Blending Whey Powder with Haricot Bean Powder for Weaning Food Production

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Abstract

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Enriched foods consist of a mixture of cereals, pulses with vitamins and minerals, intended to provide a balanced intake of essential nutrients for vulnerable groups. Whey is the milk component remaining after removal of casein, as by curdling. The large amount of whey generated by dairy industries during cheese production currently is discharged into nearby streams, causing environmental pollution. The main aim of this study was to produce weaning food by blending whey powder with haricot bean powder. The raw materials used were whey and haricot bean (Phaseolus vulgaris), Roba 1 variety. Whey was filtered through an ultra-filtration membrane, permeate was stored at 5 °C and then pre-heated at 55 °C for 40 min. Vacuum pump was used to concentrate the soluble solid to 60%, and it was cooled to 5 °C. Finally, it was dried by means of a pressure-nozzle spray dryer, and powder product was obtained. The bean seeds were weighed into a plastic pan and soaked in distilled water for 12 h at 25 °C. The pan was covered with clean muslin and kept in darkness at 25 °C to sprout. Seeds that sprouted within four days were dried, ground and stored at 4 °C. The whey and haricot bean powder were mixed in various mix ratios and analyzed for their physico-chemical and bacteriological quality. The protein, iron and fat content of whey powder was 7.9%, 1.14% and 1.12% respectively. The fat was increased to 1.95%, protein to 13.94% and iron to3.02% through blending with haricot bean powder. The microbiological results of all fortified products were within the limits recommended. This study clearly indicates that there is a possibility of blending of whey powder with the high protein content of haricot bean powder, providing an option to develop low-cost weaning and supplementary foods from pulses. The nutritional content of a mix of whey: haricot bean powder in the ratio 80:20 was the best composition for fulfilling the nutritional requirements for young children.

Key words: fat, haricot bean, protein, whey, whey powder

INTRODUCTION

Whey is the milk component remaining after the removal of casein, as by curdling. Whey contains approximately half of the original nutrients of milk (Omar et al, 2005). When humans began to make cheese, the secondary product, whey, presented a challenge for utilization (Webb, 1998). Whey has a high biological oxygen demand (BOD), and must be treated prior to disposal. Bioconversion of lactose in whey to ethanol, yeast biomass or methane has been used to produce saleable products, while reducing the organic load by >75% (Karand Misra, 2000).

The food industry produces a considerable amount of waste products that are still rich in organic substances, although they are a potential source both for the extraction of valuable compounds and for the production of edible biomass (Carlo, et al. 1999). This is also true of cheese whey, which is the major by-product of the cheese-making industry, and which in the past was often regarded as a waste product, and caused environmental pollution by its disposal. Considerable efforts have recently been made to find new outlets for whey, and to reduce environmental pollution (Carloet al., 1999; Karand Misra, 2000). Whey is a strong pollutant when discharged into streams, its high organic matter content leading to a high BOD (Webb, 1998). Much has been done to concentrate or modify whey into another form, but such efforts have been inadequate, especially in developing countries, and utilization of whey is perhaps the most serious problem facing the dairy industry worldwide (Kosikowski, 2000).The bioconversion of whey is an important process from the viewpoint of human nutrition, especially for therapeutic purposes, with regard to economy, and is advantageous for reducing environment pollution (Karand, Misra, 2000).

Food products should be available to young children, which can provide a wide range of naturally occurring essential nutrients. When this is not possible, enrichment and fortification of foods becomes an option (World Food Programme (WFP), 1991). Close to 20 years ago, efforts were begun to develop, produce, and distribute protein-rich foods to alleviate protein malnutrition in developing countries (Beaton, 1995). The high price of proprietary fortified and enriched weaning foods and of vegetable and animal proteins, and the non-availability of low-priced nutritious foods, combined with faulty feeding practices and late introduction of supplementary foods, are mostly responsible for aggravating protein malnutrition among children (Wondimu and Nagappa, 2004). Enriched foods consist of a mixture of cereals, pulses, added fats, added vitamins, and added minerals, intended to provide a balanced intake of essential nutrients for vulnerable groups (Beaton,1995; Combs.et al., 1994).

Malnutrition among infants and young children is common in developing countries (Wondimu and Nagappa, 2004). Protein-energy malnutrition generally occurs during the crucial transitional phase when children are weaned from liquid to semi-solid or fully adult foods. During this period, because of their rapid growth, children need nutritionally balanced, calorie-dense supplementary foods in addition to mother’s milk (WFP, 1991; Wondimu and Nagappa,. 2004). Consequently, the World Food Program (WFP) has supported the manufacture of locally designed and produced fortified blended foods, including

Unimix in Kenya, Famix in Ethiopia, LikinPhali in Malawi, and Indiamix in India (WFP, 1991).

Anti nutrients are some of the undesirable components in beans that could limit utilization of their protein and carbohydrate content. Inactivation or removal of undesirable components is essential in improving the nutritional quality and organoleptic acceptability of beans, and in turn helps the effective utilization of their potential as human food (Shimelis and Rakshit, 2005). Different processing methods such as boiling, hydration and germination have been used to increase the utilization of kidney beans. Germination has often been proposed as a means by which the nutritional quality of bean seeds might be improved (Shimelis and Rakshit, 2005).

A report of a widely representative regional committee in the United States of America recently identified the researchable problem of whey processing and utilization as having the greatest urgency for solution among all the current problems in dairy technology and production (Kosikowski,. 2000). Hitherto in Ethiopia, dairy industries have sold whey as animal feed or discharged it to the sewerage system, and consequently have caused ecological disturbance through the pollution of water bodies. Furthermore, there is no utilization of whey in the forms of value-added products. The main aim of the study was to investigate the utilization of whey as powder enriched with haricot bean, *Roba 1* variety.

MATERIALS AND METHODS

The raw materials used for the study were whey and haricot bean (*Phaseolus vulgaris* L.), variety *Roba 1*. Whey was collected from Lame Dairy PLC (Sholla milk enterprise) and haricot bean from Ethiopian Agricultural Research Organization, Awash Melkasa Agricultural research centre. The experimental work was carried out in the laboratories of Ethiopian Health and Nutrition Research Institute (EHNRI), Department of Chemical Engineering of Addis Ababa University, Kotebe teachers' training college and Lame Dairy PLC.

Whey Powder Preparation

The collected whey was filtered through an ultra-filtration membrane (Armfield FT-18 ultra filtration system micro-240 model; U.K. 2007); the permeate was stored in refrigerator at 5 °C, then pre-heated at 55 °C for 40 min. A vacuum pump (IP 54, LBi 07; FDR Germany 2007) was used to concentrate the soluble solid to 60%, and it was cooled to 5 °C. Finally, it was dried by means of a pressure-nozzle spray dryer (Tall Form spray drier FT80/81; FDR Germany 2006).

Haricot Bean Powder Preparation

The *Roba 1* bean seeds were weighed into large plastic pan and soaked in distilled water (bean:water ratio of 1:5 w/v) for 12 h at 25 °C (Shimelis and Rakshit, 2005). The pan was covered with clean muslin and kept in darkness at 25 °C for four days to allow the seeds to sprout. Seeds that sprouted within four days were dried on a tray dryer (Tauro-Padova, B/05EC, No. 11; U.K. 2006). The dried seeds were then ground in a mill (Retsch Gmbh, 5657 HAAN, Type S41; FDR Germany 2003) to pass a 150 µm sieve, and stored at 4 °C until analyzed.

Mixing Ratio

The whey and haricot bean powders were mixed in various ratios and analyzed for physico-chemical and bacteriological quality. The mixing ratio is shown in Table 1.

Table 1. Mix ratio of whey and haricot bean powder

Whey powder (%)	Roba-1 haricot bean (%)	Sample code
100	_	S_1
95	5	S_2
90	10	S_3
85	15	S_4
80	20	S_5
_	100	S_6
Fresh liquid whey		$\mathbf{S_7}$

Methods of Analysis

Physicochemical and Proximate Analysis

The raw materials and blends were subjected to physicochemical and bacteriological analysis. The pH of samples was measured by digital pH meter. The meter was standardized using buffer solutions of pH 4, 9 and 10. Specific gravity was measured by lactometer and viscosity by viscometer. The moisture, ash, crude fibre and fat contents were determined according to AOAC (2003), and total protein by the semi-micro Kjeldahl method, using a conversion factor of 5.78. Carbohydrate was measured as the percentage difference from 100%. COD was measured on a multi-parameter bench photometer and BOD_5 by BOD reader. Metals were determined by means of a spectrophotometer (JENWAY model 6305UV/Visible range spectrophotometer, UK 2006).

Bacteriological Analysis

Bacteriological analyses for *E.coli* (faecal and total coliform), aerobic plate count, molds and yeasts, *Salmonella*, *Shigella*, *Clostridium botulinum*, *Bacillus cereus*, *Staphylococcus aureus*, were conducted by the methods set out in the Federal Drug Administration or Bacteriological Analytical Manual (2006). All tests were conducted at Ethiopian Health and Nutrition Research Institute laboratory.

Experimental Design and Data Analysis

Laboratory experiments were carried out using whey, haricot bean and the new products in duplicate, under a completely randomised design (CRD). Statistical analyses for the results of proximate, bacteriological and mineral analyses were performed with SPSS (Statistical Package for Social Sciences) version 16.0. For the analysis of variance (ANOVA) procedure, significance was set at the 5% level. Each value was the mean ± SD of duplicate samples.

RESULTS AND DISCUSSION

Fresh liquid whey had a mean density of 1.02 g cm^{-3} density at 20 °C, 0.985 mpas of viscosity at 15 °C, 72,514 g ml^{-1} of COD and a BOD_5 of 48,538 g ml^{-1} (Table 2). The density and viscosity were in the normal range for fresh whey as indicated by Holsinger (2003), but COD and BOD_5 were higher than recommended According to Kailasapathy and Chin, 2000, the recommended values for fresh whey are 46,000 g ml^{-1} for COD and 40,000 g ml^{-1} for BOD_5, respectively. (Kosikowski, 2000) indicated that the greater the deviation of the value above the guidelines, the greater is the increase in pollution load on a water body.

Table 2. proximate composition of whey powder, haricot bean powder, enriched products and raw liquid whey

Samples	Moisture (%)	Protein (%)	Fat (%)	Crude fibre (%)	Ash (%)	Total Carbo-hydrates (%)
S1	11.25±0.35	7.99±0.04	1.12±0.06	1.63±0.37	17.23±0.24	60.87
S2	10.90±0.02	9.16±0.07	1.53±0.11	1.41±0.09	15.49±0.23	61.57
S3	10.60±0.54	10.13±0.36	1.83±0.16	1.64±0.16	13.63±0.24	62.17
S4	10.46±0.38	13.19±0.08	1.92±0.05	1.68±0.06	12.69±0.31	60.77
S5	10.22±0.21	13.94±0.05	1.95±0.08	1.72±0.01	6.73±0.21	65.48
S6	9.70 ±0.18	23.29±0.09	2.04±0.04	2.75±0.04	6.12±0.13	56.09
S7	93.020.13	0.75±00.24	0.1±0.21	0.32±0.02	0.55±0.12	5.26

N.B. All values are means ± SD

The moisture content of whey powder (S_1) was 11.25%, which is higher than the US standard for dried whey powder 5% (US standard for dry whey, 2000). This might be due to the hygroscopic nature of whey powder (Gharsallaoui et al., 2007). Ordinary whey powder is hygroscopic and will therefore form cakes. The type of whey and local climatic conditions influence hygroscopicity and caking. Hygroscopicity, caking, and all the problems associated with the stickiness of ordinary whey powder are mainly due to lactose being present in an amorphous glassy state (Gharsallaoui et al., 2007). In the spray-drying of milk products, lactose is in an amorphous state and is not stable in atmospheric air or at normal humidity. The only form of lactose that is stable to humidity is α-lactose monohydrate. Since the lactose content of whey powder comprises more than 70% of the total solids in comparison with 30% in whole milk, the problem of the lactose content in whey powder is more severe in higher humidity (Sudika, 2004).

The moisture content of the other products decreased as the haricot bean content in the mixture increased, *i.e.* from S_2 to $S_{5.}$ This might be due to the non-hygroscopic nature of the haricot bean powder component in the mixture. Monitoring of the moisture content of fortified commodities is important for assessing the potential for loss of micronutrients. A low moisture content of food retards losses of micronutrients, spoilage from moulds and insects, and adverse changes in flavor and color (WFP, 1991). Calculations of the moisture content of fortified products indicate that fortified, blended foods make an important contribution towards meeting the recommended nutrient content. Information on actual food intakes is limited, however, and it is recognized that fortified foods targeted for consumption by various vulnerable groups are shared among other family members or may be traded (Hoppe et al. 2006).

A WFP report recommends that the moisture content of cereal-based fortified foods should not exceed 10% (WFP, 1991). Studies have indicated that low moisture content can be achieved, and it has been recommended to maintain the moisture content of blended foods at 10% or less (Combs et al., 1994). The specifications set up by the European Commission for cereal-based weaning food require that the moisture content be governed by good manufacturing practices, to ensure minimum nutritive loss and to prevent the multiplication of microorganisms (Official Journal of the European Communities, 1991).

Proteins represent the main component of haricot bean and other pulses. This makes them preferable for the fortification of foods which have low protein content (WFP, 1991). The protein content of whey powder was 7.9%, which is less than the minimum value recommended by codex standard and the US standard for dried whey powder (10% and 9.5 %, respectively) (WHO 2003, US standard, 2000). As table 3 shows, the protein content of fortified whey powder was increased by fortifying it with various proportions of haricot bean. The increase was from 7.99% for the whey powder alone, to 13.9% in the fortified product. This increase was due to the high protein content of haricot bean, as described by other researchers (Dawit and Demelash., 2003).

The fat content of the whey powder was 1.12%, which is below the minimum recommended value of FAO and US (2% and 1.5%, respectively) (WHO 2003; US standard, 2000). Beaton (2005) recommended that fortification of this product with the high fat contents of cereals or pulses is necessary. The fat content of the fortified product increased from 1.12% in the whey powder alone, to 1.95% in S_5 This indicate that, as the amount of haricot bean in the mixture increased, the amount of fat in the product also increased. Some researchers have suggested that fats may be added (no more than 10 g per 100 g of product) to increase energy density and palatability. High-fat-content pulses, vegetable oils and fats containing polyunsaturated fatty acids are preferred, but especially for vegetable oils and fats containing polyunsaturated fatty acids are preferred, where advanced packaging technology is available, otherwise they may reduce the shelf life because of oxidative changes (rancidity). Therefore, in many situations it is more advisable to add fat to the supplementary food at the time of feeding (Hofvander, 2005).

The powdered whey had a mean value of 17.23% ash, which is above the standard recommended by Codex Alimentarius (9.5%) (WHO, 2003). The ash content of the fortified product was reduced as the proportion of haricot bean in the mix was increased, *i.e.* 15.49% in S_1 to 6.12 % in S_5. This might be due to the lower ash content of haricot bean, which is in line with the results of Dawit and Demelash (2003).

Because dietary fiber are slowly absorbed and fermented by intestinal flora, thus causing a laxative effect, the crude fiber content of the supplementary food should not exceed 2% (WFP, 1991). The mean value of the crude fiber content of all mixed powder ranged from 1.41 % in S_2 to 1.72 in S_5, was in the recommended guideline range of the codex standard (WHO, 2003).

Table 3. Mineral contents of whey, haricot bean powder and blended product

Samples	**Mineral content (mg/100g) on dry basis**					
	Mn	**Zn**	**Fe**	**Ca**	**Mg**	**K**
S_1	0.08±0.02	1.31±0.01	1.41±0.01	33.93±0.01	3.64±0.02	15.13±0.06
S_2	0.10±0.02	1.22±0.01	2.12±0.01	30.35±0.02	3.62±0.01	18.05±0.01
S_3	0.13±0.01	1.10±0.02	2.36±0.01	33.46±0.13	3.60±0.01	18.31±0.06
S_4	0.22±0.01	1.12±0.02	2.75±0.01	32.36±0.01	3.43±0.01	19.15±0.02
S_5	0.21±0.01	1.93±0.02	3.02±0.01	24.34±0.02	3.45±0.04	19.33±0.04
S_6	0.30±0.01	3.23±0.08	7.58±0.02	15.83±0.01	3.51±0.01	20.21±0.08

As may be seen from Table 3, the haricot bean powder had a higher content of Mn, Zn, Fe and K, the values of which were 0.30, 3.23, 7.58 and 20.21 mg/100 gm, respectively, compared with the whey powder, while the Mg content of haricot bean was less than that of whey powder.

With regards to the Fe content of the mixed products, *i.e.* from S_2, 2.12 to S_5, 3.02, it was increased. This increase is a result of the high Fe content of the haricot bean, as indicated by Dawit and Demelash (2003). According to the Codex standard for infant formulas (codex stan.72-1981), the Ca, Mg and K content of the enriched foods of sample $\mathbf{S_5}$ was within the recommended value (50, 5 and 80 mg/100 gm, respectively), while Mn, Zn and Fe were above the recommended value (5 µg, 0.5 mg and 1 mg per 100 gm, respectively).

Table 4 Microbial analysis of whey powder enriched products and liquid whey

Parameters	Result							Acceptable limit (cfu/ml)
	$S_{1(cfu/ml)}$	$S_{2(cfu/ml)}$	$S_{3(cfu/ml)}$	$S_{4(cfu/ml)}$	$S_{5(cfu/ml)}$	$S_{6(cfu/ml)}$	$S_{7(cfu/ml)}$	
Moulds and yeasts	$<1\times10^1$	$<1\times10^1$	$<1\times10^1$	$<1\times10^1$	$<1\times10^1$	$<1\times10^1$	$<1\times10^1$	< 50
APC [a]	1.2×10^3	1.4×10^2	2.4×10^4	1.5×10^6	2.3×10^7	4.8×10^7	1.48×10^4	$< 2\times10^4$
Fecal coliform	Nil	Nil	Nil	Nil	Nil	Nil	Nil	< 10
E. coli type 1	Nil	Nil	Nil	Nil	Nil	Nil	Nil	< 40
Staphylococcus aureus	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Shall not be detected
B. cereus	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Shall not be detected
Streptococcus species	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Shall not be detected
Salmonella sp	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Shall not be detected
Shigella spp	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Shall not be detected

[a] APC Aerobic bacteria plate count at 37 °C for 48 h

N.B. All values are means ± SD

Microbial Data Analysis

The aerobic bacteria or total plate count of the fortified product indicates that there was a deviation from the acceptable limits as the proportion of haricot bean powder increased relative to the whey powder (S_1).This may have been due to unhygienic preparation, especially during the milling of the haricot bean. The results for molds and yeasts were within acceptable limits for all fortified products, as recommended by the US Standard for dry whey powder and National Standard of the People's Republic of China, Hygienic Standard of Whey Powder (50 and 40 cfu ml^{-1}, respectively) (United States Standards for Dry Whey, 2000; Ministry of Health of the People's Republic of China, 1999). Other parameters, *i.e. Salmonella, Shigella, Staphylococcus* species, *Bacillus cereus*, fecal coliform and *E. coli*, were nil,

which is in agreement with the standards recommended by WFP, US and China for dried whey powder (WFP, 1991; United States Standards for Dry Whey; Ministry of Health of the People's Republic of China, 1999).

CONCLUSIONS

The traditional technology mostly applied by Ethiopians, such as germination of cereals, legumes and pulses, could be used for increasing the nutritional value and palatability of food products, and as is indicated by most researchers, by reducing the anti nutritional factors which are present in them, is easily adapted to the development of low-cost weaning and supplementary foods from pulses. This study clearly indicates that there is a possibility of enriching whey with the high-protein-content haricot bean.

The results of this study also showed that the nutritional content of a blend ratio of whey: haricot bean powder of 80:20 is the best composition for fulfilling the nutritional requirements of children, especially in terms of protein and fat. Therefore, the utilization of whey as a value-added product is crucial from a nutritional and medicinal point of view. The advantage of blending whey powder with haricot bean, as compared with other cereals and pulses, lies in its economy, since haricot bean is locally produced in sufficient quantity, and contains a significant amount of protein and fat. Utilization of whey for various value-added products in the Ethiopian dairy industry can contribute to food security via strong marketing systems and advertising programs closely linked to the good health of consumers.

REFERENCES

Association of Official Analytical Chemists (AOAC), 2003. Official methods of analysis Vol. II 17th edition) of AOAC International, Washington, DC, USA

Beaton H. 1995. Fortification of Foods for Refugee Feeding. Final Report to the Canadian International Development Agency and Technical Background Report: Derivations and Analysis. Willowdale, Ontario

Carlo M., Giorgio L., Alessandra D. Anjella I., Gian M. R., Raffaello P., 1999 High-level production of heterologous protein by engineered yeasts grown in cottage cheese whey *Applied and Environmental Microbiology*, 65 (6) 2745-7

Combs G., Dexter P., Horton S. 1994. Micronutrient Fortification and Enrichment of PL 480 Title II Commodities: Recommendations for Improvement. Technical Review Paper.

Dawit A., Demelash S., 2003 Haricot bean marketing and export performance, constraints and opportunities, research paper number 54, EARO, Addis Ababa.

European Commission 1991. Fifth Amendment to the Commission communication on the characteristics of products to be supplied as food aid. Official Journal of the European Communities. No C 114. Food and Drugs Administration / Bacteriological Analytical Manual.

Hofvander Y., 2005. Processed supplementary foods for older infants and young children, with special reference to developing countries University of Uppsala, Uppsala, Sweden. http://archive.unu.edu/unupress/food/8F091e/8F091E01.htm , accessed date Oct.2009

Gharsallaoui A., Roudaut G, Chambin O, Voilley A., Saurel R. 2007. Applications of spray-drying in microencapsulation of food ingredients: An overview. *Food Research International* 40 (9):1107–1121

Holsinger V.H., Posati L.P., DeVilbiss 2003 Whey Beverages: A Review, *International dairy journal*. 23:46-56

Hoppe C., Gregers A., Stine J., Christian M., Henrik F, Per S., Kim M. 2006. The Use of Whey or Skimmed Milk Powder in Fortified Blended Foods for Vulnerable Groups, *J Dairy Sci* 81(3), 597-608.

Kailasapathy K., Chin J., 2000. Survival and therapeutic potential of probiotic organisms with reference to *lactobacillus acidophilus* and *bifidobacterium* species, *Immunology and cell biology*, volume78, 80-88.

Kar T., Misra A. K. 2000, Enhancement and stabilization of diacetyl produced by cultures grown in whey. *Annals of Microbiology* 50 (1) 33-41

Kosikowski F.V. 2000 Whey utilization and whey products, *J. of Dairy Sci.* 62, (7):1149–1160

Ministry of Health of the People's Republic of China, 1999. National Standard of the People's Republic of China, Hygienic Standard of Whey Powder, Beijing. China

Omar A., Ara D., Adrian C., 2007. Investigation on whey proteins profile of commercially available milk-based probiotics health drinks using fast protein liquid chromatography. *British Food Journal* 109 (6): 469-480

Shimelis E. A., Rakshit S.K 2005. Anti nutritional factors and *in vitro* protein digestibility of improved haricot bean (*Phaseolus vulgaris* L.) varieties grown in Ethiopia, *International*

Journal of Food Science and Nutrition, 56(6): 377-/387.

Sudika K. 2004 Whey utilization and whey products. *International Dairy Journal.* 11**,** pp. 1–17.

United States Standards for Dry Whey14, 2000. United States Department of Agriculture, Washington, D.C. 20250-0230. http://www.ams.usda.gov/AMSv1.0/getfile?dDocName=STELDEV3004469, accessed date,Oct.2009

Webb B.H. 1998. The utilization of whey: A review, Dairy research laboratories, Bureau of Dairy industry, Agricultural research administration, U.S. Department of Agriculture, USA.

WFP 1991 Guidelines for calculating Food Rations for refugees, World Food Program, Rome

WHO 2003 Codex standard for whey powders codex stan a-15-1995, Rev. 1-2003

Wondimu A., Nagappa M. 2004 Development of weaning foods based on malted, popped, and roller-dried barley and chickpea, *J. African Crop Sci.*19: 29-32.

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