Food Science and Quality Management
ISSN 2224-6088 (Paper) ISSN 2225-0557 (Online)
Vol.39, 2015

Development of a Bovine Blood Enriched Porridge Flour for Alleviation of Anaemia among Young Children in Kenya

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The research is funded by the Kenya National Council for Science and Technology and University of Nairobi, Dean's Committee.

Abstract

The prevalence of iron deficiency and anaemia among young children in Kenya is high. This is because the main diet is porridge prepared from cereal flours which have very low iron levels and poor bioavailability. This study was designed to investigate the effect of enriching porridge flour mixes prepared from cereals and cassava with iron from bovine blood meal, for use in an intervention for anaemic children. The mixes were formulated to provide approximately 50% of the recommended daily allowance of iron for children.The mix was tested for nutritional, sensory and storage properties. The most acceptable mix contained sorghum, finger millet, cassava, and blood powder in the ratios of 6:14:10:0.5 providing 54% of the recommended daily intake of iron for 1-3 year old children, when consumed per day as porridge containing 30.5g flour. The mix could be stored at 35ºC for up to three months without adverse changes in microbiological quality. The study concludes that acceptable and shelf-stable porridge mix can be formulated from local cereals and cassava flour with iron enrichment from bovine blood meal.

Keywords: Bovine blood, Composite flour, Anaemia, Young children, Kenya

1. Introduction

Iron deficiency with its attendant anaemia is the most prevalent micronutrient disorder worldwide and is considered to be among the top ten contributors to the global burden of disease (UNICEF/UNU/WHO, 2001; Maclaren, 2008). In Kenya, a national micronutrient survey indicated that iron deficiency is widespread and is higher among children under five years with an anaemia prevalence rate of 60%. In malaria endemic areas, anaemia has the highest prevalence and is rated among the most serious health problems after malaria (Mwaniki *et al*, 2001; KDDP, 2002).

The main causes of anaemia include inadequate dietary intake and low bioavailability of iron (Lung'aho and Glahn, 2009). Interventions to reduce Iron Deficiency Anaemia (IDA) have thus mainly focused on food supplementation, industrial food fortification and dietary diversification; a term often used interchangeably with food to food fortification (IFPRI, 2001). However, iron supplementation in the malaria endemic regions of developing countries is limited, due to its potential negative side-effects (Schumann and Christ, 2007). Food fortification, which has contributed to significant reduction of IDA in developing countries, poses major challenges due to technical, operational, industrial financial constraints and requires significant planning, management, monitoring and government support to succeed (Imungi, 2006). The available fortified foods are usually unaffordable by most vulnerable groups. Food based approaches offer alternative means of improving iron intake; however the bioavailability of iron from plant foods, which form the bulk of the diet in developing countries, is low, and animal sources of food which are rich in highly bioavailable haem-iron are often unaffordable by the poor families of vulnerable children (Allen, 2003).

Blood, obtained during slaughter of domestic animals is consumed as food by many communities all over the World in traditional sausages, soups and as a thickener for sauces. In Africa many pastoral communities, such as the Maasai of Kenya and Tanzania, consume fresh bovine blood regularly ((Sehmi, 1993; Ferraro, 2001). Most communities in Kenya traditionally prepared and consumed bovine blood in various forms, though documented literature is scanty.

Research at the Voronezh Technological Academy in Russia, showed that bovine blood is rich in iron and proteins, and that the proteins are assimilated by the human body twice as fast as chicken-egg proteins. Technologies for manufacturing milk, yoghurt, chocolate, and coffee simulates from bovine blood were developed and acceptability studies indicated that the products tasted the same as the ones prepared using orthodox methods (Mosnews, 2007). Standards for safe collection and spray drying of bovine blood have been issued as well as the safety and challenges of bovine blood as human food (NASDBPPA, 2012; Ofori and Hsieh, 2011). In Chile, the National School Breakfast and Lunch programme introduced Bovine Haemoglobin Concentrate (HBC) fortified cookies to reduce iron deficiency among school children (Walter *et al*, 2001). The lack of acceptance of blood in food products among some cultures and religions must be respected and products containing blood must be declared. The Kenya national micronutrient survey of 1999 showed that porridge made from sorghum, finger millet, cassava and maize are commonly consumed by children. This study was therefore designed to develop porridge flour enriched with iron from bovine blood, for use in alleviating iron deficiency in children.

2. **Materials and methods**

2.1 Composite flour ingredients and preparation

The composite flours were formulated from flours made from sorghum (*Sorghum bicolor*), finger millet (*Eleusine coracana*), cassava *(Manihot esculenta)*, maize (*Zea mays*) and bovine blood based on the findings of the 1999 micronutrient survey, on flours commonly used for making porridge for young children (Mwaniki *et al*, 2001).

i. Sorghum, millet and maize flour preparation

Sorghum, millet and maize grains were purchased from an open air market in Nairobi. The grains were sorted to remove extraneous matter, washed with cold tap water and dried in hot sunshine for six hours. The grains were then milled into a fine flour to pass through 150µ sieve (Fritsch Analysette 3.502) using a hammer mill (DFH48, No. 282521/UPM 6000; Glen Creston Ltd, London, England). The milled flour was stored at 20-25°C, in food grade plastic containers.

ii. Cassava flour preparation

Freshly harvested cassava roots from the University of Nairobi farm were washed, peeled, grated and dried in a tent solar drier at approximately 55-58°C. The dried cassava flakes were milled to fine flour to pass through150µ sieve which was of similar particle size as the other cereal flours. The milled cassava was stored in similar containers and temperatures as the other cereal flours.

iii. Blood powder preparation

Prior to blood tapping, the animals were certified as healthy through pre-slaughter inspection, at the University of Nairobi's veterinary farm and evaluated for haematological status.

Two animals with normal haematological status were selected for blood collection. The neck area from where the blood was collected was shaved using a blade and disinfected using 70% alcohol. The containers and equipment used for blood collection were cleaned using food grade washing detergent and sanitized with hot water. During slaughter, fresh blood was collected directly from the jugular vein.

The fresh blood was transported to the Department of Food Science, Nutrition and Technology laboratory within 30 minutes and allowed to stand for about 15 minutes to facilitate separation of serum. The serum was drained off and the coagulum cut into cubes of approximately 2cm cubes using a kitchen knife then cooked at 92-93°C for 40 minutes. The cooked cakes were broken into smaller pieces with a wooden spoon, ground to coarse particles in a coffee grinder (Sinbo SCM 2923 P.R.C.), and dried overnight in a hot air oven at 55-58°C. The dried blood was milled to a fine powder to pass through 150µ sieve and stored in sealed zip-lock polythene paper bags, prior to further analysis.

b. Determination of hydrogen cyanide, proximate composition and mineral content

The sorghum millet maize and cassava flours and the blood powder were analysed for proximate composition and minerals. The cassava flour was analysed for Hydrogen cyanide. Proximate analyses were performed in duplicate, while mineral analyses were performed on triplicate samples. The results obtained from the analyses were used to formulate the composite flours.

2.2.1 Hydrogen cyanide

The Hydrogen Cyanide (HCN) content was analysed according to the Association of Analytical Chemists (AOAC, 1984). Approximately 20g of cassava flour were mixed with 200 ml distilled water in a distillation flask. The mixture was allowed to stand for at least two hours and steam distilled. Approximately 200ml of the distillate was collected in a volumetric flask containing 25ml of 2.5% sodium hydroxide solution. An aliquot of 8ml of 5% potassium iodide solution was added to 100ml of distillate and titrated against 0.02N silver nitrate solution until a faint but permanent turbidity was obtained. Hydrogen Cyanide content was calculated as: 1ml of 0.02N silver nitrate, being equivalent to 1.08 mg of HCN per 20g.

2.2.2 Proximate composition

Crude protein, crude lipid, crude fibre and total ash were analysed using AOAC methods (AOAC, 1980). Soluble carbohydrates were calculated as a difference. The moisture content of cereal flours and bovine blood powder were determined by drying at 105^0C in an air oven to constant weight. Soluble total energy was calculated from fat, carbohydrate and protein contents using Atwater's conversion factors. To determine Free Fatty Acids (FFA) 2g of the sample was dissolved in a mixture of diethyl ether and alcohol and titrated with 0.1M sodium hydroxide. The FFA figure was calculated as oleic acid (1ml 0.1M sodium hydroxide≡ 0.0282g oleic acid).

2.2.3 Mineral analysis

The specific minerals iron, calcium, potassium, zinc and selenium contents of the flours were analysed using x-ray fluorescence technique. The samples were first digested with nitric acid and perchloric acid, and the digests used for the analysis of the minerals using the bench top Total Reflection X-ray Fluorescence technique (ED-XRF model SL80175, S/No. 0701983. USA).

2.3 *Formulation of the composite flours*

Five composite flours, aimed at providing approximately 50% Recommended Daily Allowance (RDA) of iron for children aged 1 to 5 years, were formulated using Nutrisurvey linear programming software. In preparing the formulations it was assumed that 1 cup of porridge, equivalent to 250mls of porridge, is prepared from about 25g-30g of flour and that a child will take 1cup per day. The contribution of the composite flours to RDA of children for specific nutrients was also calculated using Nutrisurvey.

2.4 Porridge Preparation and Sensory evaluation

Porridge was prepared from each of the 5 different composite flours and subjected to sensory evaluation. To prepare the porridge 300g of each of the composite flours were mixed with 5.5 litres of water to form slurry. The slurry was heated while continuously stirring with a wooden spoon until it boiled. The heat was lowered and the porridge left to simmer for 20-25 minutes. The cooked porridge was then moderately cooled and served in styrofoam cups to 24 panellists who were familiar with quality characteristics of porridge. The panellists were provided with coded porridge samples

The 5 porridges were evaluated for colour, taste, mouth-feel and overall acceptability using a seven point hedonic rating scale, with 1= dislike very much, and 7=like very much. The panellists were asked to rinse their mouths with clean water before tasting the next sample. The flour that produced the most acceptable porridge was subjected to microbiological quality assessment and shelf-life evaluation.

2.5 *Microbiological analysis*

The microbiological analyses were done for the most preferred composite flour. The tests done were total viable count using plate count agar, total coliform count using violet red bile agar and yeasts and moulds counts using potato dextrose agar according to standard microbial techniques (AOAC, 1980).

2.6 Shelf life evaluation

The most preferred composite flour was subjected to shelf life evaluation. The composite flour was divided into 25 portions of 200g each. Six of these portions were packaged in 6 different gunny bags, plastic bottles, kraft paper and polythene bags. A set of three of the different packaging materials were stored in 2 differently thermostatically controlled air ovens set at temperatures of 25°C and 35°C for a period of three months. Initial microbial quality was determined at the beginning of the shelf life study. Thereafter a sample in the different packaging materials, stored at different temperatures, was removed and analysed after every month for changes in microbial quality according AOAC method (AOAC 1980. Shelf evaluation was also done by accelerated methods (one day equivalent to one month). Sixteen (16) samples packaged in 4 different packaging materials were placed in an air oven at 55°C and one set of the different packaging materials removed and analysed for moisture content and free fatty acids content at the beginning of the shelf life study and after every 24 hours for 3 days.

2.7 Statistical analysis

Data were analysed using Statistical Package for Social Sciences (SPSS) for windows, version 16. One-way analysis of Variance (ANOVA) was done for proximate composition, minerals, free fatty acids, moisture content and sensory evaluation data. Then the means were tested for significant differences at P ≤0.05. Microbiological counts were transformed into log number and analysis of variance (ANOVA) performed on log count.

3.0 Results and Discussion

3.1Hydrogen cyanide content of cassava flour

The hydrogen cyanide content of the cassava flour was found to be 8.1±0.3mg/100g,which is within the acceptable recommended level of less than10mg/100g for human consumption (KBS, 2003).

3.2 Proximate composition of the flour ingredients

The results of proximate composition and energy analyses of the composite flour ingredients are shown in Table 1. The moisture content ranged from 7.0% in cassava and millet flours to 12.3% in maize flour. These moisture contents were below the 13% mc, recommended for optimum storage of cereal flours (KBS, 2003). Bovine blood powder had a moisture content of 8.5% which is comparable to that determined by Kikafunda and Sserumaga of 6.85±1.51 (Kikafunda and Sserumaga, 2005). The bovine blood powder had a higher protein content at 80g/100DM compared to cereal and cassava flours whose protein content ranged from 8.5g/100DM (in maize) to11.81g/100DM (in millet).

Table 1: Proximate composition of sorghum, cassava, millet and blood powder used to formulate the composite flour (g/ 100g DM).

Ingredient	Moisture (%)	Protein	Lipid	Crude Fibre	Total Ash	Soluble Carbohydrates	Energy (kcal)
Sorghum flour	7.2±0.2[a]	8.14±0.1[a]	4.67±0.3[d]	2.37±0.0[c]	1.35±0.01[a]	77.80	379
Millet flour	7.0±0.1[a]	11.81±0.6[b]	3.54±0.2[c]	2.85±0.0[d]	5.22±0.13[c]	72.96	358
Cassava flour	7.0±0.3[a]	8.97±0.3[a]	1.13±0.1[a]	1.5±0.0[b]	2.36±0.04[b]	80.22	366
Maize flour	12.3±0.1[c]	8.5±0.2 [a]	3.8±0.1[c]	9.2±0.4 [e]	2.2±0.01 [b]	64.7	331
Blood powder	8.5±0.0[b]	80.1.0±0.0[c]	2.58±0.3[b]	0.25±0.0[a]	3.98±0.01[d]	4.64	362

Means in the same column with different superscripts are significantly different (P ≤ 0.05).

The protein content of the bovine blood powder was also higher than the protein content of bovine liver (17mg/100g), the food source normally recommended for use in dietary protein supplementation. Other studies have also reported that bovine blood powder is a rich source of protein (Allen, 2003; Mosnews, 2007; Kikafunda and Sserumaga, 2005). The protein content of the millet flour at 11.8g/100DM was significantly higher than that of the maize, sorghum and cassava flours; however the protein content of maize, sorghum and cassava flours were not significantly different.

The lipid and crude fibre contents of composite flour ingredients ranged from 4.7% in sorghum flour to 1.1% in cassava flour and from 9.2% in maize flour to 0.25% in blood powder, respectively. The carbohydrate content of bovine blood (4.64g/100) was lower than the carbohydrate content in the sorghum, millet cassava and maize flours (64 - 80g/100).

The low dietary fibre content of the composite flour ingredients makes it ideal for diets of malnourished children, as it increases nutrient and energy digestibility, as opposed to high dietary fibre content which increases bulk and satiety while reducing digestibility (MIYCN working group, 2009).

3.3 Mineral contents of composite flour ingredients

The results for iron and the specific minerals:- potassium, calcium, manganese, copper, zinc and, selenium in composite flour ingredients are shown in Table 2. The iron content of the bovine blood powder at 121.0±5.6mg/100g was highly significantly higher than that of other composite flour ingredients (P = 0.00). The iron content of bovine blood powder was higher than the commonly recommended animal food sources such as liver (10mg/100g edible portion) or beef (3.79mg/100g). Similar levels of iron in bovine blood has been reported in other studies (Walter *et al*, 2001; Kikafunda and Sserumaga, 2005; Mosnews, 2007). Widely consumed plant foods such as sorghum, millet and cassava contain iron comparable to that of meat and liver. However the bioavailability of iron is limited in plant foods, making them a poor source of the mineral.

All composite flour ingredients were found to be high in potassium, which was highest in cassava flour at 485 mg per 100g and lowest in millet flour at 210 mg per 100g.This implies that the flour would also make a good supplement for hypertensive persons. Millet and cassava flours contained significantly higher calcium than the other flours (P= 0.000). This would contribute to meeting part of the daily calcium requirement for young children consuming the porridge. The composite flour ingredients contained trace levels of manganese, copper, zinc and selenium.

Table 2: Mineral content of the flour ingredients for formulation of the composite flour (mg/100g)

Ingredients	Potassium	Calcium	Manganese	Iron	Copper	Zinc	Selenium
Sorghum flour	311.0±4.8	< 20	< 5	4.17±0.09	0.52±0.0	1.63±0.1	< 5
Millet flour	207.2±4.8	102.3±14.4	8.6±0.8	9.5±0.12	0.53±0.0	0.96±0.0	< 5
Cassava flour	484.7±63.8	82.9±4.6	< 5	5.42±0.03	< 0.5	0.80±0.1	< 5
Maize flour	330±31.1	<20	<5	1.5±0.01	0.50±0.0	2.5±0.1	<5
Bovine Blood	210.0±10.5	< 20	< 5	121.0±5.6	< 5	< 5	< 5

3.4 Composite flour formulations

Ingredient proportions of the different composite flours and the achieved RDA for the children as generated from Nutrisurvey are shown in Table 3. Children in the 1-3 age categories are all able to meet 50% of their iron RDA from the five formulations, while in the 4-6 age category two of the five formulations provide slightly less than 50% of the RDA for iron. The contribution of specific nutrients in the composite flours to the RDA of children is shown in Table 4. The RDA of the specific nutrients ranged from 0–13%, which although low, contributes towards meeting part of the daily nutritional needs. This is acceptable since the children are not limited to a cup of porridge and usually consume more than one cup of porridge in the course of the day in addition to other meals and snacks .The children are thus able to obtain additional nutritional requirements (Mwaniki et al, 2001; Mbagaya, 2009).

Table 3: Composite flour formulations from the ingredients

Composite flours (1 cup)	Sorghum flour (g)	Millet flour (g)	Cassava flour (g)	Maize flour (g)	Bovine blood (g)	RDA for 1-3 years old (%)	RDA for 4-6 year old
CF 1	8	8	8	0	1	53.8	48.9
CF 2	9	9	6	0	1.5	66.4	60.3
CF 3	6	14	10	0	0.5	53.8	48.9
CF 4	8	8	0	8	1.5	59.7	54.2
CF 5	9	9	0	6	1.7	66.5	60.5

Table 4: Contribution of the flours to the RDA's of children

Composite flour	Macronutrient Content					Vitamin A and Minerals					
	Protein (g)/ %RDA	Fat (g)/ %RDA	CHO (g)/ %RDA	Dietary fibre (g)/ %RDA	Energy (kcal/ %RDA	Vitamin A (µg)/ %RDA	Ca (mg)/ %RDA	P (mg)	Fe (mg)/ %RDA		Zn (mg)/ %RDA
									1-3 yrs	4-6 yrs	
CF 1	3.1/5	0.8/1	18.5/6	0.6*	91.8/5	0.4/0	6.0/1	52.6*	2.7/54	2.7/49	0.6/11
CF 2	3.5/6	0.8/1	18.5/6	0.6*	93.7/5	0.3/0	5.8/1	58.1*	3.3/66	3.3/60	0.7/11
CF 3	3.2/5	0.9/1	23.2/8	0.7*	111.1/5	0.5/0	7.3/2	65.8*	2.7/54	2.7/49	0.7/12
CF 4	3.5/6	1.0/1	17.3/6	1.2*	90.8/4	14.8/4	4.7/1	70.0*	3.0/60	3.0/54	0.8/13
CF 5	3.7/6	1.0/1	17.5/6	1.1*	92.3/5	11.1/3	4.8/1	71.2*	3.3/67	3.3/66	0.8/13

3.5 Selection of the most preferred composite flour

The results of sensory evaluation of the porridge from the five composites are shown in Table 5. All the porridges scored more than 4 in all attributes. They were therefore all acceptable. The most acceptable porridge was from CF3. This composite flour was selected for the study, and was subjected to shelf-life evaluation. However the fact that all the porridges were acceptable implies that all the mixes could be commercialized.

Table 5: Sensory attribute mean score of the composite flours

Sample	Colour	Taste	Mouth feel	General acceptability
CF1	4.8 ± 1.3^{ab}	5.0±1.23ab	5.2±1.4ab	5.1±1.2a
CF2	4.7 ± 1.3^{ab}	4.71±1.54ab	4.8±1.5ab	4.8±1.4a
CF3	5.6 ± 1.4^{b}	5.33±1.5b	5.4±1.3b	5.5±1.3a
CF4	4.1±1.7a	4.0±1.6a	4.0±1.4a	4.5±1.2a
CF5	4.5±2.0ab	4.8±1.6ab	4.5±1.5ab	4.6±1.6a

Means in the same column with different superscripts are significantly different ($P \leq 0.05$)

There was significant difference ($p < 0.01$) in colour, mouth feel and general acceptance among all composite flours. CF 3 had the highest scores and was significantly different from the lowest, which was CF4. However CF4 was not significantly different from CF2, CF1 and CF5, which were also not significantly different from each other. The taste and mouth feel followed the same trend as the colour perception. In terms of overall acceptance, there was no significant difference among all the flours. The most preferred composite flour CF3 was highly ranked in all attributes. CF4 and CF5 had the lowest sensory acceptability, this can be attributed to the fact that the dark colour of bovine blood powder was more visible in the mixes which had maize meal flour, thus making them less acceptable compared to the cassava mix porridges. Based on the results of sensory evaluation the composite flour, CF3, was selected for further testing.

3.6 Shelf -life evaluation

The shelf-life evaluation was based on changes in moisture content, free fatty acids and microbial quality of the selected composite flour

3.6.1 Changes in moisture content of the composite flour during storage

The moisture content of bovine blood powder was 8.51%, which was comparable to an average moisture content of 6.85% reported by Kikafunda and Sserumaga (2005). The initial moisture content of the composite flours was 10.23%. There was significant reduction ($p<0.05$) in moisture content of the composite flour during the three month storage period. The reduction was significant during the first and second month of storage but not significant during the third month of storage (Figure1). However, Kaced *et al.* (1984) reported that millet meal stored at 30ºC and relative humidity of 80% showed an increase in moisture content during initial 60 hours (2.5 days) of storage and then levelled off. This difference could be attributable to high temperatures used during the accelerated storage in this study. However, there were no significant differences in reduction of moisture among the different packaging materials used.

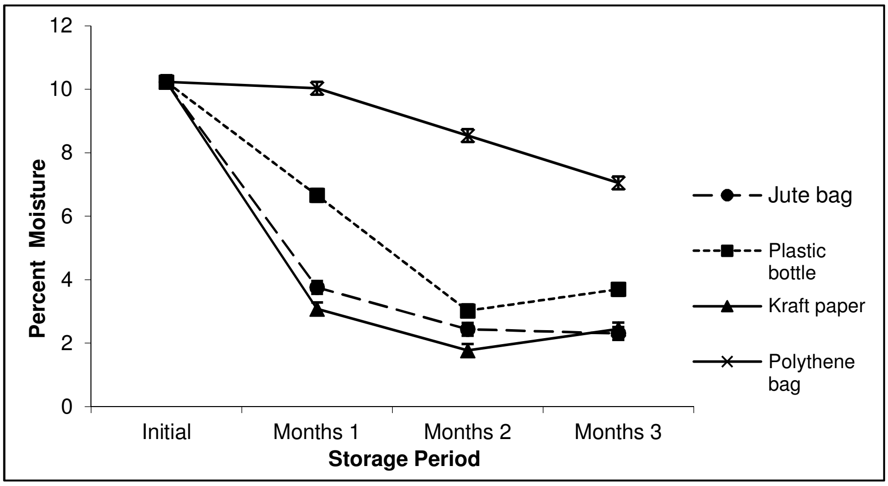


Figure 1: Moisture content of selected composite flour during three months of storage in different packaging materials

3.6.2 Changes in the free fatty acid content of the composite flour during storage

Changes in the free fatty acids content of the most preferred composite flour is shown in Figure 2. The composite flour showed a rapid increase in free fatty acid during first 2 months of storage in all packaging materials; which were a highly significant (p <0.01) however there was no change in free fatty acids during the third month of storage. The free fatty acids of composite flour increased at different rates for samples stored in different packaging materials; however the rates did not show a significant change during storage. The initial rapid increase in fat acidity (expressed as free fatty acids) in the composite flour are similar to the results obtained in a study by Kaced *et al* (1984) based on millet meal stored in polythene and cotton bags at 27ºC which also showed a rapid increase in fat acidity followed by a levelling-off phase.

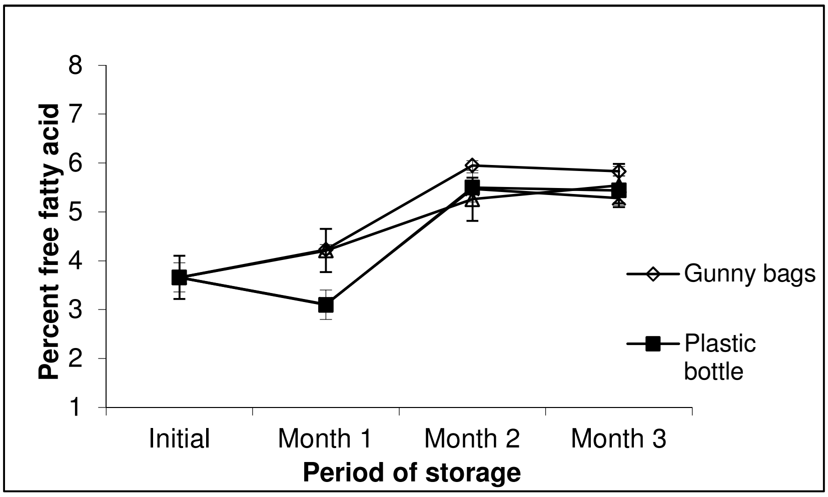


Figure 2: Free fatty acid of selected composite flour during three months of storage in different packaging materials

3.6.3 Microbiological quality

The total viable count and total coliform counts for bovine blood powder were less than 50 cfu/g. *Staphylococcus aureus*, Salmonella and *Escherichia Col*i were absent. These results are in agreement with Kikafunda and Sserumaga (2005) who similarly reported low and safe microbial counts of dried bovine blood powder.

3.6.3.1 Changes in total viable count of the composite flour

The initial total viable count, total coliform count, and yeast and mould count in the composite flour were log 6.65, 4.32 and 4.48cfu/g respectively. Despite washing and sun drying of the various flour ingredients**,** these counts were above the recommended Kenyan standards levels of log 5, 1 and 3cfu/g of composite flour for total aerobic count, coliform, and yeast and moulds respectively (KBS, 2003). This was attributed to the slow sun drying rate (2-3 days), which could allow for microbial growth. The composite flour tested negative for *Escherichia coli, Staphylococcus aureus* and Salmonella, showing that the composite flour was safe before commencement of the shelf life study.

The results for total viable counts for composite flour are shown in Figure 3. The initial total viable count in the composite flour was log 6.65 cfu/g of the flour sample. The total viable counts were above the recommended Kenyan standards level of 5 cfu/g of composite flour. There was apparent decline in total viable counts in all packaging materials during storage at 25°C and 35°C; however there was no significant difference in total viable counts between the various packaging materials during the storage period. These findings differ from the findings of Kikafunda and Sserumaga who reported a significant increase ($p<0.05$) in total viable count in bovine blood stored in polythene bags for a period of three months at room temperature.

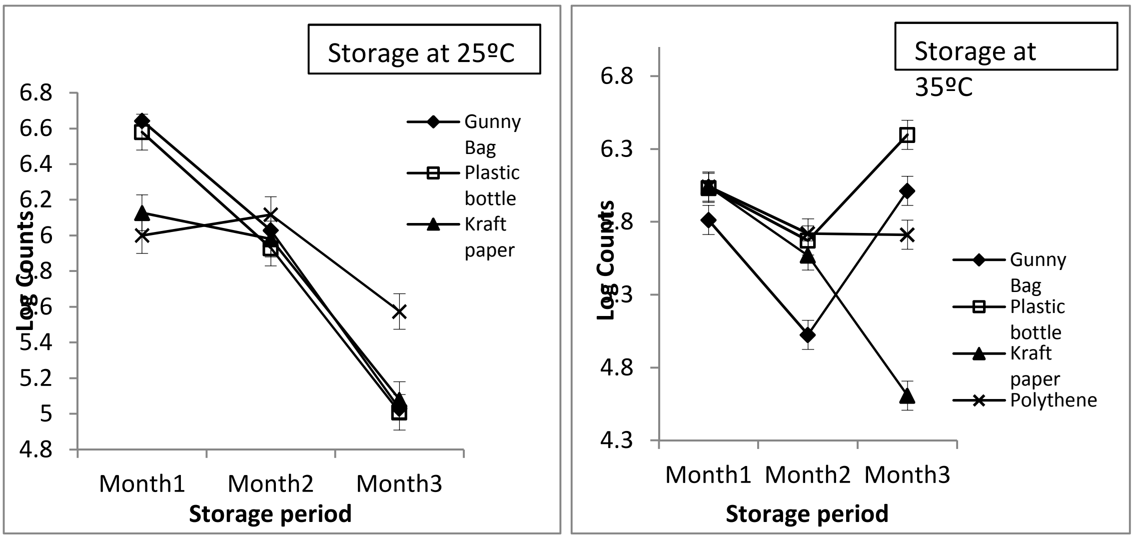
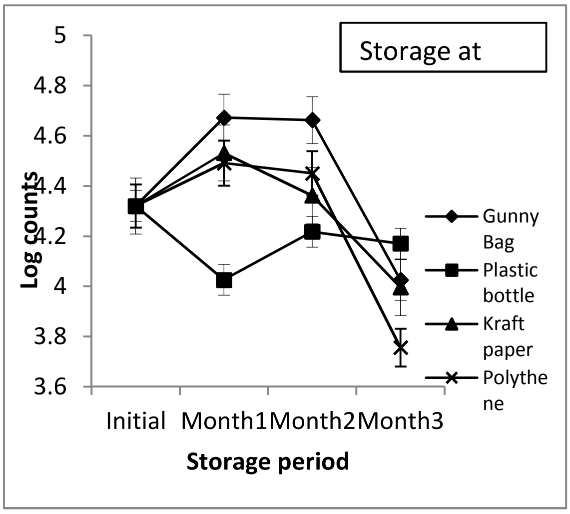


Figure 3: Total viable counts for most preferred composite flour during the three months of storage in different packaging materials

3.6.3.2 Changes in total coliform count of the composite flour

The results for total coli forms counts are shown in Figure 4. The initial total coli forms count was log 4.32 cfu/g. The total coli form counts were above the recommended Kenyan standards levels of log 1 counts per gram of composite flour (KBS, 2003). There was no significant change in total coliform count in all packaging materials during the first 2 months of storage, but the counts decreased by approximately log 0.5 cfu/g,during the third month of storage at 25°C and 35°C, which was not significant.



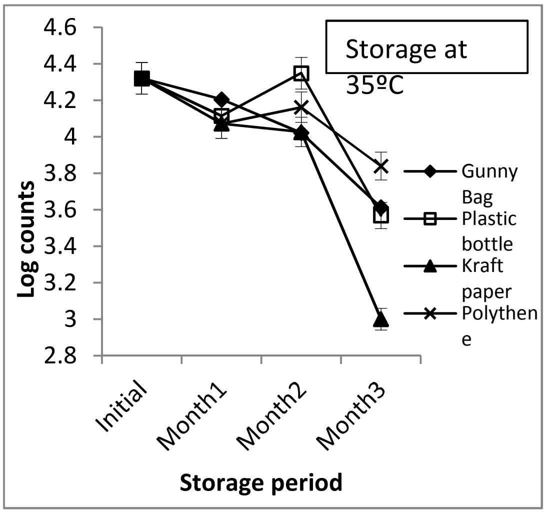


Figure 4: Total coliform counts of most preferred composite flour during three months of storage in different packaging materials

3.6.3.3 Changes in yeast and moulds in the composite flour

The results for yeast and moulds counts are shown in Figure 5. The initial yeast and mould count, in the composite flour was log 4.48cfu/g. The yeast and moulds counts were above the recommended Kenyan standards levels of log 3 cfu/g of composite flour. There was a significant difference (P<0.05) in yeast and mould counts in composite flours stored at 25°C in the various packaging material but there was no significant difference in composite flours stored at 35°C. The yeast and mould count decreased during the first month of storage at 25 and 35°C except for the plastic bottle sample which showed a slight increase that was not significant. There was an increase in yeasts and moulds count in composite flours stored in jute and polythene packaging materials during the second month of storage at 25°C, but the counts decreased during the third month of storage this could be due to deletion of air (oxygen) in the packaging materials. However there was a decrease in yeasts and moulds count in composite flours stored in gunny bags and plastic bottles packaging materials during the second month of storage at 35°C, but very slight decrease during the third month of storage at 35°C. The yeast and mould count decreased during the second and third month in composite flours stored at 35°C. There was a notable decrease in yeasts and moulds count in flours stored in plastic bottles and Kraft packaging materials during the second and third month of storage at 25°C and in flours stored in polythene packaging material during the third month of storage at 35°C. The decrease in the yeast and mould counts in theses packaging materials is attributable to anaerobic conditions created in plastic bottles and kraft packaging materials due to tight closing of the bottle lid and impermeability of the kraft materials.

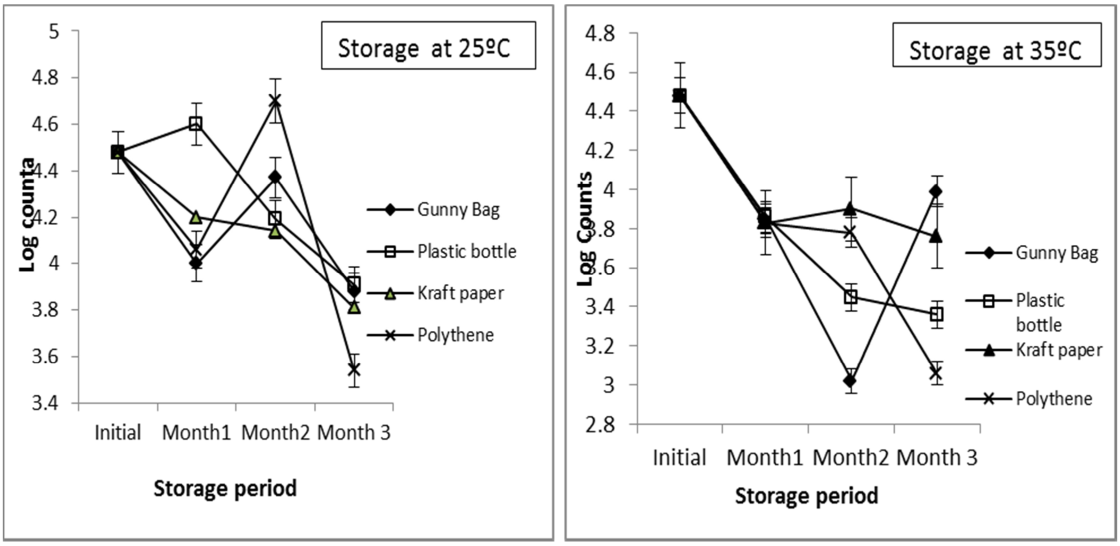


Figure 5: Yeast and mould counts of most preferred composite flour during three months of storage in different packaging materials

4.0 Conclusions and recommendations

4.1 Conclusions

Bovine blood is a rich source of iron and protein and has great potential for improving the nutritional content of diets, including composite flours, widely used in Kenya and many developing countries, to wean young children from full milk to solid food diets. Using bovine blood powder to enrich composite flours and other foods may significantly contribute to reducing iron deficiency anaemia among young children and other vulnerable populations.

The shelf life of bovine blood is greatly improved when it is hygienically cooked and dried, soon after slaughter and it becomes safer for use over a longer period of time. Without processing, bovine blood deteriorates quickly unless consumed soon after slaughter.

Bovine blood powder can be prepared locally, and in resource limited settings using, simple appropriate technology and thus has a high possibility of adoption and diffusion as an intervention at community level.

Bovine blood powder interventions can be upgraded to industrial level, for greater production and value addition. This would help address the problem of inadequate consumption of haem iron rich foods, while building capacities and empowering vulnerable populations.

The dark colour of bovine blood powder is an important factor to be considered where it is incorporated in foods such as porridge mixes, suitable foods must thus be carefully selected.

4.2 Recommendations

There is a need to carry out further studies to determine other possible method(s) for the preparation of bovine blood powder for local use and different foods. Further tests on product qualities such as bioavailability can also be conducted.

References

Allen, L. H. (2003), "Animal source foods to improve micronutrient nutrition and human function in developing countries", *Journal of Nutrition,* **133,** 3875S – 3878S.

AOAC. (1980), "Official methods of analysis of the Association of Official Analytical Chemists", (10th ed.), Washington DC.

AOAC. (1984), "Official methods of analysis of the Association of Official Analytical Chemists", (13th ed.), Washington DC.

Ferraro, G. P. (2001), "Cultural Anthropology, An Applied Perspective", (4thed.), Australia, Wadsworth Thomson Learning.

GOK. (2002), "Kisumu District Development Plan 2002-2008", *Ministry of Planning and National Development*, Nairobi, Kenya, Government Printer.

IFPRI. (2001), "Can Food Based Strategies Help Reduce Vitamin A and Iron Deficiencies?", *A Review of Recent Evidence*, Ed. Ruel M.T.

Imungi, J. K. (2006), "Limitations to Impact of Fortified Food", *Nutriview*, 2:8

Kaced, I., Hoseney, R.C. & Varriano-Marston, E. (1984), "Cereal chemistry, Factors affecting rancidity in ground pearl millet", **61**(2) 187-192.

Kenya Bureau of Standards (2003), " Composite four specifications", Ed 1 (SAC draft).

Kikafunda, J. K. & Sserumaga, P. (2005), "Production and use of a shelf-stable bovine blood powder for food fortification as a food based strategy to combat iron deficiency anaemia in Sub-Saharan Africa", *Africa Journal of Food, Agriculture, Nutrition and Development*5 (1),1-18.

Lung'aho, M. G. & Glahn, R. P. (2009), "In vitro estimates of iron availability in some Kenyan complementary foods", *Food and Nutrition Bulletin*, **30** (2),145-152.

MIYCN Working Group. (2009), "Formulations for fortified complementary foods and supplements", Review of successful products for improving the nutritional status of infants and young children, *Food and Nutrition bulletin*, **30** (2) S239-S255, United Nations University Press.

Mbagaya, G. M. (2009), "Child feeding practices in a rural Western Kenya community", *Africa Journal of Primary Health Care and Family Medicine* [Online], Available at http://www.phcfm.org/index.php/phcfm/article/view/15 (August 2nd, 2013).

McLaren, D. S. (2008), "Multiple micronutrients and "hidden hunger", A discussion paper, *Sight and Life*, **3**, 6-12.

Mosnews (2007), "Foods developed from blood developed in Voronezh", [Online] www.mosnews.com/news/2004/2007/blood.shtml (July 21st, 2007)

Mwaniki, D.L., Omwega, A.M., Muniu, E.M., Mutunga, J.N., Akelola, R., Shako, B., Gotink, M.H. & Pertet, A.M. (2001), "Anaemia and the Micronutrient Status of Iron, Vitamin A and Zinc in Kenya" *The 1999 National Micronutrient Survey Report,* MOH/KEMRI/UoN/SOMANet/UNICEF.

North American Spray Dried Blood and Plasma Producers Association (2012), Response to Advanced Notice of Proposed Rulemaking, Federal Measures to Mitigate BSE Risks"'Considerations for Further Action [Online] *www.fda.gov/ohrms/dockets/dockets/02n0273/02N-0273-EC202*(August, 2012).

Ofori, J. A. & Hsieh, Y. H. P. (2011), "Blood-derived products for human consumption", Department of Nutrition, Food and Exercise Sciences, Florida State University, Tallahassee, Florida 32306, USA , *Revelation and Science,* **1** (1)14-21.

Schumann, K. and Christ F. (2007), " Did the iron-age end in Pemba?", *Sight and Life.* **3**,6 – 13.

Sehmi, J. K. (1993), "National food composition tables and the planning of satisfactory diets in Kenya", Nairobi, Kenya, Government Printer.

Sewald, M. and DeVries, J. (2012), "Analytical progress, Food product shelf life", Medallion laboratories [Online] http:// *www.medallionlabs.com.* (July 14th, 2012)

UNICEF, UNU.; WHO. (2001), "Iron Deficiency Anaemia, Assessment, Prevention and Control*", A guide for programme Managers*, Geneva, Switzerland.

Walter, T., Olivares, M., Pizarro, F. & Hertrampf, E. (2001), "Fortification. School age children, Cookies fortified with bovine haemoglobin", In *Nutritional Anemias*, Edited by Usha Ramakrishan,CRC Press, **9**:164-167.

Acknowledgement

With gratitude to all who have supported this work. Thanks to Dr Ambrose Kipyegon, from Department of Clinical Studies and the entire laboratory staff in the Department of Food Science Nutrition and Technology, University of Nairobi, for work done during preparation and analysis of the porridge product. To all the university colleagues and friends who took part in the sensory evaluation and to Joan Waluvengo and Joseph Mugo from the unit of Applied Human Nutrition, University of Nairobi, for their assistance. Thanks also to Francis Oduor and Father Katabaro, for their support and imput to this work.

This work was supported by funds from the Kenya National Council of Science and Technology and the University of Nairobi's Deans Committee.