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Preparation, nutrient composition and digestibility of fermented shrimp head silage

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Raw heads of the river prawn, *Macrobrachium vollenhovenii*, were fermented with *Lactobacillus plantarum* at 30°C using molasses or cassava starch as the carbohydrate source. After incubation for 7 days, a desirable and stable pH < 4.5 was attained, and the carbohydrate source did not affect non-protein nitrogen (NPN) content or the proximate composition of the liquid silage after 30 days incubation. Hydrolysed feather meal, poultry by-product meal or soybean meal, used as alternative filler, was blended with the liquid silage (85:15, w/w) and solar-dried. The dried shrimp head silage meals were incorporated as protein supplements into pelleted semi-purified diets for catfish, *Clarias gariepinus*. Apparent digestibility coefficients of dry matter, crude protein, gross energy and essential amino acids in the silage by catfish fingerlings was high (> 70%). It was concluded that dried fermented fish silage is suitable and has a potential as a protein feedstuff in fish diets. © 1997 Elsevier Science Ltd

INTRODUCTION

Macrobrachium vollenhovenii (Herklots 1857) is the dominant shrimp species in the commercial inland fisheries in Nigeria (Bello-Olusoji *et al.*, 1995), and vast quantities of by-products (head, hull) are generated in the shrimp peeling/processing industry. The heads comprise > 33% of the whole raw shrimp production and are discarded as waste (Balogun & Akegbejo-Samsons, 1992) and only small volumes are converted to shrimp head meal for use in animal feeding. Considerable product variability may occur depending on the species of shrimp used and whether the waste comprises heads alone, hulls or total wastes recovered from peeling operations (Cruz-Suarez *et al.*, 1993). Sun-drying or cooking followed by coarse milling are the most common techniques for shrimp head meal production. However, there are limitations—cooking requires excessive use of firewood or other scarce fuels while sun-drying is frequently carried out under unhygienic conditions leading to meals with high microbial loading (Barratt & Montano, 1986; Hall & De Silva, 1994). Shrimp meal production requires fresh raw materials in view of the active proteolytic enzyme and microbial degradative processes that occur, which result in poor quality meals with significant protein losses (> 10%) and short storage life (Meyers, 1986). Prompt preservation through lactic acid fermentation represents an artisanal and cheap technique which will stabilize and retain the nutritional quality, and enhance the exploitation of this resource for aquaculture feed. The resulting liquid silage could be blended with fillers (dry protein feedstuffs) to aid drying but the choice is determined by cost and local availability (Lopez, 1990). This study reports the proximate composition and amino acid composition of lactic acid-fermented shrimp head silage co-dried with three alternative fillers (hydrolysed feather meal, poultry by-product meal, soybean meal). The apparent digestibilities of nutrients (protein, energy, essential amino acids) in the lactic acid-fermented shrimp head silage meals by the African clariid catfish (*Clarias gariepinus*) fingerlings were determined and compared with those of solar-dried shrimp head meal used as reference.

MATERIALS AND METHODS

Preparation and characterization of fermented shrimp head silage

Raw heads of *M. vollenhovenii* discarded as waste from small-scale processors were minced and distributed into two 1-kg batches (A, B). To batch A was added 150 g of molasses and 50 ml of inoculum while 150 g of cassava starch and 50 ml of inoculum was added to batch B. Both mixtures were stirred and incubated in sealed containers at 30°C for 30 days, after which they were

heated to 90°C in a temperature-controlled water bath and maintained for 30 min to halt autolysis, and centrifuged. The silages were swirled daily and triplicate 30 g samples were taken after 2, 4, 7, 15 and 30 days for analyses. Later, a 5 kg of batch B silage was prepared and blended with soybean meal, poultry by-product meal, or hydrolysed feather meal (85:15, w/w) and dried for 24 h under a solar simulator (1 kW/m^2) to obtain a moisture level below 10%. As a reference, 5 kg of solar-dried shrimp head meal was prepared by solar-drying the raw shrimp heads.

The pH of the liquid silage samples was determined (Table 1) and the non-protein nitrogen (NPN) content was analysed by the trichloroacetic acid (TCA) precipitation technique (Backhoff, 1976). Moisture content was determined by oven-drying at 105°C for 24 h; lipid content by extracting the residue with 40–60°C petroleum ether for 8 h; ash content by ignition at 550°C for 24 h; total N content by the Kjeldahl method and crude protein estimated as N×6.25 (AOAC, 1990). Protein content was corrected for the N content of chitin determined by gravimetry (Black & Schwartz, 1950). Amino acid composition was determined after treating the hydrolysate with 6 mol l^{-1} HCl under reflux for 24 h at 110°C using an Nc-2P TECHNICON amino acid auto analyzer. Tryptophan was determined colorimetrically (Fischl, 1960). Free fatty acid content was determined after extraction with methanol/chloroform mixture (Pearson, 1971). Gross energy was determined by bomb calorimetry.

Digestibility of nutrients in fermented shrimp head silage meals

Four diets were formulated to contain 70 g/100 g dry weight of fermented shrimp head silage meals or solar-dried shrimp head meal and 30 g/100 g dry weight of a purified reference diet (Table 2). Chromic oxide was incorporated as the inert digestibility marker. Dry extruded diets (5-mm diameter, 1-cm long) were prepared and stored (−20°C) in air-tight polythene bags until fed. Groups of 15 *C. gariepinus* fingerlings (68–73 g), obtained by artificial reproduction, were distributed in into 120-l rectangular indoor glass aquaria (75 cm×40 cm×40 cm) supplied with fresh aerated water (flow, 1 l/min; pH, 6.6–7.5; temperature, 28°C; dissolved oxygen, 6–8 mg/l). Each diet was assigned to triplicate aquaria. Catfish were fed to satiation twice daily (8.30–9.00 h and 16.00–16.30 h) for 14 days. On the last day, faeces were collected from each catfish 8 h after feeding by the rectal dissection method (Henken *et al.*, 1985) from the terminal 2.5 cm of the intestine after anaesthetizing catfish in quinaldine (2.5 ml), and pooled for each treatment. Dry matter and crude protein were analysed in triplicate samples of feed and faeces according to AOAC (1990) methods. Gross energy was determined by bomb calorimetry. Chromic oxide content of diets and faeces was determined (Furukawa & Tsukahara, 1966). Apparent digestibility coefficients (ADC) of dry matter, protein, energy and amino acids in the test diets were estimated thus:

$$ADC_{nutrient} = 10^2 - [10^2 \times (l_d/l_f \times N_f/N_d)]$$

where N_d = nutrient in diet; N_f = nutrient in faeces; l_d = Cr_2O_3 in diet; l_f = Cr_2O_3 in faeces.

The $ADC_{nutrient}$ in test feedstuff was calculated as follows:

$$ADC_{nutrient} = 100/30(ADC_{test\ diet} - 70/100 ADC_{referencediet})$$

ADC values were statistically analysed by the analysis of variance (ANOVA) test and Duncan's multiple range test was used to compare ADC values between meals ($P < 0.05$) (Zar, 1984).

Table 1. pH of fermented shrimp head silages prepared using cassava starch or cane molasses as carbohydrate source

Carbohydrate source	Fermentation period (days)					
	0	2	4	7	15	30
Cassava starch	6.7	4.8	4.7	4.5	4.3	4.0
Cane molasses	6.7	4.6	4.4	4.3	4.1	4.0

Table 2. Formulation of reference diet

	g/100 g
Casein	40
Gelatin	10
Alpha starch	40
Mineral mixture[a]	6
Vitamin mixture[b]	3
Chromic oxide	1

[a,b]According to Haylor (1992).

Table 3. pH, NPN and proximate composition (g 100 g^{-1} DM) of minced shrimp heads and fermented shrimp head silages prepared using cassava starch or molasses as carbohydrate source (after 30 days)

	Minced shrimp heads	Fermented shrimp head silage	
		Cassava starch	Cane molasses
pH	6.4	4.0	4.0
NPN (g. 100 g^{-1} TKN)	15.2	50.6	50.6
Dry matter (DM)	69.2±3.6	35.3±4.1	33.1±3.2
Crude protein	50.6±2.5	41.8±1.8	42.7±2.1
Crude lipid	18.1±1.7	15.0±1.3	13.6±1.4
Total ash	25.3±1.9	12.1±1.2	13.7±1.5
Tryptophan (mg.g^{-1} protein)	17.3	16.3	15.4
Free fatty acid (% oleic acid)	0.92	1.13	1.65

Table 4. Proximate (g/100 g dry matter) and essential amino acid (EAA)(g/100 g protein) composition of solar-dried shrimp head meal and fermented shrimp head silage meals

	SHM[a]	FSS:HF-M[b]	FSS:PB-M[c]	FSS:SB-M[d]
Dry matter[e]	91.2	91.6	92.4	90.1
Crude protein	51.3	65.6	52.1	44.7
Crude lipid	9.2	8.3	14.8	9.8
Crude fibre/chitin	12.6	6.8	7.5	9.6
Total ash	23.9	8.7	15.2	10.3
Gross energy	3.91	4.58	4.28	4.14
Arginine	6.2	6.4	6.3	6.7
Histidine	2.4	2.0	2.3	2.7
Isoleucine	2.9	4.9	3.5	2.9
Leucine	6.5	7.3	7.1	6.9
Lysine	6.3	6.1	8.1	6.3
Methionine	1.7	2.0	1.9	1.8
Phenylalanine	4.3	3.8	4.2	4.7
Threonine	4.1	6.3	6.0	4.7
Tryptophan	0.6	1.1	1.0	0.8
Valine	4.3	4.7	4.8	4.3

[a]Solar-dried shrimp head meal.
[b]Fermented shrimp head silage:hydrolysed feather meal blend.
[c]Fermented shrimp head silage:poultry by-productmeal blend.
[d]Fermented shrimp head silage:soybean meal blend.
[e](kcal/g DM).

RESULTS AND DISCUSSION

Effect of carbohydrate source on fermentation

After incubation for 7 days, a desirable and stable pH < 4.5 was attained. Molasses gave a more rapid pH decline than cassava starch (Table 3). The slow initial pH decline in the cassava treatment could be due to a high proportion of unhydrolysed starch in cassava, as suggested by Van Wyk & Heydenrych (1985). Hence, addition of an amylolytic enzyme source may be necessary so that the starch can be hydrolysed to sugars, suitable carbohydrate substrates for fermentation. Fermented shrimp heads were incubated for 30 days without deterioration in nutritional characteristics and the carbohydrate source did not affect NPN content (Table 3). The low NPN content in fermented shrimp head silage (45%) was similarly reported for snipefish (*Macrorhamphosus* spp.) fermented for 32 days (Batista *et al.*, 1989) and was attributed to the adsorption of enzymes and proteins by carbohydrates which prevented their interaction. However, Lindgren & Pleje (1983) reported higher NPN values (> 50%) in Baltic herring (*Clupea harengus*) over a similar period of fermentation. The differences are probably due to variations in enzymatic activity between species. Free fatty acid content doubled after fermentation for 30 days (Table 3) as a result of lipolysis. A sharp fall (5.8%) occurred in the tryptophan content within 7 days of fermentation and a further decrease (11.0%) after 30 days (Table 3). Tryptophan is labile under acid conditions and, generally, loss of amino acids in fermented silage has been attributed to their interaction with sugars in the unutilized molasses (Kompiang *et al.*, 1980). Both carbohydrates tested as fermentation substrates were suitable but, considering the need to minimize costs in developing countries, the use of cane molasses is particularly appropriate for tropical regions where it is frequently available as an industrial by-product. Moreover, cassava starch has competitive uses in human diets and may prove uneconomical. In addition, Dhatemwa (1989) warned of a risk of cyanide poisoning if the proper variety of cassava was not used.

Nutritional composition and digestibility of fermented shrimp head silage

Preparing artificial feeds using shrimp processing wastes (containing large amounts of chitin and ash) can lead to weakening of the pellets (Meyers, 1981). The fermentation process reduced the ash and chitin content of shrimp heads (Table 4) which may improve pellet stability in water, hence reducing the likelihood of depriving the cultivated aquatic animal species of essential nutrients. A similar phenomenon of lower chitin and ash contents was reported by Meyers & Benjamin (1987) and Fox *et al.* (1994) after formic acid ensilation of shrimp heads. Nutrient composition of the dried fermented shrimp head silage meals (Table 4) shows similar gross energy content and amino acid profiles that are comparable to those of the solar-dried shrimp head meal. Protein, lipid, fibre and ash contents were different, and reflected the corresponding constituents of the fillers.

Table 5. Apparent digestibility coefficient (%) of solar-dried shrimp head meal and fermented shrimp head silage meals

Meals	DM[e]	Protein	Energy	ARG	HIS	ILE	LEU	LYS	MET	PHE	THR	TRY	VAL
SHM[a]	68.8b	76.2c	59.9b	84.7b	78.6b	84.5b	84.8b	88.5ab	88.6a	78.7b	87.5ab	79.0a	82.2b
FSS:HFM[b]	79.5a	81.4bc	82.2a	80.0b	74.5b	78.8b	80.8b	79.7c	90.5a	75.0b	81.6b	73.6a	81.9b
FSS:PBM[c]	75.7a	82.4b	86.4a	82.3b	79.8b	82.2b	83.3b	85.7bc	91.3a	79.7b	83.3b	78.3a	80.8b
FSS:SBM[d]	77.3a	90.8a	84.9a	94.3a	91.1a	92.2a	90.3a	93.7a	87.6a	91.4a	90.8a	78.6a	90.3a

[a]Solar-dried shrimp head meal.
[b]Fermented shrimp head silage: hydrolysed feather meal blend.
[c]Fermented shrimp head silage: poultry by-product meal blend.
[d]Fermented shrimp head silage: soybean meal blend.
[e]Dry matter.
a,b,c, mean values in the same column with different letters are significantly different (P < 0.05).

$ADC_{dry\ matter}$ values of the shrimp head silage meals were higher ($P < 0.05$) than that of solar-dried shrimp head meal (Table 5) which in turn was lower than that reported for solar-dried shrimp (*Penaeus notialis*) head meal by the catfish *Clarias isheriensis* (Fagbenro, 1996). This may be due to differences in species and size of catfish, species of shrimp (head) used and whether the waste comprises heads alone, hulls or total wastes recovered from peeling operations as suggested by Cruz-Suarez *et al.* (1993). $ADC_{protein}$ values followed a similar trend as $ADC_{dry\ matter}$ (Table 5) and are similar to $ADC_{protein}$ of fermented fish silage meals reported for other catfishes—*C. gariepinus* (Fagbenro & Jauncey, 1995); *C. batrachus* (Wee *et al.*, 1986); *C. macrocephalus* (Edwards *et al.*, 1987). Of the shrimp head silage meals tested in this study, FSS:SBM had the highest $ADC_{protein}$ and these are probably related to ingredient (filler) quality. The solar-dried shrimp head meal had the highest crude fibre/chitin (12.6%, Table 4) and the amine fraction in the chitin would lower the estimated $ADC_{protein}$ (Akiyama *et al.*, 1989) but should have little influence on the ADC_{EAA} (Clark *et al.*, 1993). The ADC_{energy} value for solar-dried shrimp head meal was lower ($P < 0.05$) than those of the shrimp head silage meals (Table 5) but is comparable to that reported for solar-dried shrimp (*P. notialis*) head meal by *C. isheriensis* (Fagbenro, 1996). However, comparing the ADC_{EAA} there seem to be significant ($P < 0.05$) differences. Generally, FSS: SBM had higher ADC_{EAA} values than other meals while the solar-dried shrimp head meal had the lowest values.

Digestibility values are an important parameter to consider in diet formulation and in determining the utilization of a feed. Feedstuffs which are poorly digested would be of limited nutritional value to an animal; thus, digestibility values reported in this study suggest that shrimp head silage meals represent an alternative form of protein feedstuff in aquaculture feeds and also provide basic information for further work using pelleted fish feeds containing a fermented shrimp head silage as protein feedstuff which will result in lower costs for aquaculture production.

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