therefore, there is urgent need to search for inexpensive and effective replacements for FSM in O.niloticus feeds.

Papaya carica is widespread in Kenya and the leaves are not used for human food. The leaves have a good crude protein (Munguti et al., 2006) and the green parts of the plant contain papain (Chaplin 2005), which degrades protein into amino acids. Furthermore papain promotes proteolytic digestion thereby increaseing utilization of protein from papaya leaf meal (Buchanan 1969). On the other hand, poultry farming is a common practice and is widespread across Kenya and generates large volumes of feathers as a by-product, which can be obtained at no cost. The protein content of feather meal is high (Hasan et al., 1997; Tacon et al., 1984), and utilization of hydrolyzed feather meal protein in tilapia feeds could economically be feasible, but studies conducted to evaluate the use of hydrolysed feather meal (HFM) in fish diets indicated low substitution levels due to low digestibility and sub-optimal levels of essential amino acids (Jauncey and Ross, 1982; Hasan et al., 1997; Falaye, 1982; Steffens, 1994; Santiago and Lovell, 1988; Mendoza et al. 2001). A combination of HFM and papaya leaf meal (PLM) may promote the feeding value of HFM and by extension promote its use in O.niloticus feeds. Liti et al. (2005) reported that in Kenya the lack of feeds specifically designed for semi-intensive culture of O.niloticus has forced farmers to utilize feeds designed for intensive culture systems and this has resulted in increased production costs. Therefore the current study was conducted to evaluate the effects of a varying rate of inclusion of papaya carica leaves and hydrolyzed feather meal on growth performance and apparent digestibility of O. niloticus under intensive (aquarium) and semi-intensive (cage-cum-pond) culture conditions.

Materials and Methods

In vitro and in vivo digestibility experiment descriptions

A pre in vitro digestibility test was conducted to determine the effect of pure papain (Carlroth - GMBH, Karlsruhe, Germany) on hydrolysed feather meal. Fifteen ml of distilled water and 20 mg of Papain were added to each of two batches of 30 g feathers, which had been hydrolysed either at pH 5 or at pH 9. The mixture was stirred and maintained at room temperature for 19 hours. An aliquot of 2 g was subjected to in vitro N-digestibility in 10 ml of diluted HCl (25 % v/v), the samples were incubated at 40°C for 24 hours. After adding 10 ml HCl (25 %) for the second time, incubation continued for another 24 hours, after which the suspension was filtrated and washed with hot water until pH 7 was reached. The filters, together with the residue were subjected to Kjeldahl analysis (AOAC, 1995). The result was interpreted as "indigestible" N and in vitro digestibility was calculated as the relative difference between total and "indigestible" N and expressed as percentage.

The in vivo studies were conducted at Sagana Fish Farm (90 km northeast of Nairobi, altitude 1230 m, latitude 0°39´S and longitude 37°12´E). Two experiments were conducted in aquaria each with a dimension of 0.45m x 0.3m x 0.3m. Net cages with a top frame of 1.2 x 0.94 m and a base (0.9x0.9m) and a height of 0.75m were installed in a 20m x 40m x 0.75m earthen pond. The aquaria were set in a thermoregulated recirculating system, comprising a settling tank for solids removal and an aerobic bio filter (tickling filter) to remove ammonia.

Experimental animals, design and feeding

The diets were randomly allocated to groups of hand-sexed male O. niloticus fingerlings that were held in aquaria and cages. The experimental diets were tested both in aquaria and cages, using three replicates per treatment. The initial average stocking weight in the aquaria and cage experiment was 23 and 30g, respectively, at densities of 6 and 10 fish per aquaria and cage respectively, respectively. Fish were acclimatised for two weeks prior to the start of each experiment. Feed was offered at 10% of body weight per day for 58 days. Sampling was done on a bi-weekly basis to monitor growth and adjust the amount of feed offered. Fish in aquaria were hand fed four times a day while those in cages were fed using the automatic feeders described by Waidbacher et al. (2006). The feeders were calibrated to deliver feed continuously between 8.00 and 18.00 hrs. At the end of the study, all fish from cages and aquaria were harvested, weighed and counted.

Pond and water quality management

The experimental pond was fertilized weekly at a rate of 20 kg N and 8 kg P ha^{-1} with urea and diammonium phosphate (DAP), respectively, and limed once with 2500 kg ha^{-1} $CaCO_3$ at the beginning of the experiment. Key water quality parameters, which included temperature, pH, dissolved oxygen (DO) and chlorophyll a were measured three times a week in the aquaria and cage experiments. DO was measured using model 57 oxygen meter (YSI industries, Yellow springs, OH, USA), while a glass electrode pH meter, Hi-9024 microcomputer (Hanna Instruments Ltd., Chicago, IL., USA), was used to take pH measurements. Chlorophyll a was determined as described in APHA (1990)

Feed ingredients and diet formulation

Green papaya leaves were collected from gardens in the neighbourhood of Sagana aquaculture centre, while chicken feathers were sourced from hotels around Sagana town and sun-dried. The chicken feathers were reduced to smaller sizes with a hammer mill, and thereafter hydrolysed by cooking them in an autoclave at mean pressure of 40-50 psi and a mean temperature of 140-150^{o} C for one hour at a pH of 9. After feather hydrolyzation, part of the wet hydrolyte was mixed with green papaya leaves using a blender-liquidizer (model A989, Hampshire, UK) and left overnight. The hydrolysed feathers as well as the mixture of papaya leaves and the remaining part of hydrolysed feathers were sun-dried and further reduced to a fine powder using the blender-liquidizer as given above. Freshwater water shrimp meal was purchased from Kisumu fish supply stores, while cotton seed cake was supplied by Goldstar Company, Nairobi. Wheat bran was purchased from Maisha millers, Nyeri. Carboxyl methyl-cellulose was used as a filler material in the diet formulation. All the ingredients were ground to fine powder before being subjected to proximate analysis.

Five experimental diets, each containing 250 g kg^{-1} of CP and 2,940 kcal kg^{-1} energy were formulated: diets of treatments 1 to 3 contained 6 % of freshwater shrimp meal (FSM) plus 4.5 % HFM (control treatment 1), supplemented either with papain (treatment 2) or 4.5 % PLM (treatment 3). In two diets FSM was substituted by 8.6 % HFM plus papain (treatment 4) or 8.6 % HFM plus 8.6 % PLM (treatment 5). In order to keep energy and CP levels constant, diets contained different levels of wheat bran and carboxyl methyl cellulose as a filler.
Each ingredient was homogeneously ground and passed through a 100µm sieve. The formulations were made by mixing the ingredients into a homogenate, which was moistened before passing through a modified meat mincer. The resulting expeller-like strands were sun-dried and stored at room temperature.

Proximate analysis

The nutrient contents of the ingredients was analysed prior to diet formulation. The proximate analysis of the feeds was carried out in triplicates (ALVA, 1983; AOAC, 1995). The analyses involved the following nutrients: dry matter (DM), crude protein (CP), ether extract (EE), ash, nitrogen free extracts (NFE) and crude fibre (CF). CP was estimated from Kjedahl nitrogen, while EE was quantified as the loss in weight after extraction of the sample with petroleum ether. Ash was determined by burning dry samples in a muffle furnace at 550 ^{o}C for 4 hours. CF was determined by a consecutive alkaline-acid digestion, which was followed by ashing the dry residue at 550 ^{o}C in a muffle furnace for 4 hours. NFE was determined by the difference method (DM-CP-EE-CF-Ash; ALVA, 1983). Amino acid content was analysed using HPLC after a 20 hour hydrolysation process with 6 molar HCl and a previous stabilisation by use of tryptophan with $Ba(OH)_2$. Separation of amino acids was made using a hyperphil ODS 250 x 4 mm-column after pre-column derivatisation with OPA (orthophtalaldehyde) (ALVA, 1983; Degussa, 1986; Altmann, 1992; Commission of the

European Union, 1998). The indigenous inert marker (insoluble ash) for digestibility was determined using the method described by Bowen (1981).

Evaluation of growth performance

The growth performance of the experimental fish was evaluated by measuring the parameters mean final weight, weight gain and specific growth rate. Specific growth rate (SGR) was calculated using the following equation:

$$SGR(\%) = \frac{(\ln Wf - \ln Wi)x100}{t}$$

Where Wi and Wf are the initial and final mean body weights, respectively and t is time in days from stocking to harvest.
The apparent digestibility coefficient of protein (ADCp) was calculated using the following formula:

$$ADCp(\%) = \frac{100x(1-(\%MD)x(\%NF)}{(\%MF)x(\%ND)}$$ (Maynard & Loosli, 1962)

Where NF is the nutrient content of faeces; ND is the nutrient content of the diet; MD, is the marker content of the diet, and MF is the marker content of the faeces.

Data analysis

Data were sorted and subjected to a one-way analysis of variance (ANOVA); Duncan multiple range test (Duncan, 1955) was applied to identify means that were significantly different from each other. A type I error of 0.05 was used to declare significance.

Results

Water quality

Water quality did not vary significantly ($P > 0.05$) among treatments, both in cages and in aquaria over the culture period. The mean values for chlorophyll a was 173.4 ± 15.2 mg m^{-3}. The ranges of DO values were 2.5 - 4.5 mg L^{-1} for morning and 6.7 - 11 mg L^{-1} for afternoon. Mean water temperature in the experimental pond and aquaria was 27.5 ± 1.0 ^{o}C. The mean pH values were 8.2 ± 0.04 in the experimental pond and 8.1 ± 0.01 in the aquaria. All the water quality parameters monitored were within the recommend values for tilapia culture (Popma and Masser, 1988).

Proximate analysis of feed ingredients

The proximate composition of the feedstuffs used in diet formulation is shown in table 1, while the composition of experimental diets and their proximate composition are shown in Table 2.

Table 1: Proximate composition of the feedstuffs used in diet formulation (g kg^{-1}); SD = Standard deviation, DM = Dry matter, CP = Crude protein, EE = Ether extracts, CF = Crude Fiber, NFE = Nitrogen Free Extract, FSM = Freshwater shrimp meal, HFM = Hydrolysed feather meal.

Ingredients	Nutrient (mean ± SD)					
	DM	CP	EE	CF	NFE	Ash
FSM, sundried	875±0.8	603±0.3	14±0.4	62±0.0	67±1.3	254±0.2
HFM, sundried	899±0.8	807±0.2	18±0.2	32±0.2	42±1.3	101±0.1
Cotton seed cake	898±0.5	349±0.2	128±0.1	258±0.4	205±0.3	60±0.1
Papaya leaf meal, sundried	901±0.5	253±0.2	97±0.1	116±0.4	395±0.3	138±0.1
Wheat bran	880±1.2	140±0.1	59±0.1	136±0.3	602±0.6	63±0.3

Table 2: Composition and results of proximate analysis (on dry matter basis) of experimental diets; DE = digestible energy, NFE = Nitrogen Free Extracts, SD = standard deviation.

Ingredients	Treatment 1 (control)	Treatment 2 (control + papain)	Treatment 3 (control + papaya leaves)	Treatment 4 (substitution of FSM + papain)	Treatment 5 (substitution of FSM + papaya leaves)
Freshwater shrimp meal, %	6.0	6.0	6.0	0.0	0.0
Hydrolysed feather meal, %	4.5	4.5	4.5	8.6	8.6
Cotton seed cake, %	25	25	25	25	25
Wheat bran, %	60	60	53	60	45
Papaya leaves, %	0.0	0.0	4.5	0.0	8.6
Carboxyl methyl cellulose, %	4.5	4.5	7.0	6.4	12.8
Papain (g/kg)	-	0.5	-	0.5	-
Proximate analysis (± SD)					
Dry matter (%)	90.2±0.7	89.9±0.4	90.1±0.8	89.7±0.5	90.5±0.3
Protein (%)	25.2 ±0.1	25.6 ±0.3	25.7±0.1	26.1±0.3	25.9±0.3
Ether extract (%)	6.1±0.4	6.0±0.5	6.2±0.1	5.8±0.1	5.9±0.5
Crude fibre (%)	14.9±1.1	15.3±1.2	16.9±1.3	15.9±1.1	16.7±1.7
NFE (%)	46.1±1.3	45.5±1.9	43.8±1.5	46.1±1.7	45.6±1.8
Ash (%)	7.7±0.3	7.6±0.2	7.4±0.1	6.1±0.3	5.9±0.2
DE (kcal g^{-1})	2.94	2.94	2.94	2.94	2.94

Growth performance

Data on fish performance in aquaria and cages are presented in Table 3 and 4 respectively. Fish that were fed diets containing 4.5 % HFM, eventually supplemented with papain or PLM (diets 1, 2, 3) in aquaria had similar mean weight, specific growth rate and percent weight gain, but grew significantly better ($P<0.05$) than those fed diets in which FSM was completely replaced by HFM and supplemented with pure papain (diet 4) or PLM (diet 5). Fish fed diets 4 and 5, in which FSM was substituted by HFM plus papain or HFM and PLM weighed significantly less than fish from treatments 1 to 3 at the termination of the experiment. There was lower weight gain in treatments 4 and 5 as compared to treatments 1 to 3, with the difference between treatments 5 and 3 being significant. However, specific growth rate was not significantly different between treatments. Survival rate was high and similar ($P>0.05$) among treatments both in aquaria and cages (Table 3 and 4). Growth trend curves for O.niloticus in cages and aquaria are presented in figures 1 and 2 respectively. In aquaria, fish fed diets containing 6.0 % FSM (treatments 1, 2 and 3) separated from treatments in which FSM was substituted by HFM or HFM and PLM (treatments 4 and 5) three weeks after stocking. In cages, a similar pattern was observed: diet 3 (containing 4.5 % each of HFM and PLM) resulted in the highest growth, while diet 5 (containing 8.6 % each of HFM and PLM) resulted in the lowest growth. The growth of fish that fed on diet 1, 2 and 4 was intermediate, although the differences were not significant ($P>0.05$).

Table 3: Growth performance of O. niloticus fed diets in aquaria containing different levels of hydrolysed feather meal and papaya leaf meal in place of freshwater shrimp meal; values with the same superscript are not significantly different at α = 0.05, SD = standard deviation of mean.

Parameter	Treatment 1 (control)	Treatment 2 (control + papain)	Treatment 3 (control + papaya leaves)	Treatment 4 (substitution of FSM + papain)	Treatment 5 (substitution of FSM + papaya leaves)	SD
Initial body weight (g)	23.1^{a}	23.0^{a}	23.1^{a}	23.5^{a}	23.0^{a}	0.17
Final body weight (g)	37.6^{bc}	36.7^{b}	39.3^{c}	31.1^{a}	34.4^{a}	3.48
Weight gain (%)	62.8^{bc}	59.6^{bc}	70.1^{c}	38.2^{a}	49.6^{a}	3.51

Specific growth rate (% day^{-1}) (SGR)	0.84 [b]	0.81 [b]	0.92 [b]	0.56 [a]	0.69 [a]	0.15
Survival (%)	99.0[a]	98.0 [a]	98.7[a]	96.1[a]	95.1[a]	0.92

Table 4: Growth performance of O.niloticus fed diets in cages containing different levels of hydrolysed feather meal and papaya leaf meal in place of freshwater shrimp meal; values with the same superscript are not significantly different at α = 0.05; SD = standard deviation of mean.

Parameter	Treatment 1 (control)	Treatment 2 (control + papain)	Treatment 3 (control + papaya leaves)	Treatment 4 (substitution of FSM + papain)	Treatment 5 (substitution of FSM + papaya leaves)	SD
Initial body weight (g)	30.5[a]	30.0[a]	30.2[a]	30.5[a]	30.2[a]	0.17
Final body weight (g)	49.2[b]	50.2[b]	51.2[b]	48.0 [a]	46.6 [a]	2.42
Weight gain (%)	61.3 [ab]	67.3 [ab]	69.5 [b]	57.4 [ab]	54.3 [a]	2.41
Specific growth rate (% day^{-1}) (SGR)	0.86[a]	0.88 [a]	0.92 [a]	0.81[a]	0.75 [a]	0.09
Survival (%)	98.2[a]	97.1 [a]	96.7[a]	98.1[a]	97.1[a]	0.42

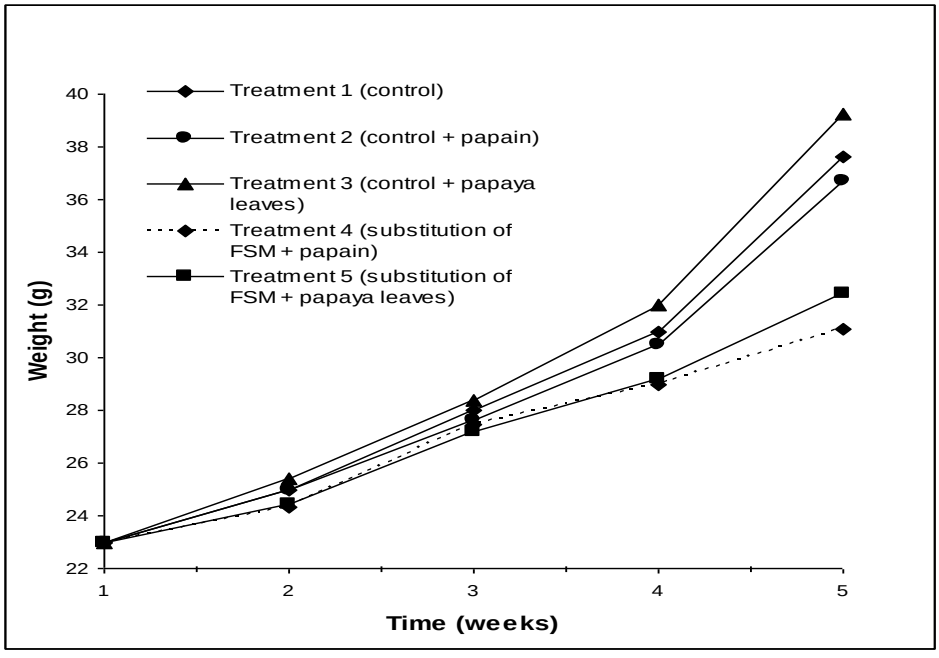

Figure 1: Growth curves for O.niloticus receiving diets with varying levels of HFM and papaya leaf meal in aquaria during 58 days culture period.

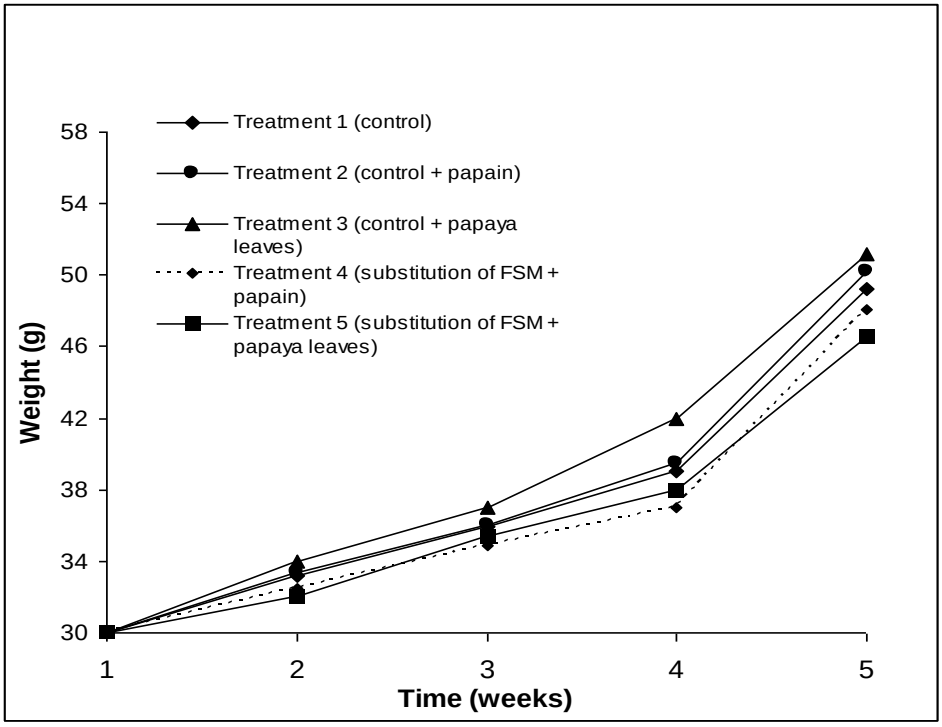

Figure 2: Growth curves for O.niloticus receiving diets with varying levels of HFM and papaya leaf meal in cages installed in a fertilized earthen pond during 58 days culture period.

Protein digestibility

Results from in vitro digestibility indicated that in vitro digestibility coefficient of protein (IDC_p) for hydrolysed feathers was best at an alkaline pH 9 which had 35 % compared to acidic pH 5 which had IDC_p of 27 %. Feathers hydrolysed without adjustment of the water pH had IDC_p of 29 %. Therefore the feathers which were used in the feeding experiment were hydrolysed at a pH of 9. The results from the in vivo experiment for apparent protein digestibility (ADC_p) in aquaria and cages are shown in Table 5. Amino acid composition of the different feed ingredients is shown in Table 6. In the aquaria experiment, significant differences ($P<0.05$) in ADC_p were observed between treatments. The ADC_p values decreased with increasing levels of HFM both in cages and aquaria (Figure 3).

Table 5: Apparent protein digestibility coefficient (APDC) in cages and aquaria; values with the same superscript are not significantly different at α=0.05; ADC_p=Apparent Protein Digestibility Coefficient.

Parameter	Treatment 1 (control)	Treatment 2 (control + papain)	Treatment 3 (control + papaya leaves)	Treatment 4 (substitution of FSM + papain)	Treatment 5 (substitution of FSM + papaya leaves)	SD
ADC_p (cage), %	68.5^a	71.5^a	72.0^a	64.5^a	65.6^a	0.39
ADC_p (aquaria), %	67.1^b	68.2^b	69.8^b	56.1^a	57.5^a	0.96

Table 6 Amino acid requirements for O.niloticus and digestible amino acid composition of feed ingredients (g kg^{-1} DM) AA=Amino acid, FSM=freshwater shrimp meal, HFM=Hydrolysed feather meal, PLM=Papaya leaf meal, WB=wheat bran, CSM=Cotton seed meal. Amino acid requirements for O.niloticus (Santiago & Lovell, 1988)

		Experimental diet ingredients				
Amino acid (%)	O.niloticus AA requirements (g/kg	HFM	PLM	FSM	WB	CSM

	DM)		
Essential			
Lysine	51.2	14.9	12.7
Methionine	26.8	3.9	4.1
Threonine	37.5	38.4	10.5
Tryptophan	10.0	5.5	4.5
Arginine	42.0	70.2	12.0
Phenylalanine	37.5	46.4	11.8
Histidine	17.2	4.7	6.4
Isoleucine	31.1	46.4	8.4
Leucine	33.9	82.9	15.5
Valine	28.0	74.5	11.2

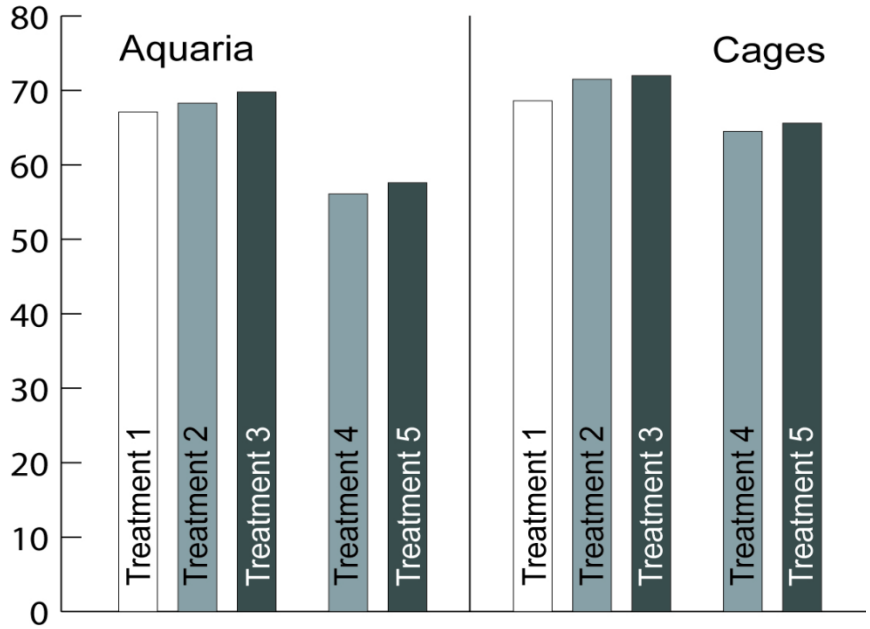

Figure 3: Apparent protein digestibility (%) of the test diets in cages and aquaria

Discussion

Results on growth performance from the present study indicated that growth of O. niloticus that were fed diets containing FSM plus a mixture of HFM and papain or PLM in cages were similar to that of the control treatment. However, there was a significant decline in growth when fish were fed diets containing 8.6 % of HFM protein (100% protein substitution level) in aquaria experiments.

Specific growth rate (SGR) of fish in cages was high and similar among all treatments. However, SGR in aquaria differed significantly between diets containing 4.5 % and 8.6 % HFM. The differences between the SGR in aquaria and in cages may be attributed to the environmental variations caused by the culture conditions. The aquaria experiment in the current study was conducted in indoor, while the cage experiment was conducted in a fertilized earthen pond. Therefore it is possible that the fish in the cage experiment got some extra nutrients from the natural pond food, a specific potential strong point of O.niloticus (Kaliba et al., 2006; Rakocy and Mcginty, 1989). This source of nutrients may have provided an extra supply of limiting essential amino acids. Furthermore, Bowen (1980) reported that tilapias can thrive well on naturally occurring dissolved amino acids. Hydrolyzed feather meal is often considered to be an inferior source of protein for fish because of its poor digestibility and unbalanced essential amino acid profile (Falaye, 1982; Hasan et al., 1997; Mendoza et al., 2001; Roley et al., 1977; Tacon et al., 1984). We therefore postulate that the interaction between the natural pond food and HFM may have resulted in an improved dietary amino acid profile, thus leading to better utilization and growth performance of fish in the cages.

Falaye (1982) did not observe a significant decrease in growth parameters (SGR, weight gain, mean weight) of O. niloticus when 50% of fish meal (FM) was replaced with HFM. These results are similar to our findings when HFM was used to substitute FSM. In relation to Tilapia diets which are commonly used in the study area, treatments 1, 2 and 3 also represent diets in which 50 % of FSM

are successfully substituted by HFM. In another study, similar results were found by Viola and Zohar (1984), who reported that up to 50% of FM, could be replaced by poultry by-products in diets of tilapia hybrids without deleterious effects on fish growth. The results of the present study were also similar to those of Abdel-Warith et al. (2001); from experiments with African catfish (Clarias gariepinus). This author could replace 40% of fish meal protein with protein from poultry by-products without a significant reduction in growth. In another study with O. niloticus, Bishop et al. (1995) reported a slightly higher level of substitution (66%) of FM-protein with HFM protein without a significant reduction in growth. This relatively high substitution level has to be related to the specific culture conditions. While part of the present experiment was conducted in aquaria indoors, Bishop et al. (1995) used concrete tanks in which additional nutrients from natural food may have been available. However, in the experiment with cages installed in the earthen pond, complete substitution of FSM by HFM and papain or PLM only moderately reduced growth. This may have been due to the extra source of nutrients from the natural pond food, as the pond was fertilized weekly.

From previous research it was concluded that hydrolysed feather meal as a potential component in fish diets contains an unbalanced amino acid pattern (Santiago and Lovell, 1988; Jauncey and Ross, 1982; NRC, 1983). Therefore, the reduced growth recorded in fish fed diets containing over 4.5 % HFM in the aquaria experiment in the present study may be attributed to sub-optimal levels of essential amino acids. This in part, might have hindered a proper utilization of protein from HFM for body protein synthesis. Fisher et al, (1981) and Harrap and Woods (1964) reported that the major component of feathers is ß-keratin which has a high degree of cross-linking of disulfide bonds, hydrogen bonding and hydrophobic interactions. Fraser et al., (1969) noted that keratin in its natural form is insoluble and difficult to digest by humans and animals.

The low nutritional value and insolubility of native feather protein is a consequence of the composition and molecular configuration of constituent amino acids that ensure the structural rigidity of the feathers (Parry and North, 1998). The use of physical and chemical treatments to convert feathers to feather meal for feeding purposes can destroy certain amino acids and decrease protein quality and digestibility (Moritz and Latshaw, 2001; Wang and Parsons, 1997). Therefore to improve feather digestibility research has to focus on microbial proteolytic systems, e.g. streptommyces fradia (Nickerson et al., 1963; Noval and Nickerson 1958), supplemented with methionine (Elmayergi and Smith (1971), Bacillus licheniformis (Williams and Shih, 1989). However, these feather degradation technologies are rather complicated for rural farmers to apply. This is specifically true for developing countries where it is desirable to utilize feathers as a component of diets produced on-farm. Therefore studies have been conducted to evaluate feather hydrolysation through cooking the feathers under high pressure and temperature (Hasan et al., 1997; Bishop et al., 1995; Papadopulos et al., 1985).
Gohl (1981) reported that feathers are insoluble due to their high content of keratin and that by autoclaving disulfide bonds are broken, thereby making the feathers more soluble and digestible. However, the content of some essential amino acids may be reduced; e.g. cystine percentage decreases from about 10 to 3.5 %. O.niloticus weight gain reported for treatment 3 in both the aquaria and cage environment in the present study may have been due to increased digestibility and nutritive value of the overall diet which also contained green papaya leaves plus ponds natural food. Green papaya carica leaves are known to contain proteases of the papain superfamily and bleomycin hydrolases which potentially improves protein digestion (Croall and Dermartino, 1991; Enenkel and Wolf 1993; Brocklehurst et al., 1987). However, a positive effect of the inclusion of PLM could not be shown for a situation in which FSM was completely substituted by HFM and PLM; treatments 4 and 5. To improve the nutritive value of diets based on hydrolysed feather meal, some researchers have suggested to supplement deficient amino acids in diet formulations (Webster et al. 1991; Murai et al. 1982; El-Sayed 1990), but given the low market prices for tilapia in developing countries, this approach does not seem economically viable in a situation which this study was related to. Miller et al., (1989) reported that papain has very broad specialties and therefore indiscrminatively breaks down major muscle (connective tissue, collagen and myofibrillar proteins). Middlebrook and Philips (1941) further reported that in presence of an alkaline (sodium bisulphate) solution wool, which is mainly keratin was rapidly attacked by papain. Therefore the use of papaya carica leaves which contain papain may be a viable way of promoting the quality of feather meal based diets.

Webster et al. (1991) suggested that utilization of amino acids is also dependent on the feeding frequency. Thebault (1985) reported that added amino acids were converted into other compounds such as methionine sulfoxide and that in methionine-supplemented fish diets plasma methionine reached its peak level substantially sooner than was the case for the rest of the EAA. Thus, the latter may not be (fully) utilized for protein synthesis when fish are fed once a day. However, in the current study the good growth recorded in the cages may have been due to amino acid utilization from the natural environment, because tilapias feed continuously in a semi-intensive environment (Getachew and Fernando, 1989; Moriarty, 1973; Zenebe and Getachew, 1998). Moreover, Bowen (1980) reported that tilapias can do well on naturally occurring dissolved amino acids. Therefore, based on the hypothesis of meal frequency, utilization of free amino acids might be enhanced in the cages were O.niloticus feeds continuously from the feed delivered by automated feeders and also from natural food. This mode of feeding may ensure that free amino acids are supplied continuously in relevant amounts at the site of tissue formation. Since no significant differences ($P>0.05$) were recorded in O.niloticus growth among all the cage treatments in the present study, the observation by Bowen (1980) would suggest that supply of amino acids from natural food present in well managed, fertilized ponds may be an effective and economical strategy of supplementing the limiting amino acids in O.niloticus diets.

Apparent protein digestibility (ADC_p) in the present study was above 65% except for treatment 4 in aquaria setup. This may have been due to a more complete hydrolysation by setting the pH to 9. However, the ADC_p declined if the inclusion rate of HFM increased from

4.5 % to 8.6 % in the diets. A similar trend was reported for Carp (Cyprinus carpio L.) and gibel carp (Carassius auratus gibelio Bloch) by Yang et al. (2006) and Degani et al. (1997), respectively who observed a decrease in ADC_p with increasing levels of feather meal. However, the ADC_p values calculated in the present study are generally slightly lower than those reported for O. niloticus by Hanley (1987). The difference may be due to the composition of the test diets. Hanley (1987) tested hydrolyzed feather meal as single (isolated) feed component, while in the present study; the diets consisted of mixtures of ingredients. It appears from the present study that the presence of natural food improved the protein digestibility in feather meal based diets in the cage experiment (Table 5, Fig. 3), so that there were no significant differences observed between diets containing FSM (treatments 1 to 3) and diets in which FSM has been completely substituted (treatments 4 and 5). The mechanism for the improvement may be linked to the presence of algae. Algae or algal based diets are reported to stimulate secretion of copious amounts of gastric acid in O. niloticus and O. mossambicus (Getachew, 1987, 1989; Bowen 1981). Bowen (1981) further reported that gastric acid at a pH uncommonly low for fishes (pH<1.5) decomposes large amounts of inorganic matter and liberates protein while Moriarty (1973) ascertained that the stomach pH is the lowest during the afternoon, when the fish's stomach is full.

In conclusion, a combination of FSM, HFM and PLM can be suggested as a locally available protein source which will allow a relatively high performance while at the same time improving the sustainability of semi-intensive O. niloticus production. The additional nutrient supply from the pond's natural food may help to avoid the growth depression caused by ingestion of formulated rations containing less digestible protein. After going through this discussion it appears more of a literature review than a discussion!. The same points are repeated using different words and this detracts the attention of the reader. There is need to refocus the discussion from story telling to the reality of our results.

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References

Abdelghany, A. E. (2003): Partial and complete replacement of fishmeal with gambusia meal in diets of red tilapia (Oreochromis niloticus x O.mossambicus). Aquaculture Nutrition, 9: 145 - 151.

Abdel-Warith, A. A., Russell, P. M. and Davies, S. J. (2001): Inclusion of a commercial poultry by-product meal as a protein replacement of fishmeal in practical diets for African catfish, Clarias gariepinus (Burchell 1822). Aquaculture Research, 32: 296 - 305.

Altmann, F. (1992): Determination of amino sugars and amino acids in glycoconjugates using precolumn derivatization with O-Phthaladehyde. Analytical Biochemistry, 204: 215-219.

ALVA. (1983): Arbeitsgemeinschaft Landwirtschaftlicher Versuchsanstalten in Österreich: Österreichisches Methodenbuch für die Untersuchung von Futtermitteln, Futterzusatzstoffen und Schadstoffen. Wien.

AOAC. (1995): Official Methods of Analysis of the Association of Official Analytical Chemists, 16th Edition. Arlington, VA, USA, 1018pp.

APHA. (1990): Standard Method for Examination of Water and Wastewater, 17th edn. Pp. 10-203. American Public Health Association, Washington, DC.

Bishop, C. D., Angus, R. A. and Watts, A. (1995): The use of feather meal as a replacement for fishmeal in the diet of Oreochromis niloticus fry. Bioresource Technology, 54: 291-295.

Borgeson, T. L., Racz, V. J., Wilkie, D. C., White, L. J. and Drew, M. D. (2006): Effects of replacing fishmeal and oil with simple and complex mixtures of vegetable ingredients in diets fed to Nile tilapia (Oreochromis niloticus). Aquaculture Nutrition, 12: 141-149.

Bowen, S. H. (1981): Digestion and assimilation of periphytic detrital aggregrate by Tilapia mossambica. Transactions of the American Fisheries Society, 110: 239 - 245.

Bowen, S. H. (1980): Detrital Non-protein Amino Acids are the Key to Rapid Growth of Tilapia in Lake Valencia, Venezuela Science, 207: 1216 - 1218.

Brocklehurst, K., Willenbrock, F. and Salih, K. (1987): Cysteine proteinases. In Hydrolytic Enzymes (Neuberger, A. and Brocklehurst K., eds). Elsevier Biomedical Press, Amsterdam. pp. 39 – 158.

Buchanan, R. A. (1969): In vivo and in vitro methods of measuring nutritive value of leaf protein preparations. British Journal of Nutrition, 23: 533.

Bureau, D. P., Harris, A. M. and Cho, C. Y. (1998): The effects of purified alcohol extracts from soy products on feed intake and growth of Chinook salmon and rainbow trout. Aquaculture, 161: 27-43.

Chaplin, M. (2005): Application of proteases in the food industry. Faculty of Engineering Science and the build environment, South bank University Press. London. UK.

Commission of the European Union, (1998): Commission directive 98/64/EC of 3.9.1998 establishing Community methods of analysis for the determination of amino acids, crude oils and fats and olaquindox in feeding stuffs and amending. Directive 71/393/EEC. Official Journal of the European Communities, L257: 16 - 23.

Craig, S. and Helfrich, L. A. (2002): Understanding Fish Nutrition, Feeds and Feeding, Cooperative Extension Service, publication. Virginia State University, USA. 420-256 pp

Croall, D. E. and Demartino, G. N. (1991): Calcium–activated neutral protease (capain) system: structure, function and regulation. Physiological Reviews, 71: 813 - 847.

Degani, G., Viola, S. and Yehuda, Y. (1997): Apparent digestibility coefficient of protein sources for carp, Cyprinus carpio L. Aquaculture Research, 28: 23-28.

Duncan, D. B. (1955): Multiple Range and Multiple F-Tests. Biometrics, 11: 1-42.

Elmeayergi, H. H. and Smith, R. E. (1971): Influence of growth of streptomyces fradiae on pepsin-HCL digestibility and methionine content of feather meal. Canadian Journal of microbiology, 17:1067-1072.

El-Saidy, D. M. S. and Gaber, M. M. A. (2003): Replacement of fish meal with a mixture of plant protein sources in juvenile Nile tilapia, Oreochromis niloticus (L) diets. Aquaculture Research, 34: 1119-1127.

El-Sayed, A. F. M. (1990): Long-term evaluation of cotton seed meal as a protein source for Nile tilapia, Oreochromis niloticus. Aquaculture, 84: 315 - 320.

El-Sayed, A. F. M. (1998): Total replacement of fishmeal with animal protein sources in Nile tilapia Oreochromis niloticus (L) feeds. Aquaculture Research, 29: 275 - 280.

Enenkel, C and Wolf, D. H. (1993): BLH1 codes for a yeast thiol-aminopeptidase, the equivalent of a mammalian bleomycin hydrolyase. Journal of Biological Chemistry, 268: 7038 - 7043.

Fagbenro, O. A. (1998): Apparent digestibility of various legume seed meals in Nile tilapia diets. Aquaculture International, 6: 83 - 87.

Falaye, A. E. (1982): The use of hydrolysed feather meal alone or in combination with supplemental amino acids as a dietary source for tilapia (Oreochromis niloticus) MSc thesis, University of Stirling, UK.

FAO 2002. The State of World Fisheries and Aquaculture 2002. FAO Fisheries Department, FAO, Rome, Italy.

FAO 2005. Regional review on aquaculture development for sub-Sahara Africa. FAO Fisheries Circular, No 1017/4. FAO, Rome, Italy.

FAO 2006. State of world aquaculture 2006. FAO Fisheries Technical Paper 500, Rome, Italy.

Fitzsimmons, K. (1997): Introduction to tilapia nutrition. In: Tilapia Aquaculture: Proceedings from the fourth International Symposium on Tilapia in Aquaculture (Fitzsimmons, K. ed.) pp. 9 -12. Northern Regional Agricultural Engineering Service (NRAES) Publication No. 106, Ithaca, NY.

Fowler, L. G. (1982): Tests with poultry by-product meal in fall Chinook salmon diets. U.S. Fish and Wildlife Service, Abernathy Salmon Cultural Development Center. Techn. Trans. Ser. No. 82, 1-14 pp.

Fraser, R. D. B., MacRae, T. P., Parry, D. A. D. and Suzuki, E. (1969): The structure of ß-keratin. Polymer, 10: 810 - 826.

Gaber, M. M. (2006): Partial and complete replacement of fish meal by broad bean meal in feeds for Nile tilapia, Oreochromis niloticus L., fry. Aquaculture Research, 37: 986 - 993.

Getachew, T. and Fernando, C. H. (1989): The food habits of a herbivorous fish (Oreochromis niloticus L.) in Lake Awasa, Ethiopia. Hydrobiologia, 194:195 - 200.

Getachew, T. (1987): A study on an herbivorous fish (Oreochromis niloticus L.,) diet and its quality in two Ethiopian Rift valley lakes, Awasa and Zwai. Journal of fish Biology, 30: 439 -449.

Getachew, T. (1989): Stomach pH, feeding rhythm and ingestion rate in Oreochromis niloticus L. (Pisces: Cichlidae) in Lake Awasa, Ethiopia. Hydrobiologia, 174: 43 - 48.

Gohl, B. (1975): Tropical feeds. Food and Agricultural Organisation of the United Nations, Rome, Italy. p 528.

Hanley, F. (1987): The digestibility of foodstuffs and the effects of feeding selectivity on digestibility determinations in tilapia, Oreochromis niloticus (L.). Aquaculture, 66: 163 - 179.

Hardy, R. W. (1996): Alternative protein sources for salmon and trout diets. Animal feed science technology, 59: 71 – 80.

Hasan, M. R., Haq, M. S., Das, P. M. and Mowlah, G. (1997): Evaluation of poultry-feather meal as a dietary protein source for Indian major carp, Labeo rohita fry. Aquaculture, 151: 47-54.

Hecht, T. (2000): Consideration on African aquaculture. World Aquaculture, 31: 12 - 19.

Hossain, M. A., Focken, U. and Becker, K. (2003): Anti-nutritive effects of galactomannan rich endosperm of sesbania (Sesbania aculeate) seeds on growth and feed utilization in tilapia, Oreochromis niloticus. Aquaculture Research, 43(3): 1171-1179.

Hughes, S. (1990): Feather meal can displace fishmeal in aquaculture rations. Feed international, 15: 13 -15.

Jauncey, K. and Ross, B. (1982): A guide to tilapia feeds and feeding. Institute of Aquaculture, University of Stirling, Scotland. U.K.

Kaliba, A. R., Osewe, K. O., Senkondo, E. M., Mnembuka, B. V. and Quagrainie, K. K. (2006): Economic analysis of Nile Tilapia (Oreochromis niloticus) production in Tanzania. Journal of the World Aquaculture Society, 37: 464 - 472.

Liti, D., Kerogo, L., Munguti, J. and Chhorn, L. (2005): Growth and economic performance of Nile tilapia (Oreochromis niloticus) (L.) fed on two formulated diets and two locally available feeds in fertilized ponds. Aquaculture Research, 36: 746 - 752.

Liti, D., Waidbacher, H., Straif, M., Mbaluka, R. K., Munguti, J. M. and Kyenze, M. M. (2006): Effects of partial and complete replacement of freshwater shrimp meal (Caridina nilotica Roux) with a mixture of plant protein sources on growth performance of Nile tilapia (Oreochromis niloticus L.) in fertilized ponds. Aquaculture Research, 37: 477 - 483.

Lovell, R. T. (1980): Nutritional value of solid by-products from ethanol production of corn. Auburn Technical Assistance Centre, Auburn, AL 7pp.

Maynard, A. L. and Loosli, K. J. (1962): Animal nutrition (5th edition) McGraw-Hill, New York, 613pp.

Mbahinzireki, G. B., Dabrowski., K. J., Lee, K., El-Saidy, D., and Wisner, E. R. (2001): Growth, feed utilization and body composition of tilapia (Oreochromis sp.) fed cottonseed meal based diets in a recirculating system. Aquaculture Nutrition, 79:189 - 200.

Mendoza, R., De Dois, A., Vazquez, C., Cruz, E., Ricque, D. Aguilera, C. and Montemayor, J. (2001): Fishmeal replacement with feather-enzymatic hydrolyzates co-extruded with soya-bean meal in practical diets for pacific white shrimp (Litopenaeus vannamei). Aquaculture Nutrition, 7: 143-151.

Middlebrook, W. R. and Philips, H. (1941): The application of enzymes to the production of shrinkage resistant wool and mixture fabrics. Journal of the Society of Dyers and Colourists, Bradford, 57: 137.

Miller, A. J., Strange, E. D and Whiting, R. C. (1989): Improved tenderness of restructured beef steaks by a microbial collagenase derived from Vibrio B-30. Journal of food Science, 54(4): 855 - 857.

Moritz, J. S. and Latshaw, J. D. (2001): Indicators of nutritional value of hydrolysed feather meal. Poultry Science, 80: 79 - 86.

Munguti, J. M., Liti, D. M. Waidbacher, H. Straif, M. and Zollitsch, W. (2006): Proximate composition of selected potential feedstuffs for Nile tilapia (Oreochromis niloticus L.) production in Kenya. Die Bodenkultur, Austrian Journal of Agricultural Research, In Press.

Murai, T., Ogata, H. and Nose, T. (1982): Methionine coated with various minerals supplemented to soybean meal diet for fingerling carp, Cyprinous carpio and channel catfish Ictarulus punctatus. Bulletin of the Japanese Society of Science Fisheries, 48: 85 - 88.

National Research Council (NRC) (1983): Nutrient requirement of warm water fishes and shellfishes. National Academic Press, Washington, D.C. USA.

New, M. B. and Csavas, I. (1993): A summary of information on aquafeed production in eleven Asian countries. In: M.B. New, A.D.J. Tacon & I. Csavas (eds) Farm-made aquafeeds. Proceedings of the FAO/AADCO Regional Expert Consultation on Farm-Made Aquafeeds, 14-18 December 1992, Bangokok, Thailand. FAO-RAPA/AADCP, Bangokok, Thailand, 434 p.

Ng, W. K. and Wee, K. L. (1979): The nutritive value of cassava leaf meal in pelleted feed for Nile tilapia. Aquaculture, 83: 45 - 58.

Olvera-Novoa, M. A., Olivera-Castillo, L. and Martinez, P. C. A. (2002): Sunflower seed meal as a protein source in diets for Tilapia rendalli (Boulanger, 1896) fingerlings. Aquaculture Research, 33: 223 - 229.

Papadopulos, M., El Boushy, A. and Ketelaars, E. (1985): The effect of different processing conditions on amino acid digestibility of feather meal determined by chicken assay. Poultry Science, 64: 1729 - 1741.

Pedini, M. and Shehadeh, Y. (1997): Aquaculture in sub-Saharan Africa: Situation and outlook, FAO Aquaculture Newsletter, 15: 3 - 6. Rome.

Philippart, J. C. and Ruwet, J. C. (1982): Ecology and distribution of tilapias. In: R.S.V. Pullin & R.H. Lowe-McConnell (Editors), The Biology and Culture of Tilapias. ICLARM conference proceedings, International Centre for Living Aquatic Resources Management, Manila, The Philippines, pp 15-59.

Popma, T. and Masser, M. (1989): Tilapia life history and biology. Southern Regional Aquaculture Center, SRAC Publication No. 283.

Rakocy, J. E. and Mcginty, A. S. (1989): Pond culture of tilapia. Southern Regional Aquaculture Center, SRAC Publication No. 280.

Rana, K. J. (1997): Global trends of aquaculture: a FAO statistical perspective. Paper presented at the First International Meeting: Population Genetics and Aquaculture in Africa, 1-4 April 1997. Grand Bassam, Cote d`Ivoire.

Robinette, H. R. and Dearing, A. S. (1978): Shrimp by-product meal in diets of channel catfish. Progressive Fish Culturist, 40 (1): 39 - 40.

Roley, D. D., Roley, S. E., Hardy, R. W. and Brannon, E. L. (1977): Feather meal and candida yeast as substitute for fish in the diets of Chinook salmon fry (Oncorynchus tshawytscha). Annual Report of the college of Fisheries, University of Washington, Seattle, WA, 70 pp.

Santiago, C. B. and Lovell, R. T. (1988): Amino acid requirement for growth of Nile tilapia. Journal of Nutrition, 118: 1540 - 1546.

Shang, Y. C. (1992): The Role of Aquaculture in the World Fisheries", Presented at the World Fisheries Congress, Athens, Greece, May 3 - 8, 30 pp

Shao, J., Den, S. and Gao, J. (2002): Detoxifying and nutrition analysing of castor beanmeal. Chinese Journal of Health Laboratory Technology, 2:157 – 158

Sklan, D., Prag, T. and Lupatsch, I. (2004): Apparent digestibility coefficients of feed ingredients and their prediction in diets for tilapia Oreochromis niloticus x Oreochromis aureus (Telepstei, Cichlidae) Aquaculture Research, 35:358 - 364.

Sokal, R. R. and Rohlf, F. J. (1981): Biometry, The principles and practice of statistics in biological research, W. H. Freeman and Company, San Francisco, USA.

Steffens, W. (1994): Replacing fishmeal with poultry by product meal in diets of rainbow trout (Onchorhynchus mykiss). Aquaculture, 124: 27 - 34.

Stickney, P. R., Hesby, J. H., McGeachin, R. B. and Isbell, W. A. (1979): Growth of Tilapia niloticus in ponds with differing histories of organic fertilization. Aquaculture, 17: 189 - 194.

Tacon, A. G. J. (1993): Feed ingredients for warm water fish. Fishmeal and other processed feedstuffs. FAO Fisheries Circular No.856, 64pp FAO Rome.

Tacon, A. G. J. (1997): Fishmeal replacers: a review of anti-nutrients within oilseeds and pulses, a liminiting factor for the aquafeed green revolution. In: Feeding Tomorrows Fish. Vol 22 (Tacon, A. and Basurco, B. Eds), Cahiers Potions Mediterraneennes, Institut Agronomique Mediterraneen de Zaragoza Spain. pp. 153-182.

Tacon, A. G. J., Jauncey, K., Falaye, A., Pantha, M., McGowen, I. and Stafford, E. (1984): The use of meat and bone meal, hydrolyzed feather meal and soyabean meal in practical fry and fingerling diets for Oreochromis niloticus. In: Proceedings of the First International Symposium on Tilapia in Aquaculture, (Fishelson, L. & Yaron, Z. eds), Tel Aviv University Press, Tel Aviv, Isreal. pp. 356 - 365.

Thebault, H. (1985): Plasma essential amino acid changes in sea-bass (Dicentrarchus lobrax) after feeding diets deficient and supplemented in L-methionine. Comparative Biochemistry and Physiology, 82: 223 - 233.

Viola, S. and Zohar, G. (1984): Nutrition studies with market size hybrids of tilapia (Oreochromis niloticus) in intensive culture. Bamidgeh, 36: 3 - 15.

Waidbacher, H., Liti, D. M., Fungomeli, M., Mbaluka, R. K., Munguti, J. M. and Straif, M. (2006): Influence of pond fertilization and feeding rate on growth performance, economic returns and water quality in a small-scale cage cum-pond integrated system for production of Nile tilapia (Oreochromis niloticus L.) Aquaculture Research, 37: 594 - 600.

Wang, X. and Parsons, C. M. (1997): Effect of processing systems on protein quality of feather meal and hog hair meal. Poultry Science, 76: 491- 496.

Webster, C. D., Tidwell, J. H. and Yancey, D. H. (1991): Evaluation of distillers' grains with soluables as a protein source in diets for channel catfish. Aquaculture, 96: 179 - 190.

Williams, C. M. and Shih, J. C. H. (1989): Enumeration of some microbial groups in thermophilic poultry waste digesters and enrichment of feather-degrading culture. Journal of applied bacteriology, 67: 25 - 35.

Yang, Y., Xie, S., Cui, Y., Zhu, X., Lei, W. and Yang, Y. (2006): Partial and total replacement of fishmeal with poultry by-products meal in diets of gibel carp, Carassius auratus gibelio Blouch. Aquaculture Research, 37: 40 - 48.

Zenebe, T. and Getachew, T. (1998): Diel feeding rhythm and assimilation efficiency of Oreochromis niloticus L, Pisces: Cichlidae) in Lake Ziway, Ethiopia. Verhandlungen Internationaler Verein der Limnologie, 26:2324 - 2328.