IMPROVEMENT IN THE NUTRITIVE QUALITY OF SOYABEAN MEAL BY CO-ENSILING WITH UNDER-UTILIZED FISH DISCARDS

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ABSTRACT

Improvement in the nutritive value of soybean meal was investigated by Co-ensiling it with underutilized trash fish discards (gizzard shad) at different proportions. The following proportions of Gizzard shad to soybean meal were used; (a) 100% gizzard shad + acid combination (b) 80% Gizzard shad + 20% soybean meal & 10%WB (c) 60% Gizzard shad + 40% soybean meal & 10%WB. (d) 100% Gizzard shad without acid combination. Co-ensiling was achieved by adding sufficient acid to produce a paste. Products were neutralized by addition of 2% (by weight) calcium hydroxide and drying was effected by freeze-drying.

The dried silage products were stored at low temperatures. Products were analysed for proximate composition and amino acid composition. The amino acid composition and ration of essential amino acid: Non essential amino acid (EAA/NEAA) was used as index of nutritive quality. Also essential amino acid profile of the co-ensiled products were compared with essential amino acid requirement of some warmwater fish species to estimate their nutritive usefulness for these species.

INTRODUCTION

The increasing world demand for high protein crops and the prospect for animal feedstuffs have prompted considerable interest in the development of legume seed crops. One of the most agronomically important legume seed crop is the soybean. Among plant proteins, soybean meal is the most promising candidate for replacing part or all of fish meal protein in fish diets. The use of soybean meal in this respect is becoming increasingly important in the developing and developed countries of the world because of dwindling fish resources from the oceans and the high price of fish meal.

Soybean meal as a protein supplement has been used with varying success in the diet of several fish species, tont (Dabrowski *et al* 1985), African Catfish (Balogun and Ologhobo, 1989) tilapia (Davis and Stickney, 1978) and Common carp (Abel *et al* 1984 Marai *et al* 1986) and grass carp (Dabrowski and Kozak, 1979).

A major problem associated with raw and inadequately heated soybean meal is the presence of protease inhibitors and phytic acid; Heating during commercial oil extraction

(105°C for 10 - 20 min) destroys most of the protease inhibitors. To the processor, maximum heat treatment represents an additional cost and to the fish farmer, the reduction in protein quality resulting from unavailability of the critical amino acid.

Methioninelhysine represents an additional increasing costs since he has to feed more to satisfy the requirement of the fish for these amino acids. Additional cost is also expended in supplementing with a high amount of zinc to neutralize the effects of plytic acid.

One processing technique that can improve the nutritive value of plant proteins significantly at high reduced cost is by co-ensiling them with whole trash underutilized fish or fish offals. The method follows the principle of silaging as a means of preserving fish and utilizing waste fish by a combination of both organic and inorganic acids. Stone et al (1984) improved the nutritive quality of canola meal significantly by co-ensiling it with fish. A better balance in the amino acid composition of canola meal was achieved.

This paper presents the investigation on the changes brought about in the overall nutritive quality of soybean meal when it is co-ensilied with gizzard shad - an underutilized freshwater clupeid.

2.0 MATERIALS AND METHODS

2.1. The selection of material for the ensiling process was carefully done given consideration to (a) the availability of large volume of uneconomic small size by-catches (trash fish) in Nigeria Marine and freshwater capture fisheries (b) the effective utilization of small size tilapia fish produced on most fish farms in Nigeria. Thus gizzard shad average weight 12.41g which represents one of the by-catches (trash fish) from commercial fisheries in Lake Erie. They were collected and kept at -5°C and transported in 250 gallon capacity tightly sealed plastic containers. The whole fish materials were minced in a Hobbert Mincer and divided into equal parts. Silaging/co-silaging was made through the addition of sulfuric acid (2.00%) and propionic acid (0.75%) as follows:

(i) 100% minced fish + 2% H_cSO_4 + 075% propionic acid.
(ii) 60% minced fish + 40% soybean (90% SB + 10% WB) + acid combination.
(iii) 80% minced fish + 20% soybean meal (90% SB + 10% WB + acid combination.
(iv) 100 minced fish without acid combination (Control 1)
(v) 100% minced fish without acid combination (freeze-dried immediately).

A 50% diluted 2.00% H_2So_4 was used. Ensiling was carried out at room temperature in 50-litre container. Anti-oxidant (0.05%) was added to each of the six containers to stabilize the lipid.

All materials were ensiled for a period of 30 days. Samples were taken every 5 days for 1st measurement. At the end of ensiling period, neutralization was carried out by adding appropriate levels of calcium hydroxide. The resulting materials were freeze-dried. The dried silage products were stored at 15°C until they were used for chemical analysis or biological evaluation.

Chemical analysis

P^H Measurement: Approximately, 29m of the liquid silage was diluted with 20mls of de-ionised water. The mixture was thoroughly mixed by vortexing for 30 seconds and centrifuged in a refridgerated centrifuge and the supernatant used for P^H measurement.

Proximate composition

Moisture and drymatter were determined by AOAC (1970) method. Ether extract was estimated from the data presented for gizzard shad by Strange and Pelter (1987). Protein was estimated as the sum of total amino acid content after amino acid analysis.

Total amino acid composition.

At each sampling period, samples were withdrawan and freeze-dried immediately. Freeze-dried duplicate samples were pulverised and amount equivalent to 40mg protein was used for total amino acid analysis.

Samples were oxidized at 0°C with performic acid oxidation for 16 hours for the conversion of Cystine to cysteic acid and methionine to mehtionine sulphone. This was followed by 6N HCI hydrolysis at 110°C for 24 hours and drying under vacuum. After acid removal by rotary evaporation. Coupling of amino acids with phenylisothiocyanate was carried out. Reverse-phase HPLC separation of PITC-derivatized amino acids was carried out according to the procedure described by Henrickson and Meredith (1984). Calibration of standards were as described by Millipore Corporation (1990).

Free amino acids (FAA)

Free amino acids were measured after extracting with 4% sulphosalicylic acid (Stone et al 1989) and filtering through a 0.45um filter. Derivatization and elution followed the procedure described by Henrickson and Menedith (1984).

3.0 RESULTS AND DISCUSSION

The average P^{H} of the ensiled products are presented in Table 1. The P^H varied between 2.01 ± 0.13 and 2.25 ± 0.17 in all ensiled products except in products VI and VII (controls) where average P^H values were 7.21 ± 0.09 and 7.11 ± 0.11. Slight variations were observed in the PH of the ensiled products over the 30 days storage period. Apart from the bacteriostatic effect at this low P^H the P^H is important because it gives an indication of the degree of neutralization for the processed ensiled products. Stone et al (1989) for example, used 1.6% of Calcium hydroxide for neutralisation after ensiling for 40 days. In the present study however, 2.0% was required to neutralise the ensiled products.

The results presented in Table 2 are the changes in dry matter and protein content of the silage/co-ensiled products. Ensiling gizzard shad alone yielded a product with 44.77% protein. Co-ensiling it with soybean meal yielded products containing 36.31% and 39.71% protein. Co-ensiling with soybean meal resulted in products that are significantly lower in protein than the originally ensiled fish product. No significant differences (P>0.05) were obtained in the dry matter of all products. Table 3 compares the amino acid composition of fish meal with that of soybean meal. Table 4 presents the total amino acid composition of the ensiled gizzard shad and shad co-ensiled with soybean meal.

The ratio of essential amino acid to non-essential amino acid (EEA:NEAA) of the ensiled products were also presented in Table 4, lysine and methionine content in soybean meal were 3.23gm/100gm DM and 0.62gm/100gm DM respectively (Table 3). When soybean was co-ensiled with gizzard shad, a significant improvement was obtained in the methionine content of product UI (80% gizzard shad + 20% soybean meal). The EEA:NEAA obtained for this product was also not significantly different from that obtained for commercially available fish meal whose EAA:NEAA was 0.87 (Table 3). Based on the EAA:NEAA values obtained for the ensiled products, co-ensiling of fish discards in the present study approved to have improved the essential amino acid balance in Soybean meal especially when a ratio of 80% discard is ensiled with a ratio of 20% soybean meal.

The essential amino acid profile of the ensiled products was compared with essential amino acid requirement of essential of Channel catfish and rainbow trout (Table 4).

The NRC, (1983) EAA requirement for these fish species showed that product II (60% fish + 40% soybean meal) is deficient in arginine and methionine for rainbow trout and not for catfish.

To further examine the changes in the nutritive value of the silage products, changes in the essential amino acid pattern were determined and presented in Table 5. In all essential amino acid measured, lossers are less than 10%.

This is an indication that losses in essential amino acid and hence in the intrinsic nutritive value of silage products are minimal for the co-ensilaging technique used in the present study.

During the process of silaging, numerous digestive and catheptic enzymes produce a liquified product which is rich in proteins, polypeptides and free amino acids (Stone and handy, 1986). Free amino acids expressed as a percentage of the total amino acids are presented in Table 6. In the final products, free amino acid levels constituted between 9% and 11% of the total amino acid. Higher levels of free amino acids were obtained in the essential amino acids of all the silage products. Although the levels were rather low, evidence has been presented (Geiger, 1947, Dabronski and Dabrowska, 1981, Stone *et al* 1989) that free essential amino acids are rapidly absorbed than amino acid from intact proteins. They (free amino acids) are irreversibly metabolised and not available for protein synthesis. The levels of free amino acids in these silage products may be too low to have elicit any serious implications on their nutritive values when fed to fishes.

TABLE 1: AVERAGE p^H VALUE OF ENSILED GIZZARD SHAD AND FISH CO-ENSILED WITH SOYBEAN AND FEATHER MEAL

Ensiled Products	Average. p^H
I	2.01 ± 0.13
II	2.25 ± 0.17
III	2.12 ± 0.11
IV	7.21 ± 0.09
V	7.11 ± 0.11

I = 100% gizzard shad + acid combination (30 days storage)

II = 60% gizzard shad + 40% soybean meal (30 days storage)

III = 80% gizzard shad + 20% soybean meal (30 days storage)

IV = 100% gizzard shad without acid (frozen for 30 days)

VII = 100% gizzard shad (minced and freeze dried immediately)

TABLE 2: Changes in Dry Matter and Protein Content of Dried Shad Silage and Co-ensiled Products at the Initial and End of Silaging Period

	Dried Products	Dry Matter		Protein	
		Day 0	Day 30	Day 0	Day 30
I	Gissard Shad Silage	95.14	96.05	44.99[a]	44.77[a]
II	Gizzard Shad (60%) + 40% Soybean Meal Silage	94.96	95.31	38.62[b]	36.31[b]
III	Gizzard Shad (80%) + 20% Soybean Meal Silage	95.04	95.12	41.71[ab]	39.71[ab]
IV	Gizzard (frozen)	95.84	95.86	45.66[a]	43.97[a]
V	Gizzard Shad (freeze-dried immediately)	95.80	----	47.18	----

TABLE 3: AMINO ACID COMPOSITION OF COMMERCIAL FISH MEAL, FEATHER MEAL AND SOYBEAN MEAL (GM/100 GM GM)

Amino Acid	Fish Meal	Soybean Meal
Aspartic acid	5.47	3.88
Glutamic acid	9.45	7.05
Serine	2.76	7.05
Glycine	6.38	2.52
Histidine	1.76	1.04
Arginine	3.23	2.98
Theronine	2.36	1.59
Alanine	5.09	3.84
Proline	4.10	3.60
Tyrosine	2.09	1.26
Valine	3.02	3.43
Methionine	2.81	0.62
Cystine	0.81	0.71
Isoleucine	2.88	2.69
Leucine	5.45	3.71
Phenylalanine	2.80	2.43
Lysine	5.25	3.23
Total Amino Acid	65.71	46.49
Total Essential Amino Acid (EAA)	30.37	22.43
% EAA of Total	46.22	48.25
% NEAA of Total	53.28	51.75
EAA:NEAA	0.87	0.93

TABLE 4: Amino Acid composition of Ensiled Gizzard Shad and Shad Co-Ensiled with Feather Meal in comparision with Fish Meal in comparison with Fish Meal and Soyabean g/100 gDm (after 30 days) in comparison with EAA requirement of Channel Catfish and trout.

Amino Acid	Silage Products (freeze dried)				
	I	II	III	IV	V
Aspartic acid	3.44	2.78	2.86	3.67	3.82
Glutamic acid	7.71	7.36	7.28	7.64	7.17
Serine	1.75	1.48	1.82	1.68	2.09
Glycine	4.23	2.31	2.85	4.47	4.41
Histidine	0.94	0.78	0.85	1.06	1.09
Arginine	3.30	1.80	1.76	3.15	4.36
Threonine	1.57	1.29	1.42	1.51	1.62
Alanine	3.29	2.29	2.52	3.08	3.01
Proline	2.30	2.32	2.60	2.26	2.21
Tyrosine	1.33	0.96	1.21	1.24	1.56
Valine	1.97	1.63	1.95	1.84	1.88
Methionine	1.46	0.92	1.19	1.57	2.37
Cystine	0.50	0.52	0.58	0.54	0.80
Isoleucine	1.50	1.40	1.58	1.27	1.57
Leucine	2.86	2.15	2.79	2.91	2.66
Phenylalanine	2.40	2.38	2.50	2.20	2.55
Lysine	4.22	3.24	3.41	3.82	4.01
Total Amino Acid	44.77	36.31	39.71	43.91	47.18
% Essential Amino Acid (EAA)	46.28	44.09	45.41	45.25	48.61
% Non-Essential Acid (NEAA)	53.72	55.91	54.59	54.75	51.39
EAA:NEAA	0.86	0.79	0.83	0.82	0.95

TABLE 5: Changes in essential Amino Acid composition of Ensiled Gizzard Shad and Shad Co-Ensiled with Soybean during 30 day storage period (%).

Amino Acid	ESILED PRODUCTS				
	1	2	3	4	5
			% Change		
Histidine	2.92	3.41	4.90	0.00	0.00
Arginine	2.77	1.89	6.25	6.62	3.11
Threonine	1.33	5.30	2.70	4.58	0.58
Valine	5.80	7.52	1.12	2.43	0.00
Isoleucine	0.68	6.54	1.88	5.04	5.88
Leucine	1.79	4.61	2.70	1.69	4.75
Lysine	1.13	2.77	1.87	5.42	2.25
Methionine	6.59	3.88	0.00	0.00	7.04
Phenylalanine	3.25	6.22	1.21	0.52	5.80

Ensiled products 1 - 5 are listed in Section 2.1

TABLE 6: Free Amino Acid composition of Gizzard Shad and Shad Co-Ensiled Soybean Meal and Feather Meal (g/100 g DM).

Amino Acid	I	II	III	IV	V
Aspartic acid	0.21(6.1)	0.18(6.5)	0.17(5.9)	0.15(4.1)	0.12(3.14)
Glutamic acid	0.58(7.5)	0.50(6.8)	0.55(7.00	0.34(4.5)	0.20(3.91)
Serine	0.12(6.9)	0.14(9.6)	0.12(6.6)	0.08(4.8)	0.06(2.9)
Glycine	0.29(6.9)	0.23(9.9)	0.20(7.0)	0.19(4.5)	0.16(3.6)
Histidine	0.06(6.4)	0.05(6.4)	0.07(8.2)	---	---
Arginine	0.24(7.3)	0.14(10.0)	0.14(7.9)	0.15(4.8)	0.11(2.5)
Threonine	0.10(6.4)	0.08(6.2)	0.09(6.3)	0.04(2.7)	0.04(2.5)
Alanine	0.16(4.9)	0.19(8.3)	0.18(7.1)	0.14(4.6)	0.12(3.9)
Proline	0.18(7.8)	0.20(8.6)	0.22(8.5)	0.11(4.9)	0.13(5.8)
Tyrosine	0.10(7.5)	0.04(4.2)	0.07(5.8)	0.05(4.0)	0.05(3.2)
Valine	0.15(7.6)	0.12(7.4)	0.14(7.2)	0.09(4.9)	0.08(4.3)
Methionine	0.07(4.8)	---	---	---	---
Isoleucine	0.16(10.7)	0.16(11.4)	0.14(8.9)	0.10(7.9)	0.08(5.1)
Leucine	0.13(4.6)	0.11(5.1)	2.12(4.3)	0.10(3.4)	0.08(3.0)
Phenylalanine	0.24(10.0)	0.20(8.4)	0.22(8.8)	0.10(4.6)	0.12(4.7)
Lysine	0.44(104)	0.46(14.2)	0.48(12.1)	0.31(8.1)	0.27(6.9)

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