GLOBAL JOURNAL OF PURE AND APPLIED SCIENCES VOL. 13, NO. 4 2007: 457- 462
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NUTRITIVE COMPOSITION AND PHYSICAL CHARACTERISTICS OF SUPPLEMENTED LMITATION MILK FROM AFRICAN YAM BEAN (*SPHENOSTYLIS STENOCARPA*)

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(Received 27 March 2007; Revision Accepted 8 May 2007)

ABSTRACT

The effect of processing techniques on the chemical composition and physical properties of imitation milk from African yam bean (AYB) was evaluated. Milks extracted from whole bean flour had a protein content of 3.7%, which was significantly ($P<0.05$) higher than milks extracted from soaked or blanched seeds. The increase in protein content of milks due to supplementation with 4% milk protein concentrate exceeded 50%. The lysine content (g/100g protein) of supplemented milks was about 18% lower than the values for the seeds. Potassium and phosphorus were the most abundant minerals in AYB seeds and milks. Production of supplemented milk resulted in reductions of 100% sucrose and 72.1 – 75.4% stachyose and raffinose. Thermal treatment increased the lightness (L-value) and reduced yellowness (b-value) of milks significantly ($P<0.05$), and dehulling the beans significantly ($P<0.05$) increased the L-values of milk. Heat processing of milks at 95°C for 20 min was suitable for color stability.

KEYWORDS: African yam bean, heat processing, imitation milk, Nutritive composition, *Sphenostylis stenocarpa*

INTRODUCTION

The shortage of animal proteins in some parts of the world has led to a high level of dependence on plant proteins. However, several legumes are under-utilized and research efforts have been geared towards the development of acceptable products to improve their utilization. A variety of dairy-like products, including imitation and fermented milk products, have been developed from legumes and oilseeds. Apart from soymilk, which is produced commercially in many countries world-wide, imitation milks have been developed from peanut (Beuchat and Nail, 1978; Bucker *et al.*, 1979) sunflower (Ferber and Cooke, 1979), cowpea and mung beans (Caygill *et al.*, 1981;Rao *et al*, 1988), and lupin (Jimenez-Martinez *et al.*, 2003). Lactic fermentation of imitation milks was found to improve the nutritional value and prolong the shelf-life of these products (Beuchat and Nail, 1978).

African yam bean is a common legume inWestern and Central Africa,but it is under exploited (Nnam, 1997). The plant produces nutritious seeds and edible tubers, and is well adapted to tropical conditions (Apata and Ologhobo, 1990). Fermented dairy-like products, including a cheese-like product (Ofuya *et al.*, 1991) and vegetable milk fermented by indigenous microflora (Nnam, 1997) have been developed from the bean. These products were both well accepted at the experimental level and the protein content in vegetable milk was comparable to soymilk. Various methods have been utilized during the production of legume-based milks. Differences in processing techniques have been reported to affect the chemical composition of soymilk and its capacity to support the growth of lactic acid bacteria (Wilkens and Hackler, 1969; Kothari, 1973). It is expected that different processing techniques would affect the quality of African yam bean milk.

There is a need to study the effects of different processing methods on the quality of African yam bean milk because it can potentially be developed into fermented dairy-like products. The objective of this study was to evaluate the nutritive composition and physical properties of imitation milk prepared from African yam bean by subjecting the bean to different processing conditions.

MATERIALS AND METHODS

Materials

African yam beans (marble variety and cream-colored seeds) were purchased from Creek Road market, Port Harcourt, Nigeria. The beans were packaged in polyethylene bags, transported to the laboratory and kept at 4°C until required.

Bean flours

Five hundred grams of each bean type was mechanically dehulled with a pearler (Strong – Scott 19810, Seedburo Equipment, Chicago, IL). The dehulled beans were sieved through No. 6 USA Standard Testing Sieve (Pore diameter 3.35mm, Fisher Scientific, Pittsburgh, PA) and the portion retained in the sieve was collected. Dehulled and whole seeds were milled in a sample mill (Brinkman model ZMI, GmbH and Co., Hann, Germany) and passed through a 0.5mm screen to obtain flours.

Soaking

One hundred grams of each bean type was soaked in triplicate in 500 mL of 0.5% $NaHCO_3$ for 12hr at ambient temperature (23 ± 1°C). The soaked beans were drained and boiled in tap water for 5 min.

Milk preparation

African yam bean milk was prepared in triplicate from both bean flours and soaked beans (Fig.1). The flours and soaked beans were blended separately with water (1:4 seed: water) in an Osterizer® Laboratory Blender (Nelson-Jameson, Marshfield, WI) for 3 and 5 min, respectively. The bean flour was blended with water at 80-85°C, and cold tap water was used for the seeds. The resulting slurry was filtered through two layers of double-folded cotton cheese cloth (Nelson-Jameson, Marshfield, WI), and coarse particles were removed by settling (10 min) or centrifugation (IEC HT Centrifuge, Damon/IEC Division, Needham Heights, Mass) at 3000 ***g*** for 5 min. The milk was heat processed at 85°C and 95°C for 20 and 30 min in a water bath, with stirring. African yam bean milk was fortified at 4% (w/w) with milk protein concentrate containing 56% protein (ALAPRO® 4560, NZMP (USA), Inc., Lemoyne, PA).

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Analytical Procedures

The pH of milk samples was determined using a combination glass electrode (Fisher Scientific, Pittsburgh, PA) and a pH meter (Corning Glass Works, Medfield, MA). Proximate analyses on the bean components were determined by methods from the Official Methods of Analysis of the AOAC International (AOAC, 2000): total solids were determined gravimetrically by force oven air drying (AOAC 925.23), moisture by forced air oven drying (AOAC 925.10), ash by gravimetric method in a furnace ≤ 550°C

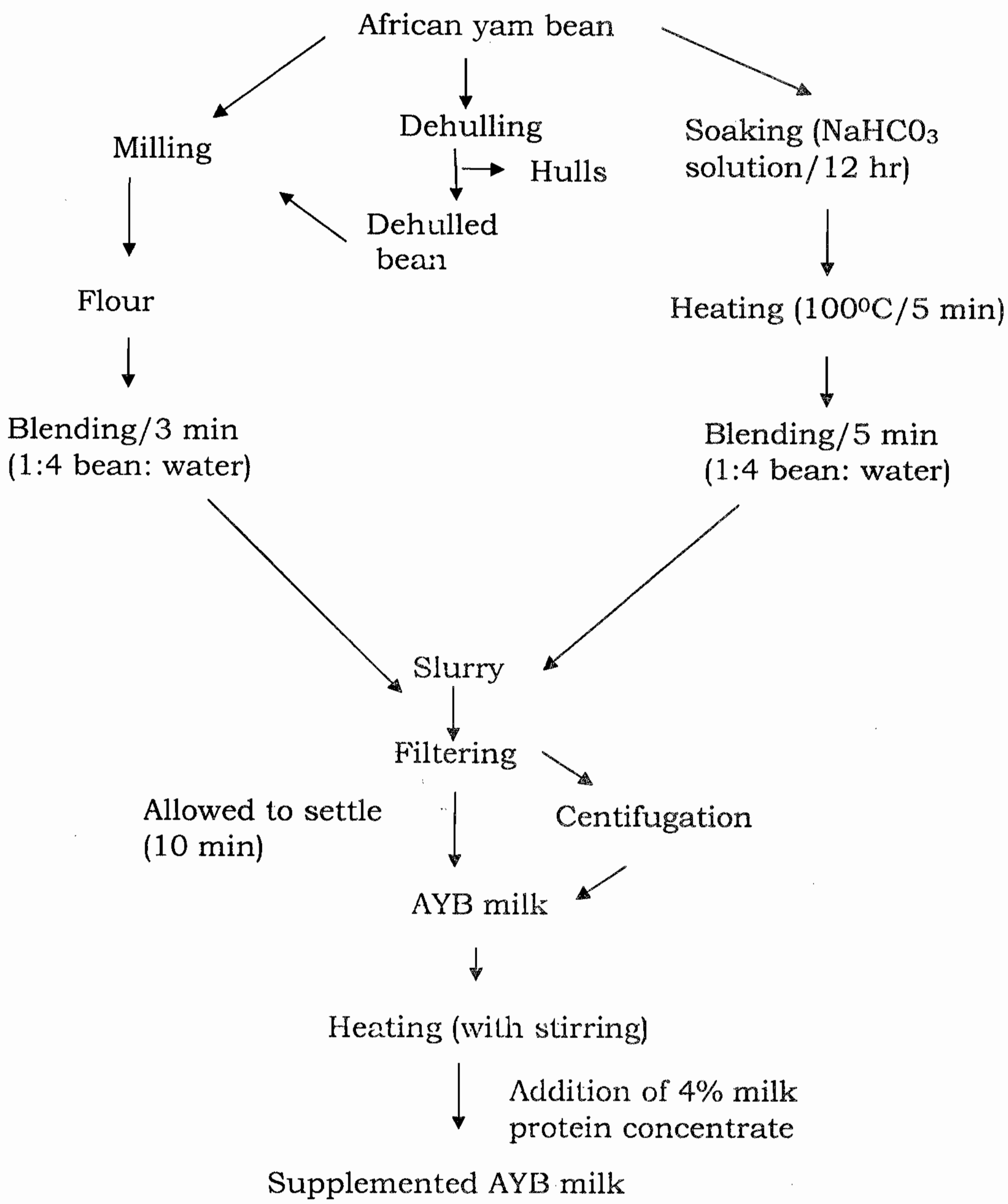

Fig 1: Flow chart for the preparation of African yam bean (AYB) milk.

(AOAC 945.46) and crude protein by Kjeldahl (AOAC 991.20). Total nitrogen was converted to protein using the factor 6.25. Fat content was determined by the Mojonnier ether extraction method (Atherton and Newlander, 1977).

Sugar, amino acid and vitamin analysis were performed using the following methods: The amino acid composition was determined by HPLC (Medallion Laboratories, Plymouth, MN). Sugar composition was determined using AOAC method 977.20. Riboflavin and thiamin contents were determined by a fluorometric procedure (AOAC 942.23, 970.65 and 981.15). Niacin content was determined microbiologically (AOAC Method 944.13), and mineral analysis was performed with Inductively Coupled Plasma Optical Emission Spectroscopy (Fassel and Kniseley, 1974).

The color of the milk samples was measured in terms of L-, a- and b-values, using a McBeth colorimeter (Minolta Co., Japan). The colorimeter was calibrated with a white tile which had color values L=97.3, a=-0.77, b=2.39. Color values were determined in triplicate.

Statistical Analysis

Data were evaluated by analysis of variance (ANOVA) using the statistical package for social science research (SPSS, 1998) software. Duncan's multiple range test was used to determine if there was a significant difference (P<0.05) between means.

RESULTS

The proximate composition of milks prepared from African yam bean is shown in Table 1. The milk that was extracted from whole flour and allowed to settle, had a significantly (P<0.05) higher volume than those extracted by other processing methods.

Table 1: Effect of processing method on proximate composition, volume and pH of African yam bean (AYB) milks

Sample	Milk volume (ml/100g	PH	Total solids %	Crude protein %	Fat %	Ash %
Marble variety						
Milk from whole flour	282^{a}	6.24^{b}	8.15^{c}	3.69^{c}	0.52^{b}	0.54^{bc}
+ supplementation	__*	6.22^{b}	12.6^{a}	5.74^{a}	0.67^{a}	0.80^{a}
Milk from dehulled flour	263^{b}	6.31^{b}	9.01^{b}	4.54^{b}	0.62^{a}	0.65^{ab}
Cream-colored Seed						
Milk from soaked/blanched seed	240^{c}	6.60^{a}	3.73^{e}	0.850^{d}	0.23^{c}	0.17^{d}
Milk from blanched seed	260^{b}	6.40^{b}	3.41^{e}	1.14^{d}	0.18^{c}	0.22^{d}
Milk from whole flour	285^{a}	6.31^{b}	8.48^{c}	3.74^{c}	0.55^{b}	0.53^{bc}
+ Supplementation	__*	6.26^{b}	12.3^{a}	5.72^{a}	0.64^{a}	0.79^{a}
Milk from whole flour (centrifuged)	265^{b}	6.31^{b}	7.53^{d}	3.92^{c}	0.58^{a}	0.48^{c}
Milk from dehulled flour	262^{b}	6.28^{b}	8.85^{b}	4.46^{b}	0.63^{a}	0.63^{b}

Values are means of 3 replicates. Means in the same column followed by different superscripts are different (P<0.05).
*Not determined.

The total solids content of milks extracted from bean flours exceeded 100% of the levels in milks extracted from blanched or soaked seeds. The increase in protein contents of milk samples due to supplementation, exceeded 50%. The pH values for the milks ranged from 6.21- 6.60 and only the extract from beans soaked in 0.5% $NaHCO_3$ had a significantly (P<0.05) higher value.

The amino acid composition of beans and supplemented imitation milks are presented in Table 2. The two types of African yam bean had similar amino acid composition and the supplemented vegetable milks also had comparable amino acid contents. The levels of methionine and lysine in the milk proteins were lower than values for the seeds, and the differences were more for lysine.

Cream-colored African yam bean had slightly higher levels of minerals than marble variety, with the exception of potassium and magnesium (Table 3). The most abundant mineral in the beans was potassium, followed by phosphorus and then magnesium. Potassium and phosphorus were also the most abundant minerals in the milks, followed by calcium. The levels of vitamins in the beans were in the order of niacin>thiamin>riboflavin. Thiamin and Niacin contents of the cream-colored bean were slightly higher than values for the marble variety. The stachyose content of African yam bean was higher than the levels of sucrose and raffinose (Table 4). The raffinose content was about 78% lower than the stachyose content.

Cow milk had a significantly (P<0.05) higher lightness (L-value) and lower yellowness (b-value) than the vegetable milks (Table 5). Non-heated AYB milk prepared from whole marble variety seeds had the lowest L-value among the AYB milk samples and did not differ significantly (P<0.05) from the value for commercial soymilk. The L-

Table 2: Amino acid composition (g/100g protein) of African yam bean seeds and milks.

Amino acid	Marble variety seed	Cream-colored seed	Milk from Marble variety (supplemented)	Milk from cream-colored seed (supplemented)	Soybean*
Essential					
Arginine	5.33	5.59	3.68	3.60	7.4
Lysine	7.54	7.83	6.29	6.39	4.1
Tyrosine	4.15	4.27	4.38	4.65	4.2
Phenylalanine	5.59	5.46	4.55	4.65	5.1
Methonine	1.54	1.52	1.42	1.62	1.3
Leucine	7.15	7.17	9.26	8.84	7.8
Isolencine	4.07	3.88	4.90	5.00	5.8
Threonine	4.08	3.58	6.05	5.34	4.2
Valine	4.47	4.24	6.29	6.39	5.3
Non-essential					

Alanine	4.42	4.42	6.05	6.04	5.2
Aspartic acid	9.99	10.1	13.8	11.8	1.3
Glutamic acid	12.3	12.6	14.5	14.8	19.4
Glycine	4.12	4.15	3.68	3.90	4.6
Proline	4.12	4.24	4.00	4.47	5.4
Serine	5.16	5.56	8.91	8.67	5.8
Histidine	4.20	4.41	2.46	2.72	2.6

[a]Source: Sattar *et al.* (1990)

Table 3: Mineral and vitamin composition of African yam bean seeds and milk (As Is)

Component	Marble variety seed	Cream-colored seed	Milk from Marble variety	Milk from cream-colored seed	Milk from Marble variety (Supplemented)	Milk from cream-colored seed (Supplemented)
Minerals (g/kg)						
Macroelements						
Ca	0.33	0.36	0.06	0.06	0.80	0.78
P	2.5	2.6	0.54	0.49	1.00	0.96
K	9.7	9.5	2.3	2.3	2.6	2.7
Mg	1.6	1.5	0.34	0.34	0.37	0.37
Microelements						
Mn	0.03	0.03	0.01	0.01	0.01	0.01
Zn	0.02	0.02	0.01	0.01	0.01	0.01
Fe	0.05	0.07	0.01	0.01	0.01	0.01
Cu	0.01	0.01	0.00	0.00	0.00	0.00
Vitamins (mg/100g)						
Thiamin	0.19	0.22	ND*	ND	0.06	0.05
Riboflavin	<0.01	<0.01	ND	ND	0.16	0.11
Niacin	3.6	4.3	ND	ND	0.89	0.92

Values are means of 2 replicates
*ND, not determined.

Table 4: Sugar profile of African yam bean seeds and milk (g/100g, As Is)

Sugars (%)	Marble variety seed	Cream-colored seed	Milk from Marble variety (Supplemented)	Milk from cream-colored bean (Supplemented)
Fructose	X	X	X	X
Glucose	X	X	X	X
Sucrose	1.2	1.9	X	X
Maltose	X	X	X	X
Lactose	X	X	X	X
Raffinose	0.57	0.65	0.14	0.17
Stachyose	2.7	2.9	0.72	0.81

X, not detected.

Table 5 – Colorimeter parameters L, a and b for cow, soy and African yam bean (AYB) milks

Milk	L	a	b
Cow	72.1^{a}	-4.67^{e}	-2.47^{d}
Soy	56.9^{e}	-4.30^{e}	4.39^{c}
Cream-colored AYB			
Milk from whole flour (non-heated)	59.2^{d}	-0.67^{c}	8.74^{a}
Milk from whole flour boiled at 95°C for 20 min	68.3^{b}	-1.48^{d}	7.53^{b}
Milk from dehulled flour (non-heated)	61.5^{c}	-1.81^{d}	8.61^{a}

Marble Variety			
Milk from whole flour (non-heated)	57.9^{e}	0.08^{b}	9.47^{a}
Milk from dehulled flour (non-heated)	60.7^{cd}	0.99^{a}	8.59^{a}

and b-values for the other AYB milk samples were significantly higher than the value for soymilk. Color changes occurred in milk processed at 85°C for 20 and 30 min and stored at 4°C (Table 6). The lightness of these milk samples increased significantly (P<0.05) during storage while changes in a- and b- values were not significant. On the other hand, milk heat-processed at 95°C for 20 and 30 min showed little or no change in color values.

Table 6: Effect of heat processing on color of African yam bean milk stored at 4°C.

Heat processing conditions	Storage time (weeks)	L	A	b
85°C for 20 min	0	53.5^{d}	0.82^{a}	14.3^{a}
85°C for 30 min		54.1^{d}	0.83^{a}	13.8^{ab}
95°C for 20 min		68.3^{a}	-1.48^{bc}	7.53^{c}
95°C for 30 min		67.3^{a}	-1.15^{b}	7.54^{c}
85°C for 20 min	2	57.6^{c}	1.05^{a}	13.5^{ab}
85°C for 30 min		58.7b^{c}	0.897^{a}	13.0^{b}
95°C for 20 min		68.4^{a}	-1.45^{bc}	7.42^{c}
95°C for 30 min		67.3^{a}	-1.09^{b}	7.60^{c}
85°C for 20 min	4	59.3^{bc}	1.10^{a}	13.0^{b}
85°C for 30 min		60.4^{b}	0.971^{a}	12.6^{b}
95°C for 20 min		68.0^{a}	-1.50^{bc}	7.52^{c}
95°C for 30 min		67.0^{a}	-1.09^{b}	7.59^{c}

DISCUSSION

Chemical composition

The most pronounced differences in proximate composition were in total solids and protein contents. The total solids and protein contents of milks extracted from whole bean flours exceeded the contents in milks from soaked or blanched seeds by 2- and 3-fold, respectively. Milks prepared from dehulled bean flours had significantly (P<0.05) higher total solids and protein contents than milks obtained from whole flour and soaked or blanched seeds. The higher total solids content results from the failure of the cheese cloth to separate much of the coarse material from the extract. The protein contents of milks prepared from AYB seed were similar to the value (approximately 1.0%) for sunflower milk (Ferber and Cooke, 1979) and cowpea milk (Rao *et al*, 1988), but the level in milk prepared from bean flour was similar to the value for soymilk (3.8%) (Pinthong *et al.*, 1980).

The amino acid content of AYB seed was comparable to that of soybean (Fig. 2). The results obtained in this study confirm the findings of Nnam (1977), who stated that the lysine and methionine contents of AYB are similar, or higher, than the contents in soybean. More than 50% of amino acids assessed had higher values per 100g protein in supplemented milks than in the seeds. This indicates that milk production from AYB and supplementation can serve as a means of protein fortification. . The levels of lysine in supplemented milk proteins were 16.6 and 18.4% lower than the protein content of the marble variety and cream-colored seeds, respectively.

Potassium and phosphorus were the most abundant minerals in AYB seeds and milks. The increase in calcium in the milks is due to supplementation with milk protein concentrate. Among the vitamins, only riboflavin contents were higher in the supplemented milk samples than in raw beans due to supplementation.

The sugars detected in AYB seeds were sucrose, raffinose and stachyose. Stachyose and raffinose are flatulence-causing oligosaccharides, which are attacked by microorganisms in the gastrointestinal tract because of a lack of α-galactosidase in the upper intestinal tract (Charley and Weaver, 1998). Sucrose was not detected in the supplemented milks, but the levels of raffinose and stachyose were reduced by 72.1-75.4%.

Milk color

Thermal treatment of AYB milk increased lightness and reduced yellowness significantly (P<0.05). This may be due to inactivation of enzymes present in the seeds. Thermal treatment at 93°C for 20 min inactivates lipoxygenase in soybean protein products (Wolf, 1977). Dehulling of seeds prior to extraction of milk also increased the lightness of milk significantly, but the reduction in b-values was not significant. Milk heat-processed at 95°C for 20 and 30 min and refrigerated showed little or no change in color values, indicating good stability in terms of color. Milk samples processed at 85°C had significantly lower lightness and higher yellowness values than those processed at 95°C. Although the color values for milk processed at 95°C for 20 and 30 min were similar, the latter treatment resulted in slightly lower L-values over the entire storage period. This may be due to heat induced browning. The findings in this study indicate that the extraction of AYB milk from bean flour and heat processing at 95°C for 20 min are suitable processing conditions.

ACKNOWLEDGEMENT

The study was supported by a Fulbright research grant.

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