FEEDING AND WASTE MANAGEMENT STRATEGIES IN THE CULTURE OF *Clarias gariepinus*

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ABSTRACT

Nutrient digestibility and amino acid availability were assessed in sharp-toothed catfish. *Clarias gariepinus*, fingerlings fed diets containing soyabean flour (SF) - Poultry meat meal (PMM) blends (25:75, 50:50, and 75:25) and 0.5 of 1.0 % Cr_2O_3. There was agreement between the pattern of overall protein digestibility and average amino acid availability despite the variability in individual amino acid availability,. The best dry matter, lipid and protein digestibility coefficients, and amino acid availability values were obtained with diets containing 0.5% Cr_2O_3. Chromic Oxide inclusion level appeared to affect nutrient availability. Increased marker level resulted into decreased nutrient digestibility coefficients. Similarly, these diets generated lower fecal crude protein than those with 1.0% Cr_2O_3. However, the latter group recorded higher protein retention efficiency. Dry matter and lipid of diets containing more soyabean flour seemed to be more digestible than those of poultry meat meal. Similar trend was observed for the apparent availability of the amino acids. This investigation has indicated that low level of marker was better in digestibility study. Utilisation of more SF than PMM in the diets of this catfish was more beneficial and should be encouraged in the feed industries producing catfish diets towards a better feed and waste management strategies in this aquaculture operation.

INTRODUCTION

Nutritional approaches to quantifying solid and soluble wastes from aquacultural operations have log been proposed. These are based on the importance of digestibility as an indicator of potentially available energy and nutrients for growth, maintenance and reproduction of the animal, as well as the levels of indigestible nutrients (solid wastes output) which account for the major portion of aquaculture wastes (Cho 1993). Cho *et a;* (1991) investigated effect of feed quality on effluent waste concentrations from diets fed to brown trout, *Salmo trutta,* using both biological (nutritional) and chemical methods. This was designed to verify the prediction model for assessing aquaculture waste output from feed quality, quantity and fish produced.

Digestibility estimations have been a major focus in aquaculture nutrition for assessing ingredient or diet quality. However, controversy exists concerning the use of internal or external markers (Bowen 1978; De Silva and Perera 1983; Tacon and Rodrigues 1984). Higher digestibility values were recorded with the use of a internal marker (crude fiber) than an external one, chromic oxide (Cr_2O_3) in blue tilapia, *Oreochromis aureus* (De Silva *et al.* 1990). Little is known with respect to digestibility and the use of chromic Oxide in the sharp-toothed catfish. *Clarias gariepinus* despite its aquaculture potential. *Clarias gariepinus* is a member of the clariid family and a warmwater fish. It is presently of commercial importance in most tropical fish culturing countries in Africa and Asia where it is a major culturable species (FAO 1994). In temperate countries, European countries in particular, it is being cultured in tropicalised aquarium with appreciable growth response.

The purpose of the present study was to evaluate the effect of chromic oxide on digestibility values in sharptoothed catfish fed diets containing soyabean flour and poultry meat meal blends. Fecal nitrogen output and protein efficiency would also be evaluated.

MATERIALS AND METHODS

Experimental Design

A 2 x 3 factorial design was used and involved 12 10-L aquaria. Each treatment was duplicated. Sharp-toothed catfish fingerlings (23.3 ± 0-6g) were stocked at five fish per aquarium in a static-water digestibility system. The system was supplied with water from an overhead tank thermoregulated at 28°C.

Diets, Feeding and Fish Husbandry

Six isocaloric (4 kcal/g gross energy) and isonitrogenous (38% crude protein) diets (Diets I-VI) were formulated on the basis of two factors: Chromic oxide level (0.5% and 1.0%) and soybean flour (SF): poultry meat meal (PMM) blends (25:75.50:50) and 75:25) (Table 1). Ingredients were mixed thoroughly and pelleted using a pellet mill (California Pellet Mill Model CL2, San Francisco, California. USA). Pelleted diets were dried by convection at 60°C overnight. After cooling, diets were packed in sealed black polyethene bags and stored for three weeks to collect sufficient fecal matter for analysis. Feed was given at 12.00 hours at 2% body weight. The fishes were allowed to feed for one hour after which feed remnants were flushed out. Fecal matter was collected at 11.00 hours the following morning, weighed and oven-dried overnight at 105°C prior to proximate and Cr_2O_3 analyses. This was done by draining from the aquarium base once the connected valve was open as done with the Guelph System (Cho *et al*, 1985).

Chemical Analysis.

Ingredients, diets and fecal matter were analyzed for proximate composition in duplicate according to ADAC (1990) methods. Protein was determined using micro-Kjeldahl technique (Kjeltec 1030. Iecator), lipid was determined by asking in a muffle furnace for 12 hours at 450°C. Anubi acids were analysed using Alpha-plus Amino Acid Analyzer (LKB Bithrom Ltd, Cambridge, United Kingdom). Energy values were calculated using gross energy values of 9.5, 5.4 and 4.1 kcal/g for lipid, protein and carbohydrate respectively (Jobling 1983). Chromic Oxide was estimated by the method of perchioric acid. Thereafter, the percentage chromic oxide was determined spectrophotometrically. From the Cr_2O_3 values of the diets and feces, apparent digestibility coefficients (ADC) and apparent amino acid availability values of the diets and feces, apparent digestibility coefficients (ADC) and apparent amino acid availability (AAAA) of the diets were determined according to Maynard et al. (1979) from the equations below:

$$ADC\ (\%) = 100\% \frac{Cr_2O_3 \text{ in diet}}{\%\ Cr_2O_3 \text{ in feces}} \times \frac{\%\text{nutrient in faeces}}{\%\text{nutrient in diet}} x$$

$$AAAA\ (\%) = 100 - (100\ \frac{\% Cr_2O_3 \text{ in diet}}{\% Cr_2O_3 \text{ in feces}} \times \frac{\%\text{amino acid in faeces}}{\%\text{amino acid in diet}}$$

The equation for fecal protein waste output (indigestible crude protein) estimation was:

(100-%protein ADC) x protein intake (g/fish/dau) = Indigestible protein (g/fish/day)

The equation for protein retention efficiency (PRE) was:

$$PRE = \frac{\text{carcass protein gain (g)}}{[\text{Protein intake (g)} \times \text{protein ADC (\%)}]}$$

Statistical Analysis

Data were analyzed using one way analysis of variance (ANOVA) for the treatments variation. Turkey's test (Steel and Torie 1968) was used for means comparison. Multifactor analysis of variance (MANOVA) was used to evaluate variation due to the factors (blending ratio and Cr_2O_3 level) and their interaction.. Probability level was 0.05 and used for the significance test of the variation. Percentage data was transformed by arc-sin transformation on according to Zar (1984). Statistical software Statgraphic (version 5.0) was used for this analysis.

Table 1: Level of inclusion of ingredients in the soybean flour-poultry meat meal blend based diets of *Clarias gariepinus* and the proximate composition of the diets. Values in parentheses are the percentages of Cr_2O_3 in the diet.

	25	75	50	50	75	25
Ingredients	I (0.5)	II (1.0)	III (0.5)	IV (1.0)	V (0.5)	VI (1.0)
Soybean flour	48.8	14.8	32.5	32.5	54.0	54.0
Poultry meat meal	44.5	44.5	32.5	32.5	18.0	18.0
Wheat flour	27.4	27.4	20.8	20.8	12.8	12.8
Soybean oil	5.2	5.2	6.2	6.2	7.3	7.3
Vitamin premix[1]	2.0	2.0	2.0	2.0	2.0	2.0
Mineral premix[2]	4.0	4.0	4.0	4.0	4.0	1.0
MC(Binder)[2]	1.5	1.0	1.5	1.0	1.5	1.0
Cr_2O_3 (Marker)	0.5	1.0	0.5	1.0	0.5	
Proximate Composition (% as fed)						
Moisture	5.2	5.8	6.1	7.5	7.4	10.2
Protein	39.1	39.4	39.2	38.5	38.8	38.3
Lipid	10.7	10.9	10.4	10.2	10.0	9.6
Ash	12.0	11.8	10.9	10.9	9.9	9.7
Energy kcal/g	4.5	4.5	4.5	4.4	4.4	4.3

[1]Vitamin premix providing the following vitamins (mg/kg premix): Vitamin A, 1000; Vitamin D, 4.0; Vitamin E, 7000; Vitamin K, 1500; Vitamin C, 37500; thiamine, 4250; riboflavine, 3000; pyridoxine, 1250; pantothenic acid, 5250; Niacin, 12500; biotin, 90; folic acid, 1000; Vitamin B_{12}, 1.25; choline chloride, 74050; inositol, 25000.

[2]Mineral premix providing the following minerals (g/kg premix): calcium orthophosphate, 727.8; magnesium sulphate, 127.5; sodium chloride, 60; potassium chlorine, 50; iron sulphate, 25; zinc sulphate, 5.5; manganese sulphate, 2.5; copper sulphate, 0.8; cobalt sulphate, 0.5; calcium iodate, 0.3; chromic chloride, 0.1

[3]CMC = Carboxymethyl cellulose

RESULT AND DISCUSSION.

Nutrient digestibility

Table 2 represents the dry matter, protein, lipid and ash apparent digestibility coefficients (ADCs) of the six diets. Lipid digestion was highest of the three nutrients. Higher nutrient ADCs were obtained for diets containing 0.5% Cr_2O_3 than 1.0% Cr_2O_3 except for the protein ADC of diets V and VI where those of VI (1.0% Cr_2O_3) was significantly higher than diet V (0.5% Cr_2O_3) (P <2 0.05). Only lipid and dry matter ADCs varied significantly with the blends. The interaction of the blending ratio and Cr_2O_3 level were insignificant for dry matter and protein ($P > 0.05$) but significant for ash and lipid ($P < 0.05$).

Dry matter ADV was higher in 75:25 SF:PMM or 0.5% Cr_2O_3 while that of protein was highest at 75 SF:PMM and 0.5% marker level while that of ash was highest at 25:75 SF:PMM and 0.5% marker level except for diets V and VI. The high lipid ADCs for all the diets may be attributed to high lipase activity in fish generally (Sargent *et al.* 1989; De Silva and Anderson 1995). Sadiku and Jauncey (1995a) observed a high lipid digestibility in *Oreochromis niloticus*. Dry matter, lipid and protein ADCs were moderately high and low for diets containing 0.5 and 1.0% Cr_2O_3, respectively, in agreement with Sadiku and Jauncey (1995a) who found that increasing Cr_2O_3 level in a diet could surpress digestibility. It was also observed that protein of diets with more SF were more digestible than those with more PMM in *Oreochromis niloticus*. This is in agreement with earlier reports that protein of dehulled solvent extracted soyabean meal was very digestible in fish - 85% digestible in channel catfish (Lovell 1977), rainbow trout (Smith 1976) and tilapia (Popma 1982), and even equally or more digestible than that of whole fish meal protein in channel catfish (Lovell 1990). Protein of 75:25 SF:PMM blend based diet was therefore expected to be more digestible than 25:75 and 50:50 SF:PMM. This was not the case in *Clarias gariepinus* which tends towards carnivory could be responsible. This is not conclusive as it will need to be further investigated.

Low ash ADCs relative to those of protein and lipid were observed here. This might be due to poor enzymatic digestion of ash, as digestion of inorganic matter is largely due to stomach acid (Lovell 1990). Very low ash digestibility values have been reported for soyabean meal and poultry by-products in rainbow trout (Alexis *et al.* 1988). Effect of Cr_2O_3 level on ash ADC pattern was in agreement with that of protein and lipid as diets containing 0.5% Cr_2O_3 better digestible than those of 1.0% except for diets V and VI.

Nutrient availability.

Table 3 shows the availability of 17 amino acids analysed. The overall pattern of apparent amino acid availability corresponds to that of protein digestibility. There was no significant difference in amino acid availability between the blending ratios of SF:PMM in the diets ($P > 0.05$). Marker level was a significant factor in the amino acid availability. Overall apparent amino acid availability was significantly higher in diets containing 0.5% than 1.0% Cr_2O_3 ($P < 0.05$). The interactions of blend and marker levels were insignificant ($P > 0.05$). Methionine and the highest availability while the least available was cysteine. Wilson *et al.* (1981) investigated the availability of amino acids in soyabean meal for channel catfish and found that arginine was most available while the least available was glycine. However, they presented the availability of methionine + cysteine in combination which have been separated in this investigation.

Prediction of faecal nitrogen waste output

Cho (1993) postulated that it is possible to determine the proportions of nutrients retained in fish and excreted as solid wastes from the apparent digestibility of feed and comparative carcass analysis coupled with measurements of feed intake and growth. (This would be helpful in developing prediction model for waste estimation in aquaculture operation. Biological (nutritional) and chemical methods of quantifying waste output in effluent have being in use. The biological method seems more reliable. *Cho et al.* (1991) reported that there was agreement in results from both methods for nitrogen waste output in brown trout culture. However, the chemical method was inappropriate for phosphorus waste output as it gave excessively high phosphorus output level. Similarly they observed that the use of fixed ADC and NRE (nutrient retention efficiency) for

0.5% Cr_20_3 while those without had 1.0% Cr_20_3.

Diets	Dry matter	Protein	Lipid	Ash
I (25:75)%	87.39±3.57ab	86.74±0.47b	87.76±0.83b	36.97±0.28b
II (25:75)	76.70±0.00a	62.44±2.65a	69.03±1.48a	20.46±6.58c
III (50:50)%	89.96±0.00b	88.06±0.57b	95.61±0.77b	36.38±2.37a
IV (50:50)%	83.61±4.74ab	73.69±6.75a	88.95±0.21b	21.40±0.95b
V (75:25)	91.10±4.19b	87.87±0.88b	96.10±0.98b	24.20±8.32a
VI (75:25)	88.17±0.00ab	70.25±1.29a	93.36±0.28b	27.42±1.58a

Table 3: Amino acid availability of soybean flour-poultry meat meal blend based its (Diets I-VI) fed to Clarias gariepinus fingerlings. M±SEM (Mean ± Pooled Standard Error of Mean). Data on the same row carrying the same letter are insignificantly different (P > 0.05).

Diets

EAA%	I(25:75 SF:PMM; 0.5%Cr_20_3)	II(25:75 SF:PMM; 1.0%Cr_20_3;	III(50:50 SF:PMM; 0.5%Cr_20_3)	IV(50:50 SF:PMM; 0.05%Cr_20_3)	V(75:25 SF:PMM; 0.05%Cr_20_3)	VI(75:25 SF:PMM: 1.0%(Cr_20_3)
Arg	86.28±0.65cd	45.89±5.76a	86.99±0.25cd	62.87±1.73b	90.65±0.46d	82.12±0.05c
His	89.12±0.49c	60.98±1.08a	89.06±0.30c	70.05±0.16b	91.25±0.49c	69.85±0.21b
Iso	85.42±0.56c	44.59±5.27a	85.22±0.64c	63.94±0.15b	89.71±0.40c	64.73±1.36b
Leu	87.04±0.93c	55.69±3.71a	86.67±0.42c	67.32±0.77b	89.04±0.33c	67.60±0.081b
Lys	89.97±0.44c	65.56±0.34a	89.31±0.34c	71.23±0.23b	91.56±0.38c	71.74±0.45b
Met	92.62±0.44d	72.14±2.36a	93.15±0.08d	82.26±0.18b	93.90±0.03d	78.56±0.25b
Phe	87.23±0.74c	57.89±0.59c	86.79±0.59c	66.83±1.16b	91.75±2.06c	68.22±0.43b
Thr	85.83±1.30c	52.34±4.02a	85.50±0.55c	6.92±0.00b	89.70±0.25c	67.71±1.74b
Val	85.29±0.89c	45.57±5.34a	84.49±0.59c	64.47±3.68b	89.12±0.56c	64.27±0.83b
Mon-EAA%						
Ala	89.04±40.40b	60.94±3.39a		67.31±1.02a	89.29±0.76b	67.28±5.80a
Asp	87.54±0.34c	54.0±3.24a	87.47±0.55b	67.59±1.29b	90.01±0.13c	69.94±1.58b
Cys	83.92±0.53d	44.12±2.01a	87.89±0.42c	67.45±1.63b	90.94±0.09e	67.22±2.68c
Glu	90.03±0.78b	61.79±0.05a	87.44±1.07d	71.20±1.03a	92.35±0.18b	72.50±0.3a
Gly	89.35±0.73c	69.77±4.03a	89.67±0.32b	69.42±2.00ab	90.13±0.34c	71.93±1.2b
Pro	88.32±0.77c	60.94±0.14c	88.04±0.54c	66.64±.1.51b	90.66±0.23c	71.62±0.00b
Ser	86.28±0.72c	85.90±0.72c	88.85±0.14c	66.76±0.93b	90.02±0.38c	68.71±1.15b
Tyr	89.00±0.39d	61.65±0.40d	85.90±0.72c	70.89±1.24b	92.17±0.20e	73.01±0.41c
			89.70±0.40d			
M:SEM	87.94±2.00b	58.62±7.50a	88.00±1.14cd	68.41±.84a	90.82±1.05d	73.01±2.33bc

*Arg, Arginine: His, Histidine; Iso, Isoleucine; Leu, Leucine; Lys, Lysine; Met, Methionine; Phe, phenylanine; Thr, Threonine: Val, Valine: Ala, Alanine: Asp, Aspartic acid: Cys, Cystine: Glu, Glutamic acid: Gly, Glycine; Pro, Proline; Ser, Serine: Tyr, Tyrosine:

Figures in the same row with the same letter are insignificantly different
(P >0.05).

Estimating waste output was equally misleading as different ingredients and diet formulations would continue to change. Digestibility and nutrient utilization studies of either an ingredient or a diet formulation are therefore necessary preambles to predicting its waste generation potential and subsequent modelling.

In this study, the nutrient digestibilities of the diets with 0.5% and 1.0% levels of Cr_2O_3 were investigated (Table 2). Protein digestibility of the diets differed significantly from each other ($P < 0.05$). Table 4 depicts that diet III (50:50 SF: PMM) had the highest protein ADC, followed by diets V(75:25 SF:PMM) and I (25:75 SF:PMM) in that order - all with 0.05% Cr_2O_3. They generated lower fecal nitrogen wastes than diets II, IV and VI with 1.0% Cr_2O_3. This was probably due to their high level of the marker. Protein retention efficiency (PRE) derived from carcass protein gain, ADC and protein intake data depicts that only marker level had significant effect on PRE ($P<0.05$). Effect of blend variation was insignificant as well as its interaction with the marker levels ($P>0.05$).

Carcasses protein gain, protein intake and apparent net protein utilisation (ANPU) data of Sadiku and Jauncey (1995b) from a previous feeding trial using the same diets were used in predicting the PRE. It is noteworthy that the feeding trial diets had no Cr_2O_3. This was due to the higher protein ADC in the latter while they shared the same carcass protein and protein intake. Sadiku and Jauncey (1995) recorded insignificantly different PRE with the two marker levels and same diets in *Oreochromis niloticus* diets.

The use of the same carcass protein and protein intake values for the two marker levels in evaluating PRE would not hold in practice if the feeding trial diets were to contain the two levels of the marker. Protein intake in diets with 0.5% Cr_2O_3 would be higher than those with 1.0% Cr_2O_3, as well as its carcass level, since it was more digestible. Consequently, insignificantly different PRE would probably have been recorded in diets with 0.5% Cr_2O_3 and 1.0%, if not higher for the former as theoretically expected. This hypothesis will need to be investigated. Therefore, it is suggestive that feeding trial diets should equally contain the marker for accurate estimation of waste generation potential and nutrient retention efficiency of diets in aquaculture operation.

Table 4: Daily individual protein intake, digestion, carcass protein and protein retention in *Clarias gariepinus* fingerlings fed soyabean flour-Poultry meat meal blend based diets. Figures in parenthesis represent the blending ratios of soyabean flour. Poultry meat meal. Imported data from previous 56 days feeding trial of Sadiku and Jauncey (1995) as a case study for predicting waste generation potential and nutritient retention of the same diets in *C. gariepinus*. Calculated values predicting indigestible protein and PRE.

Diets	Diet ration(Protein/ first/day)	Protein ADC (%)	Indigestible Protein (g/fish/day**	Carcass Protein (g/fish/day)	ANPU (%)	Protein Retention Efficiency
I (25:75)%	0.39±0.05a	86.76±0.34c	0.06±0.01a	0.18±0.00a	40.00±2.0a	0.46±0.03ab
II (25:75)	0.39±0.05a	62.44±1.88a	0.15±0.02b	0.18±0.00a	440.00±2.0a	0.64±0.05b
III (50:50)%	0.47±0.04b	88.06±0.40c	0.06±0.01a	0.23±0.02b	47.50±0.5a	0.55±0.01ab
IV (50:50)%	0.47±0.04b	73.69±4.78b	0.13±0.01b	0.23±0.02b	47.50±0.5a	0.65±0.00b
V (75:25)	0.38±0.04a	87.87±0.62c	0.05±0.01a	0.19±0.00a	47.00±0.5a	0.40±0.01a
VI (75:25)	0.38±0.04a	70.25±0.91b	0.11±0.01b	0.19±0.00a	40.00±6.5a	0.58±0.09ab

Figures in the same column with the same letter are insignificantly different ($P > 0.05$).

Finally, the effect of digestibility studies marker level needs be established. It appears from this study that increasing the levels of Cr_2O_3 from 0.5 to 1.0% apparently depressed protein digestion and amino acid availability. Whether, this would affect diet intake and carcass composition will need to be studied. This maker level differential would obviously affect the value of faecal nitrogen waste output from these ingredients and diets. Estimation of waste output of different ingredients, feed combinations and nutrients other than those investigated here would be beneficial to expanding the scope of this investigation and developing comprehensive nutritional model for quantifying waste output generation in aquaculture operation.

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