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Maize gluten meal as a protein source in the diets for African catfish *Clarias gariepinus* (Burchell, 1822) and its effect on liver glycogen and histology

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ABSTRACT

This study was conducted to assess the potential use of maize gluten meal (MG) as a substitute protein source for the African catfish, *Clarias gariepinus*. Four experimental diets formulated with different substitution levels of MG (control 0, 25, 50 and 75%) for fish meal (LT94), were fed to African catfish. The African catfish that were fed the high inclusion levels of MG showed significant differences ($p>0.05$) in gwoth performance. Weight gain ranged from 86.73 g for the control group to 19.50 g for the high inclusion level group (75% MG); specific growth rate (SGR) ranged from 4.89 to 2.59; feed conversion ratio (FCR) ranged between 0.85 and1.58; and the apparent net protein utilisation (ANPU) values ranged from 51.53 to 25.32%. In all cases, significant lower performances were recorded in the 75% MG substitution group ($p>0.05$). Furthermore, liver glycogen increased with increasing MG levels. Further, histological examination of liver tissue revealed alterations in the hepatic structure associated with higher levels of MG. These data suggest that no more than 25% of fishmeal should be substituted with maize gluten meal under the conditions tested in this study.

Keywords: African catfish, Growth performance, Histology, Liver glycogen, Maize gluten

Introduction

The African catfish, *Clarias gariepinus,* is a warm water aquaculture species occurring in countries in Africa and Asia and has been recently introduced in Europe and Latin America (FAO, 1997; Ali, 2001). The superior performance of *C. gariepinus* compared with other *Clarias* species in terms of growth rate has likely contributed to the fact that *C. gariepinus* has been widely introduced to areas outside its natural range (Verreth-Huskens and Segner, 1993). The African catfish is a heavily exploited food source in many parts of the world. Annually, in terms of global aquaculture production of fish, catfish is ranked as the fourth most widely cultivated fish species after carp, salmon and tilapia (FAO, 2002). Fishmeal, which has been conventionally used as the main source of animal protein in catfish feeds, is becoming increasingly expensive for a viable and beneficial aquaculture enterprise worldwide because of a reduction in supply from capture fisheries, particularly in the tropics (Naylor *et al.,* 2000; Hardy and Tacon, 2002). Although there is evidence of some differences in the utilisation of plant products between and within fish species, most studies support the potential of successful replacement of fishmeal by plant protein ingredients with a high protein component (Goda *et al.,* 2007; Davies and Gouveia, 2008; Imorou Toko *et al*., 2008) for African catfish (Mente *et al.,* 2003), Atlantic salmon (Amirkolaie *et al.,* 2006; Abdel-Warith, 2008; Al-Asgah *et al.,* 2011) and Nile tilapia.

Maize gluten meal is being used in livestock production worldwide (Parson, 1998). However, its use in fish feed has been limited to amounts that serve as a source of energy and filler in diet formulation. Because fish require more protein than land animals (Wilson, 1991), more attention has been focused on high quality protein sources to meet these requirements. Maize gluten meal is an important byproduct of maize processing, during which most of the starch, bran and germ are removed (Hardy, 1989). Maize gluten has considerable potential to replace fishmeal in diets up to a certain level without any effect on the growth and health of rainbow trout (Moyano *et al.,* 1991) or Atlantic salmon (Mente *et al.,* 2003). Additionally, maize gluten does not contain anti-nutritional factors (Parson, 1998) like most grains and seed oils that are used as feed ingredients in catfish diets. However, diets containing maize gluten are known to be deficient in lysine (Regost *et al.,* 1999). It is therefore an attractive proposition to use high lysine corn instead of normal corn to reduce the need for other protein supplements (Wu *et al.,* 1999).

Previous investigations have reported on a range of different fish species that were fed relatively high inclusions of plant proteins, including African catfish fed maize gluten with amino acids supplementation (Falaye and Oloruntuyi, 1998; Fasakin *et al.,* 2006; Goda *et al.,* 2007), tilapia fed corn gluten (Wu *et al.,* 1999), channel catfish (Robinson *et al.,* 2001) and Atlantic salmon fed Maize gluten (Mente *et al.,* 2003). Corn gluten meal, which contains more than 60% protein, is the major protein portion obtained from the wet milling process used to separate corn into its starch, germ, protein and fiber components.

The objectives of this study were to investigate the effects of partial substitution of fishmeal (LT94) with plant protein sources (maize gluten) for African catfish to determine maximum inclusion levels for this species and to study the effect of maize gluten on fish growth, feed utilisation, liver glycogen and histology.

Materials and methods

Experimental fish

African catfish with an average weight of 5.97 g were graded and stocked in fiber glass tanks. All treatments were tested on duplicate groups of fish. Another group of twenty fish were sacrificed using a lethal concentration of benzocaine and kept frozen at -20°C for later determination of initial carcass composition.

Experimental diets

Four experimental diets were formulated containing variable proportions of maize gluten meal (MG). The control diet (D1) contained 0% MG and used fishmeal (LT94) as its protein source. Each remaining diet replaced 25%, 50% or 75% of the fishmeal with equivalent amounts of MG (D2, D3 and D4, respectively). The four experimental diets were isoenergetic and isonitrogenous and were adjusted to contain 35% crude protein and 16% lipids. Table 1 shows the feed formulation and proximate composition of the experimental diets.

Feeding regime

Fish were fed on Trouw Aquaculture pellet for one week to acclimate them to the treatments, the system and to clean their gastrointestinal tract from the pre-experimental diet. At the end of the acclimation period, the fish were weighed prior to initiation of feeding the experimental diets. During the experiment, fish were fed at 4% of their body weight twice a day and fish were weighed, every fourteen days. Daily feed intake was recorded throughout the eight weeks study period. On termination of the experiment, the final weight of each fish was taken following 24-h starvation. Five fish from each treatment group were randomly selected from the experimental tanks and sacrificed for carcass analysis. Another set of ten fish from each treatment group were euthanised and the liver was removed for glycogen determination, and general hepatic morphology as well as for histological studies.

Proximate composition

The proximate chemical compositions of moisture, protein, lipid, ash and gross energy for both the diets as well as fish tissue were determined according to AOAC (1995).

Determination of amino acids

The amino acid contents of the diets were determined following acid hydrolysis nethod of McCullagh *et al.,* (2006).

Table 1. Ingredients composition and proximate content of the control and test diets (g100 g^{-1} dry weight).

Ingredients	Diets			
	D1	D2	D3	D4
Fish meal[1]	42.00	31.50	21.00	11.00
Maize gluten[2]		10.50	21.00	32.00
Wheat meal[3]	35.00	35.00	35.00	35.00
Blood meal	2.00	2.00	2.00	2.00
Corn oil[4]	4.40	4.45	4.57	4.65
Cod liver oil[5]	4.40	4.45	4.57	5.65
Vitamin premix[6]	2.00	2.00	2.00	2.00
Mineral premix[7]	1.00	1.00	1.00	1.00
Binder[8]	2.00	2.00	2.00	2.00
αCellulose[9]	7.20	7.10	6.86	4.70
Proximate composition (% as fed)				
Moisture	3.73	4.98	6.72	4.11
Protein	35.22	35.72	35.46	34.96
Lipid	16.24	16.66	16.71	17.86
Ash	7.30	6.16	4.95	3.90

[1] Fish meal LT94, Trouw Aquaculture (Nutreco Company); [2] Maize gluten, Cargill Ltd.; [3] Wheat meal, KalproS™., Orsan, Paris, France
[4] Mazola- pure corn oil; [5] Fish oil- seven pure cod liver oil; [6] Vitamin premix,Trouw Aquaculture (Nutreco Company);
[7] Mineral premix, Trouw Aquaculture (Nutreco Company); [8] Carboxymethyl cellulose (CMC). [9] Sigma Chemical Co., Poole, Dorset

Approximately 20-25 mg each of ground sample was weighed into 5 ml vials with 4 ml (6.6 M) of HCl and 1 ml (0.1 M) phenol, each vial was sealed and placed in an oven for 22 h at 110°C.

Amino acids were assayed using a Kontron Chromakon 500 automatic amino acid analyser [column 250 × 4.6 mm, cation ion-exchange resin material (AS70)]. The mobile phase was a gradient of sodium citrate-based buffers having the following composition: (i) 0.22 M sodium citrate, adjusted to pH 3.2 with concentrated HCl, +8% v/v methanol; (ii) 0.067 M sodium citrate + 0.5 M sodium chloride adjusted to pH 3.79 with concentrated HCl; (iii) 0.067 M sodium citrate + 1.4 M sodium chloride adjusted to pH 4.3 with concentrated HCl.

Detection was by a post column reaction with ninhydrin (in 4 M lithium acetate buffer, pH 5.2, flow rate 12 ml h^{-1}) at 115°C in a reaction oven followed by visible absorption measurement at 570 nm and 440 nm to produce a mean signal for quantitative integration. Dilution was made by loading buffer (2.2 pH sodium citrate buffer) and suitable 100 ml aliquots injected into the rheodyne automatic injection valve. The amino acid profiles are presented in Table 2.

Histological examination

On termination of the feeding trial, five fish from each group were sacrificed, and their livers were removed for histological examination. Hepatic tissues of the control and the treated fish were fixed in 10% neutral buffered formalin and the samples were then processed for standard histological evaluation. Sections of 5µm were prepared and stained with hematoxylin and eosin stains as described by Luna (1968) and Bernet *et al.* (1999). The histological slides were observed in a VANOX model AHBT Olympus microscope and photomicrographs were taken.

Determination of liver glycogen

Glycogen content was determined using a method derived from Plummer (1987). Glycogen was liberated from the tissue by heating with KOH, and then precipitated with ethanol using sodium sulfate as a co-precipitate to give a quantitative yield.

Statistical treatment of data

Data were analysed using a one-way analysis of variance (ANOVA) technique. The means were separated by Fisher's LSD test and compared using Duncan's Multiple Range Test, as described by Snedecor and Cochran (1989).

Results

Growth performance

The growth data for the African catfish fed the four different diets are displayed in Table 3. There was significant difference ($p>0.05$) in the final average body weight between the experimental groups. Fish fed the fishmeal-based (D1) diet exhibited the highest increase in final average body weight (92.70 g), and the lowest growth was observed for fish fed the D4 diet, which contained the highest amount of maize gluten in place of fish meal (75% replacement) (25.47 g). These fish had only an approximate 4-fold increase in weight after 8 weeks of feeding. These trends were also evident with respect to absolute weight gain, which decreased as the level of plant protein inclusion increased. The control diet resulted in the greatest weight gain of 86.73 g, while the MG 75% diet produced the lowest weight gain of 19.50 g.

The specific growth rate (SGR) values further support this trend with significant differences ($p>0.05$). The SGR decreased from 4.89 for the control fish to 2.59 for the fish fed 75% MG diet. Fish fed on 50% MG diet (D3)

Table 2. Essential amino acid composition (expressed as % of protein) of control and test diets containing different levels of maize gluten meal fed to African catfish.

Amino acids	Diets				African catfish Requirements*
	D1	D2	D3	D4	
Arginine	5.65	4.92	5.19	4.37	-
Histidine	2.81	2.58	3.31	2.81	-
Isoleucine	3.64	3.58	4.59	3.86	-
Leucine	7.37	8.90	16.54	13.87	-
Lysine	6.39	4.24	4.21	3.58	5.70[1]
Methionine	2.38	2.23	2.82	2.35	3.20[1]
Methionine + Cysteine	2.71	2.65	3.63	3.01	-
Phenylalanine	3.91	4.74	7.29	6.19	-
Phenylalanine + Tyrosine	6.48	8.02	12.94	10.76	-
Threonine	4.00	4.17	4.63	4.33	-
Valine	4.42	4.14	5.29	4.39	-
Tryptophan	ND	ND	ND	ND	-

*Requirements for all amino acids have not been determined for African catfish.
[1]Fagbenro *et al.* (1998a, b).
ND : not detected.

performed better than those on the 75% MG (D4) diet, although there was little difference between the 25% inclusion level (D2) and the control diet (Table 3). No mortality was observed during the course of the experimental period.

Feed consumption and feed utilisation

All diets were well accepted by the fish except the D4 diet, which had the highest level of maize gluten (75% MG). The mean daily feed intake ranged between 1.32 and 0.55 g fish^{-1} day^{-1}. There was significant differences ($p>0.05$) in feed intake, with more D1 feed consumed than D4 feed due to the inclusion of alternative protein sources, as shown in Table 3.

The feeding rates of the control group were higher compared to those of the fish fed diets containing MG. The feed conversion ratio (FCR) differed significantly ($p>0.05$) between treatments, and the values followed the same apparent trend as with final weight gain and specific growth rate. The lowest FCR was obtained for catfish fed the 75% MG diet, with a value of 1.58, while significantly greater values ($p>0.05$) were obtained for the other three groups. Fish fed the control diet had the best FCR (0.85), compared to those receiving the maize gluten diets.

Protein efficiency ratio (PER) also showed significant differences ($p>0.05$) between treatments, with fish fed the control and 25% MG diets having the highest PER (3.33) and those fed high levels of plant protein MG having the lowest values (1.81). Apparent net protein utilisation (ANPU) was also significantly different ($p>0.05$) between treatments in the same manner as PER. Catfish fed the control diet exhibited the highest ANPU value (51.53) compared to the treated groups; in particular, catfish fed the high inclusion level diet (75%MG) yielded the lowest ANPU (25.32) (Table3).

Hepatosomatic index and liver glycogen

The hepatosomatic index (HSI) also supports this trend. Fish fed the highest level MG had the highest HSI value (2.71), while the fish fed control diet had the lowest HSI values (1.11). Additionally, the liver glycogen content supports these trends. Fish fed control diet and 25% of maize gluten had lower wet weight values of glycogen (34.02 and 37 mg g^{-1} respectively) than fish fed 50% and 75% maize gluten diets (50.83 and 53.07 mg g^{-1} respectively) (Table 3).

Fish body composition

Initial and final carcass compositions based on proximate chemical analysis of the fish fed the experimental diets are presented in Table 4. Total lipid levels increased significantly ($p>0.05$) with higher levels of MG inclusion which ranged from 8.76% in control diet to 13.02% in fish fed diet D4 (75% MG). Moisture, protein and ash levels decreased with increasing MG inclusion. Fish fed diets supplemented with MG showed a peculiar yellow - orange coloration on the skin, operculum and base of the fins, with increasing color intensity as the amount of MG increased in the diet.

Histological studies

Sections of liver tissue showed differences in the liver structure of fish fed diets with higher MG

Table 3. Growth parameters, feed utilisation and hepatosomatic index (HSI) in African catfish fed different diets (mean±SD; n=2)

Parameters	Diets			
	D1	D2	D3	D4
Number of fish	60	60	60	60
Mean initial weight (g)	5.97	5.98	5.95	5.97
Mean final weight (g)	92.70±0.59^{d}	78.34±1.52^{c}	59.59±2.01^{b}	25.47±1.10^{a}
Mean weight gain (g)	86.73±1.60^{d}	72.36±1.53^{c}	53.64±2.01^{b}	19.50±1.03^{a}
Daily feed intake (g fish^{-1} day^{-1})	1.32±0.02^{d}	1.17±0.04^{c}	0.98±0.02^{b}	0.55±0.02^{a}
SGR (%)[1]	4.89±0.01^{d}	4.60±0.04^{c}	4.11±0.06^{b}	2.59±0.07^{a}
FCR[2]	0.85±0.01^{a}	0.91±0.01^{a}	1.02±0.01^{b}	1.58±0.02^{c}
PER[3]	3.33±0.02^{c}	3.09±0.03^{c}	2.76±0.04^{b}	1.81±0.02^{a}
ANPU (%)[4]	51.53±0.98^{c}	47.25±0.55^{c}	40.68±0.61^{b}	25.32±0.19^{a}
Liver glycogen (mg g^{-1} wet weight)	34.02±1.66^{a}	37.00±1.10^{a}	50.83±1.70^{b}	53.07±1.80^{b}
HSI (%)	1.11±0.07^{a}	1.15±0.05^{a}	1.74±0.21^{b}	2.71±0.79^{c}
Survival rate (%)	100	100	100	100

Values in the same row with the same superscript are not significantly different ($p> 0.05$)

[1]SGR: [Ln final bw (g) - Ln initial bw (g)]/feeding days ×100

[2]FCR: feed intake (g)/body weight gain (g)

[3]PER: body weight gain (g)/protein intake (g)

[4]ANPU (%) = (% final body protein × final body weight) - (% initial body protein × initial body weight) / total protein intake (g) × 100

Table 4. Fish body composition (g100 g^{-1} wet weight) of whole fish fed experimental diets (mean ±SD; n=2, 5 fish per duplicate).

Parameters	Initial values	Diets			
		D1	D2	D3	D4
Moisture	77.2	73.18±0.84^{b}	72.76±0.92^{b}	70.93±1.05^{a}	70.81±0.82^{a}
Protein	11.5	15.21±0.57^{c}	14.99±0.41^{c}	14.43±0.38^{b}	13.40±0.40^{a}
Lipid	6.77	8.76±0.81^{a}	9.20±0.64^{a}	12.02±0.80^{b}	13.02±0.63^{c}
Ash	2.07	2.51±0.08^{c}	2.31±0.14^{b}	2.08±0.15^{a}	2.00±0.14^{a}

Values in the same row with the same superscript are not significantly different (p> 0.05).

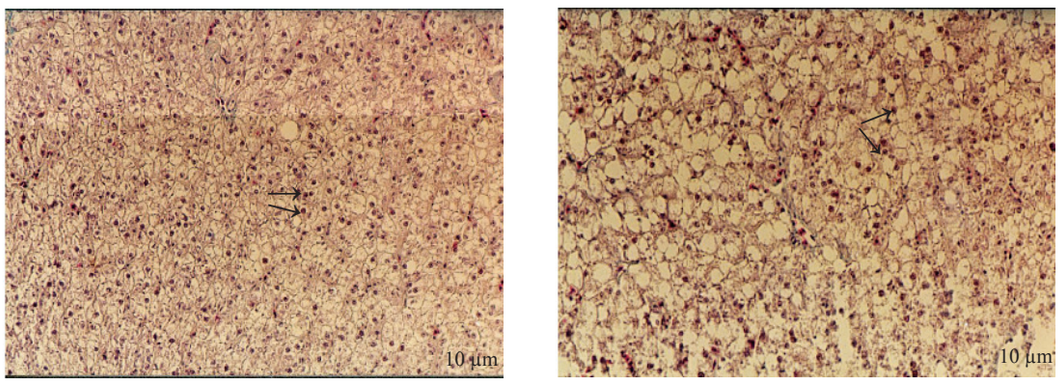

Fig. 1. a: Histological section of liver from *Clarias gariepinus* fed fishmeal-based control diet shows no visible intracellular lipid deposition and normal nuclei (arrow), b : Photomicrograph of histological section of liver from fish fed diet high in MG (75%) shows severe intracellular lipid deposition and abnormal nuclei. (H&E; ×200)

Fig. 1a and b depict typical gross architecture of hepatic tissue from representative fish. Hepatocytes of fish fed control diet were well-defined in shape, well–organised and there were no signs of loss of architecture or necrosis. Catfish fed diets with high inclusion levels of MG (75%) showed appreciable alterations in the liver tissue. The relative size of hepatocytes increased as the proportion of the MG in the diets increased, and this was associated with a much greater hepatic lipid deposition. Polarisation and isolated necrosis in hepatocytes were also observed in fish fed the diet D4.

Discussion

Previous studies have extensively investigated the use of alternative protein sources to substitute fishmeal in fish feeds. The consensus view is that increasing plant protein levels to replace fishmeal has a negative effect on growth rate and feed utilisation. However, a number of researchers have demonstrated the feasibility of using a variety of plant protein at moderate inclusion levels to feed fish. In the present work with African catfish, the results demonstrated that plant protein sources such as maize gluten can effectively replace up to 25% of high quality fish meal protein (LT94) without any serious depression of growth rate. Other studies have also recommended gluten levels of 20-25% (Alexis *et al.,* 1985) and even 40% (Morales, 1993) in diets for rainbow trout. However, Robinson *et al.* (2001) suggested that corn gluten can be used as an effective plant protein source for channel catfish *Ictalurus punctatus* at levels up to 50% without affecting feed palatability, weight gain or feed utilisation, though high replacement levels with plant protein sources radically reduced growth rate in channel catfish. The possible reason for the poor performance associated with the higher substitution of plant proteins than the fishmeal-based control diet is the imbalance of nutrients, particularly protein composition. This may be related to a less adequate dietary amino acid profile when maize gluten is added to the formula because this ingredient is considered deficient in lysine. However, Fasakin *et al.* (2006) found an improvement in the performance indices of African catfish with increasing levels of L-lysine supplementation in maize gluten-based diets.

There are a number of other causes for decreased growth rates and feed consumption observed, at higher levels of MG substitution. The palatability of MG by catfish was certainly lower compared to fishmeal based diets. Our results are in agreement to those obtained from a similar study with yellowtail sea bream, *Seriloa quinqueradiata* by Shimeno *et al.* (1993). These investigators observed that diets containing animal protein sources, such as meat and bone meal, supported higher feed intake than those containing a similar level of corn gluten meal. A second reason for decrease in feed utilisation of MG is the lower digestibility of plant proteins as well

as carbohydrates and the interaction of specific nutrient components during the course of digestion (Davies *et al.,* 2011).

MG meal is a high protein byproduct of the milling industry (Davies *et al.,* 1997) and because of this, MG may be considered a partially purified ingredient. Due to the application of heat treatment and subsequent extraction of the carbohydrates, maize gluten has relatively lower levels of anti-nutritional factors (ANFs) compared to other plant protein sources, such as soybean meal. Furthermore, in relation to the present study, a decrease in growth rate was observed in all fish fed high concentration maize gluten diets when compared with the LT94 fishmeal based control diet. While the ANF content of maize gluten is at least partially responsible for the poor growth performance of tropical fish fed the MG test diets, it should not necessarily be interpreted as a primary reason for the suppression of growth. Fagbenro (1999) reported that the presence of anti-nutritional factors in winged beans did not pose a nutritional problem if the beans were adequately heat treated before inclusion in fish diets.

In the present investigation, the protein efficiency ratio (PER) of the control group was 4.21 compared to the fish fed on MG 75% diet, which was only 2.09. The PER showed a linear trend with the feed conversion ratio (FCR), which increased with each inclusion level of plant protein. The reduction in apparent net protein utilisation (ANPU) supports the reduced protein efficiency with respect to utilisation. This indicated a progressive reduction in food quality, as can be confirmed from the lipid content in the carcass composition. The results of the body composition of fish in this experiment are in accordance with the growth performance results and show an overall reduction in protein and energy retention.

Fish fed the 75% MG diet produced lower body moisture but higher fat content, which is a typical relationship in most fish proximate composition analyses. Instead of utilising the proteins for growth, the proteins may be diverted for energy storage purposes. Storing the unused energy of the plant proteins in the fish body may also increase the fat content. Wu *et al.* (1995) found that when 32 and 36% corn gluten was incorporated into diets fed to Nile tilapia, these diets produced weight gains and feed conversion ratios similar to a control diet of a commercial (36% protein) catfish feed. Therefore, channel catfish can efficiently utilise a 50% corn gluten substitution in the diet without adversely affecting feed palatability, weight gain or FCR (Robinson *et al.,* 2001). Furthermore, Jahanbakhshi *et al.* (2012) reported that the replacement of fish meal with corn gluten meal caused significant differences in growth, SGR, FCR, PER and body compositions of moisture and lipid content, while significant differences were not found in ash and protein composition in common carp, *Cyprinus carpio*. However, African catfish fed a diet containing maize and plantain peel meal showed decrease in weight gain with increased inclusion levels of these ingredients compared with control diet (without plantain peel meal), and PER as well as PPV were also affected (Falaye and Oloruntuyi, 1998). Fagbenro (1999) showed that the African catfish was capable of digesting the energy and protein in winged bean meal as effectively as soybean meal in experimental diets.

A high level of body fat is undesirable from the feed storage point of view because high levels of body fat can increase the rancid off flavour and thus reduce consumer acceptance. In this study, the growth and final lipid content of the fish were affected by the diet composition. Additionally, the final protein deposition in the fish, post-treatment was higher than in untreated fish. Furthermore, there was a trend between protein content in fish fed different amounts of MG, with those receiving the highest amount having the lowest protein content in the whole body.

The protein content values in treated fish only indicate that protein was partitioned throughout the body. It does not; however, provide an indication of how protein is utilised for maintenance. A further reason for poor growth performance and nutrient utilisation is the possible imbalance of fatty acids between animal and plant oil in the different feeds. However, one reason for oil supplementation is that when plant proteins such as maize gluten are employed to replace fish meal on an isonitrogenous basis, oil must be added to the diet to elevate the energy level because plant meals are lower in digestible energy than fish meal. The yellow-orange coloration observed on the operculum, skin and fins of fish fed high levels of MG could be explained by the presence of carotenoids in this meal, mainly zeaxanthin and lutein (Latscha, 1990). Robaina *et al.* (1997) found similar color changes in gilthead sea bream fed diets containing high levels of corn gluten meal. The accumulation of lipids observed in the liver sections of African catfish fed high MG diets could be related to a higher available carbohydrate component in this ingredient. These results agree with those reported by Shimeno *et al.* (1993) who found elevated liver lipid content in yellowtail sea bream, *Seriola quinqueradiata*, fed meat and bone meal compared to fish fed corn gluten meal diets at the same dietary inclusion level. Additionally, our data agree with results obtained by Russel *et al.* (2001) who reported that high inclusion levels of plant ingredients in carnivorous fish (rainbow trout) diets can increase glycogen deposition in the liver after a long feeding period. This increase in gluconeogenesis has

been related to the low carbohydrate digestibility (Hemre *et al.,* 2002). Likewise, Farhangi and Carter (2001) reported increasing glucogenic activity as the inclusion levels of lupin increased in the diet. However, Borquez *et al.* (2011) found that increasing the levels of dietary lupin led to a significant decrease in the hepatosomatic index (HSI) and slight lipid infiltration into hepatocytes in rainbow trout. These observations are in agreement with those of Robaina *et al.* (1995), who found no alterations in lipid and glycogen storage in hepatocytes from *Sparus aurata* fed with a diet supplemented with up to 30% of dehulled lupin seed meal. Moreover, in our work, African catfish fed with high levels of MG supplementation up to 75% resulted in alteration of lipid and glycogen deposition in hepatocytes. This could be attributed to high level of carbohydrate in D4 compared with other two diets D2 and D3. Interestingly, in the present experiment, isolated areas of necrosis in hepatocytes were located in the livers of fish fed higher levels of MG, indicating possible irreversible effects on fish health due to nutritional imbalances (Mosconi-Bac, 1990). Robaina *et al.* (1998) reported that the increase in the n-3/n-6 fatty acid ratio with approximately 30% soybean meal improved the utilisation of liver lipids, thus reducing liver histological alterations in gilthead sea bream. Similarly, the supplementary fish/ vegetable oils remained relatively high in our diets and would have provided the n-3/n-6 fatty acid requirements for this species.

The results of this study was able to show that African catfish responded to diets of varying levels of maize gluten incorporation and grew favourably up to an inclusion level of about 25%, replacing the fishmeal component. However, higher inclusion levels affected growth adversely and also resulted in substantial changes in the hepatic tissue of these fish.

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