IMPROVING THE IRON STATUS OF CHILDREN IN KISUMU COUNTY KENYA USING PORRIDGE FLOUR ENRICHED WITH BOVINE BLOOD

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DECLARATION

This thesis is my original work and has not been presented for a degree in any other university

This thesis has been submitted for examination with our approval as university supervisors.

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ABBREVIATIONS and ACRONYMS

ASF	Animal Source Foods
CDC	Centre for Disease Control
CHW	Community Health Worker
DGLV	Dark Green Leafy Vegetables
DMT1	Divalent Metal Transporter 1
EDTA	Ethelene Diamine Tetra-acetic Acid
FAO	Food and Agriculture Organization of the United Nations
FBP	Food by Prescription
FFQ	Food Frequency Questionnaire
FGD	Focus Group Discussion
GOK	Government of Kenya
HIV/AIDS	Human Immune deficiency Virus/ Acquired Immune deficiency Syndrome
Hb	Haemoglobin
HCP1	Haem Carrier Protein 1
IDA	Iron Deficiency Anaemia
ID	Iron Deficiency
IFPRI	International Food Policy Research Institute
KDHS	Kenya Demographic and Health Survey
KEMRI	Kenya Medical Research Institute
KII	Key Informant Interview
KMIS	Kenya Malaria Indicator Survey
KNBS	Kenya National Bureau of Statistics
MI	Micronutrient Health
MMS	Multiple Micronutrient Supplements
MUAC	Mid-Upper Arm Circumference
N-CRSP	Nutrition Collaborative Research Programme
RDA	Recommended Dietary Allowance
SD	Standard Deviation
SDA	Seventh Day Adventist
SF	Serum Ferritin
SPSS	Statistical Package for the Social Science
sTfR	Soluble Transferrin Receptor

UNICEF	United Nations Children Fund
UNU	United Nations University
WHO	World Health Organization

DEDICATION

To my parents Bridget and Austin Orowe who are my foremost nutritionists,

My husband Fred and our children Linda, Jaqueline, Martin, Joy and Patricia

For their support and encouragement,

My mother-in-law, Felista Awour Ochieng, the inspiration behind bovine blood.

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To my best friend I say **THANK YOU HEAVENLY FATHER!**

ABSTRACT

There is a high prevalence of iron deficiency and anaemia in Kenya with young children and mothers bearing the greatest burden. Bovine blood is acceptable food in Kenya but it largely goes to waste. It therefore could be used to improve local diets as a strategy for preventing anaemia. This study evaluated the effect of consumption of bovine blood enriched porridge, on the iron status of anaemic preschool children in a malaria endemic region of Kenya.

A rapid assessment was conducted to determine knowledge, attitudes and practices related to anaemia and bovine blood use in East sub-countyof Kisumu County. This involved four key informant interviews, four focus group discussions, free listing and observations of slaughter houses. This was followed by biochemical screening of 270 children for haemoglobin status. A total of 140 children were tested and out of these a sample of 102 children were selected for the household survey and the intervention. A structured questionnaire was used in the baseline householdsurvey to collect background data on socio-demographic and socio-economic characteristics, food consumption, anthropometric measurements and morbidity characteristics of the children. Iron status was also assessed.

Based on the knowledge from the rapid assessment, porridge flour was developed from sorghum, millet and cassava flours and enriched with bovine blood powder. The flours and bovine blood powder were analysed for proximate composition and specific mineral contents. Using the results, Nutri-survey modelling was used to formulate the composite porridge flours to provide approximately 50% of the iron RDA for young children. Sensory evaluation was performed to determine the most acceptable porridge flour. The most acceptable flour was then subjected to shelf life evaluation tests.

During the intervention phase of the study, children were randomly allocated to two feeding groups. One group was fed with the porridge containing the blood (intervention

group) and the other with the porridge without blood (placebo group). The feeding period lasted six weeks, after which the children were re-evaluated for haemoglobin and iron status.

Qualitative and quantitative data were analysed with the help of ATLAS *ti*, Nutrisurvey and IBM SPSS Statistics Base, version 22 respectively. Summary statistics for age, sex, socio-demographics, anthropometry, and biochemical indicators at baseline were determined to describe the study population. Analysis of covariance was used to determine any differences between treatment groups, and group-specific effect sizes calculated and

$P < 0.05$ was considered significant.

Results of the rapid assessment indicated that the community was aware of anaemia and its main causes. Bovine blood is well-accepted as food and traditional methods exist for it's preparation for consumption and preservation.

The most acceptable porridge mix contained sorghum: finger millet: cassava: blood powder in the ratios of 6:14:10:0.5 respectively, andprovided 54% of the recommended daily intake of iron for 1-3 year old children, when consummed as one cup of porridge daily. The mix had a better shelf life when stored at 35ºC for up to three months.

Out of 270 children screened, 51.86% were anaemic and 34.7% suffered from iron deficiency anaemia. There were no significant differences in the baseline characteristics, morbidity status and nutritional status of children in both feeding groups ($p>0.05$). The dietary recall indicated that most children (88.9%) were receiving less than the RDA foriron. The prevalence rates of stunting, wasting and underweight were 24.5% and 10.8% and 16.7% respectively.

After six weeks of intervention 52.6% of the children achieved normal haemoglobin levels, mostly from the intervention groupwhich had significantly($p= 0.000$) higher levels compared to the placebo group.There was also a significant ($p < 0.05$) difference in soluble transferrin receptor levels between the two groups. However, the mean change in serum ferritin between the groups was not significant ($p = 0.080$).

The study concludes that porridge flour formulated from sorghum, millet, cassava and bovine blood can be effectively used to improve the iron status of anaemic children living in malaria endemicregions of Kenya.

CHAPTER 1

GENERAL INTRODUCTION

Iron deficiency and specifically iron deficiency anaemia, remains one of the most severe and important nutritional deficiencies on a global basis (UNICEF, UNU, WHO, 2001; IFPRI, 2001; Kennedy *et al*, 2003) and is considered to be among the top ten contributors to the global burden of disease (McLean *et al*, 2007). Iron deficiency disorders affect populations in both developed and developing countries and all age groups are vulnerable (WHO, 2008). The prevalence of iron deficiency in developing countries is however, much higher than in developed countries. The most vulnerable population groups are women in the reproductive age group and young children with 40-60% of the children in many developing countries, reported to suffer from iron deficiency anaemia. A joint global report described the occurrence in developing countries as '*the rule rather than the exception*' (UNICEF and MI, 2008). A national micronutrient survey in 1999 revealed that iron deficiency is widespread in Kenya, with the moderate and severe forms being more common than mild forms of the deficiency (Mwaniki *et al*, 2001).

Children who are iron deficient tend to be weak and tire more easily, they eat less, are easily irritable, have short attention spans, fall ill frequently and fail to grow normally (both mentally and physically) compared to children who are not iron deficient. In adulthood iron deficiency causes fatigue and reduced work capacity. In pregnant women, severe anaemia may cause foetal growth retardation or low birth weight and is responsible for a large share of maternal deaths (Ruel, 2001).

Awareness about the magnitude and health implications of iron deficiency has led to the commitment of world organizations and governments to focus on addressing the problem to reduce the high prevalence rates, especially among vulnerable populations (WHO, 2008). Iron deficiency interventions targeting young children are important because they have the potential to improve iron status, and thus enable them to develop optimally and live healthier, productive lives. Food based strategies or approaches, have received strong support as sustainable means of meeting the nutritional needs of this population group.

Although iron can be obtained from plant and animal foods, evidence from several studies, such as the Nutrition Collaborative Research Programme (N-CRSP), indicates a strong positive association between Animal Source Food (ASF) intakes and micronutrient status when compared with other interventions (Allen, 2003). However, in the developing countries, the most vulnerable and malnourished groups, suffering from iron deficiency, often cannot afford sufficient amounts of animal source foods (Ruel, 2001). Bovine blood, a cheap by-product of beef slaughter, but which is rich in iron was used to enhance a local, low iron porridge.

Factors Influencing Iron Status

Adequate iron status ensures that the body is able to meet all the required physiological functions. Iron plays a vital role in oxygen transport and storage, oxidative metabolism, cellular proliferation and other physiological processes in the body. Approximately 70% of the human body's 2.5- 4g of iron is present in haemoglobin, the oxygen carrying pigment of the red blood cells. Myoglobin, the oxygen binding storage protein found in muscles, contains about 4% of body iron. Trace amounts of iron are also associated with electron transport and several enzymes, such as the haem containing cytochromes, iron sulphur proteins and hydrogen peroxidases. Storage iron constitutes about 25% of body iron, mainly in the liver, with smaller amounts in the reticulo-endothelial cells of the bone marrow, spleen and muscle tissues (Lynch, 2007; Gibson 2005).

Iron balance in the body is maintained through the control of absorption. Iron absorption has been known to occur primarily in the proximal small intestine, through mature enterocytes located at the tips of the duodenal villi. Two transporters, Haem Carrier Protein 1 (HCP1) and Divalent Metal Transporter 1 (DMT1), are believed to mediate the entry of almost all dietary iron into these mucosal cells. In more recent years the discovery of the enzyme hepcidin, a small cysteine-rich, cationic peptide, produced in the liver, has been identified as a key regulator of iron metabolism (Lonnerdal and Kelleher, 2007; Lynch 2007).

The main factors that affect iron balance and metabolism are intake, stores and losses. Intake is determined by the quantity and bioavailability of dietary iron as well as the capacity to absorb iron. The amount of dietary iron absorbed is in turn influenced by individual nutritional needs and factors influencing bioavailability. Haem-iron is more bioavailable compared to non-haem iron. Ingestion of dietary inhibitors such as phytates and polyphenols also limit absorption in non haem iron. Calcium inhibits both haem and non-haem iron absorption. Enhancers of iron absorption include vitamin C, source of foods (animal or plant) human breast milk and organic acids The amount of iron absorbed can vary from less than 1% to more than 50% depending on the source (Lynch, 2007; Gibson,2005, Bowman and Russell, 2001).

The amount of iron in stores varies widely depending on sex and iron status. Chronic ID leads to depletion of stores in the liver, spleen and bone marrow whereas positive iron balance increases stores gradually (Gibson, 2005). Total daily iron losses are small (except for menstrual blood loss in women), occurring mainly through faeces, desquamated skin cells, sweat and urine. Most of the iron in erythrocytes is recycled for haemoglobin synthesis at the end of their approximately four month lifespan. Each day 0.66% of the body's total iron content is recycled in this way (Gibson, 2005, Lynch, 2007).

Causes of Iron Deficiency Anaemia (IDA)

Maternal iron deficiency, prematurity and low birth weights are significant causes of IDA. During pregnancy iron requirements increase from approximately 18 mg to 27 mg for iron replete women. Women who start pregnancy with low iron stores are at high risk of developing anaemia, during their second and third trimesters. Short intervals between pregnancies, adolescent pregnancies and multiple births are also risk factors for anaemia (Gleason and Scrimshaw, 2007; Allen and Casterline-Sabel, 2001).

Infants of anaemic mothers are likely to have low iron reserves and thus start life disadvantaged. Anaemia in pregnancy is associated with increased maternal morbidity and mortality. Impacts on infants include premature birth and low birth weight, which is a risk factor for anaemia. Premature births are also associated with inadequate iron stores due to shortened gestation period resulting in anaemia (Thurnham and Northrop, 2007; Worthington-Roberts and Williams, 2000; Allen and Casterline-Sabel, 2001).

A major determinant of nutritional iron deficiency is insufficient dietary intake to meet daily requirements and bioavailability factors. Depletion of iron stores at 4 to 6 months and inadequate supplies from breast milk is a cause of deficiency, hence the recommendation for additional iron in diets of children by 6 months (Thurnham and Northrop, 2007). The iron content in human milk, though lower than in cow's milk or infant formula, is significantly more bioavailable at 45 to 100 percent. Iron intake and bioavailability among older children also relates to its absorption in the presence of inhibiting and enhancing factors. Total nutrient intake is important and underweight and wasting were significantly associated with anaemia during the 1999 survey in Kenya (Kennedy *et al*, 2003, Mwaniki *et al*, 2001).

Frequent exposure to inflammation and infection increases the risk of developing anaemia in infants and young children. Parasitic infections also contribute to inflammation and anaemia. Thus anaemia is common in many developing countries where there is a high prevalence of diarrhoea, vomiting, fever, malaria and helminth infection (Thurnham and Northrop C., 2007). Of particular concern in Kenya are malaria, diarrhoel diseases, acute respiratory infections, HIV/AIDS and TB. Genetic conditions affecting haemoglobin include sickle cell and thalassaemia and are important contributing factors (Mwaniki *et al*, 2001).

During periods of increased growth, altered metabolism and during illness, additional iron is required by young children. Anaemia often develops due to iron deficiency, and is exacerbated when both these conditions occur together.

Methods for Assessing Iron Status and Anaemia

The World Health Organization estimates that the primary cause of most anaemia globally is nutritional deficiencies. However, detection is often complicated by the high prevalence of infectious diseases that cause anaemia in many tropical countries. The most important are malaria and hookworm (Lynch and Green, 2001).

Since iron deficiency is known to respond to supplementation or fortification, assessment of iron status is crucial in the evaluation of nutritional anaemia (Biesalski and Erhardt, 2007). There are clinical indicators of iron deficiency, such as chronic fatigue, whose symptoms are unspecific. Evaluation of iron intake, which differentiates between haem and non-haem iron may be useful, but diagnosis relies mainly on biochemical indicators.

In most cases iron deficiency occurs in three sequential stages. Depleted iron stores represent the first phase. This indicates the earliest evidence of iron deficiency. Though no functional consequences occur, there are no iron reserves to meet future physiological and pathological requirements. Early functional iron deficiency or iron-deficient erythropoiesis is the second stage where the supply of iron to the bone marrow and other tissues is sub-optimal, but not reduced sufficiently to cause measurable anaemia. In the third phase of Iron Deficiency Anemia (IDA) there is a measurable deficit in the most accessible functional compartment, the erythrocyte (Biesalski and Erhardt, 2007; Lynch and Green, 2001).

The three stages in the development of iron deficiency anaemia can be analyzed biochemically. There is an agreement that the measurement of Hb, Serum Ferritin (SF) and Soluble Transferrin Receptor (sTfR), complemented with indicators of acute and chronic infections, is the best procedure for evaluating iron status (Biesalski and Erhardt, 2007). These methods are also viewed as difficult and costly, especially in the context of developing countries. Based on these recommendations, this study used Hb, SF and sTfR biochemical analyses at the baseline and after intervention to evaluate change. Screening for malaria and deworming was also done in line with recommendations (Lynch and Green, 2001).

Strategies to Address Iron Deficiency Anaemia

The main strategies used to control iron deficiency anaemia are supplementation, food based approaches such as fortification, dietary diversification and dietary modification and public health measures. (Kennedy*et al*, 2003; Allen, 2003; Allen and Gillespie, 2001;IFPRI, 2001; WHO/CDC, 2007).

Experiences indicate that although each one of these strategies can contribute to the control of micronutrient deficiencies, none can do the job alone (UNICEF and MI, 2008; Zimmermann 2007).

Supplementation, a technical approach where high dose preparations of nutrients are delivered by means of oral preparations as syrups, pills or parenteral preparations as oil solutions, is usually used as a short term measure, for targeting populations with a high risk of deficiency and is then later replaced with long term measures (GOK, 2008). Iron supplementation is indicated when diet alone cannot restore deficient iron levels to normal within an acceptable timeframe. The two forms of supplemental iron available are ferrous and ferric. Ferrous iron salts (ferrous fumarate, ferrous sulfate and ferrous gluconate) are the best absorbed forms of iron supplements.

Benefits of iron supplementation include a rapid increase in iron as haemoglobin usually increases within two to three weeks of starting supplementation. The intervention is also relatively simple to implement and monitor. Therapeutic doses of iron supplements, which are prescribed for iron deficiency anaemia, may lead to undesirable side effects among some people, such as nausea, vomiting, constipation, diarrhoea, dark coloured stools, and/or abdominal distress (Schumann and Solomons, 2007). The necessity for haematological screening in malaria endemic regions, to avoid negative outcomes, makes supplementation a costly intervention. When excess supplementary iron is taken, there is formation of free iron radicals leading to increased bacterial replication and iron toxicity. Blanket supplementation among vulnerable populations located in plasmodium falciparum malaria endemic regions has negative consequences, unless WHO recommendations are followed (Schumann and Christ, 2007).

In recent years, Multiple Micronutrient Supplements (MMS) have been advocated, as opposed to isolated supplements for each micronutrient. A number of commercial formulations of multiple micronutrients have been developed and numerous trials in developing countries continue to be carried out. Examples include Sprinkles, Foodlets and iron-fortified spreads. Sprinkles, is a blend of micronutrients which is added to food just before consumption, it has proven to be highly effective in reducing anaemia among poor children in rural Haiti (McLaren, 2008; IFPRI, 2007). A major concern about these supplements is the fact that they are manufactured in developed countries then imported into developing countries, hence their sustainability is doubtful (Stacia Nordin, 2005).

Fortification of weaning foods is one of the key strategies of preventing iron deficiency in young children growing in developing countries. This strategy has been used with varying degrees of success, in different countries. In some developed countries like America, well designed policies on universal fortification and supplementation for young children, based on recommendations of the American Association of Pediatrics (AAP), and has ensured that iron deficiency anaemia is limited (www.aapnews, 2010).

In many developing countries like Kenya, the administrative, technical and financial challenges presented by fortification have hindered the provision of adequate amounts of micronutrients, including iron, to young children (Mwaniki *et al*, 2001; Zimmermann, 2007). Furthermore the main staples in developing nations, such as maize and sorghum are often processed in small, rural mills which are not centralized, making fortification logistically challenging (WHO, 2008). Fortification in these countries requires careful planning, management and monitoring for effective impact, and can only be achieved through the sufficient will and commitment of governments (Imungi, 2006).

Food based approaches, also referred to as dietary diversification and modifications, encompass a variety of interventions that aim to; increase the production, availability and access to foods high in iron, through agricultural programmes and policies; increase consumption of micronutrient/iron rich foods, through social marketing, behavior change and preservation programmes; improve the bioavailability of iron through home processing techniques and food combinations that increase iron bioavailability and enriching staple crops with iron through bio-fortification (UNICEF/UNU/WHO, 2001; IFPRI, 2001; Kennedy *et al*., 2003).

Reduction of concurrent infection is important for the success of IDA control interventions. The Kenya National Technical Guidelines for Micronutrient Deficiency Control (MDC) recommends specific control and/or treatment where hookworm infestation, urinary schistosomiasis and plasmodium falciparum malaria are endemic. Other diseases which may lead to the problem of iron deficiency anaemia must also be addressed. In Kenya the use of combined therapy drugs, distribution of bed nets and indoor spraying with pyrethroids, has contributed to reducing mortality from malaria among children less than 5 years of age (GOK, 2008; Mannar 2007).

Improving Iron Intake Using Food Based Strategies

Food based approaches represent the most desirable and sustainable method of reducing and preventing iron deficiency. Food based approaches including fortification of staple foods and condiments for the general populace, as well as home fortification for specially targeted groups, are perceived to have great potential to address iron deficiency as they are more sustainable; are less perceived as treatment of a condition; and are applicable for use in malaria endemic areas (De Pee *et al*, 2007).

Whereas food based approaches offer the most effective means of addressing the problem at source, identifying suitable, affordable and effective foods at household level, poses major challenges, especially as most animal source foods rich in haem iron, are too expensive for the families of the most vulnerable. Furthermore, food avoidances worldwide most frequently relate to animal foods, mainly due to religious and cultural reasons (Hartog *et al*, 2006; Sehmi 1993).

The iron requirement of young children aged 12-60 months varies from 7-11mg/day depending on age, sex and physiological status (Food and Nutrition Board, 2001). On average, most Kenyan children are introduced to semi-solid foods between the ages of four to twelve months (Mwaniki *et al*, 2001). Which are mainly starch based, of high bulk and low nutrient density, including low iron content. Young children, from the age of 6 months to 60 months, are thus rendered particularly vulnerable to iron deficiency (Agostoni *et al*, 2008; UNICEF/MI, 2008; Mwaniki *et al*, 2001).

Perception of Animal Blood as Food

Animal blood is an underused and widely available source of food (Ofori and Hsieh, 2011). A preliminary survey of slaughter houses in Kenya, indicated that blood from public and private slaughter houses is largely disposed of, with the largest meat plant in Kenya disposing of about 600 litres of bovine blood daily, except when they have requests for its use in animal feeds (Kenya Meat Commission, 2008).

Nutritionally, cow's blood, is a richer, cheaper source of haem iron than liver or eggs. It is also a rich source of protein, calcium and phosphorus. The high nutrient content and functional properties of bovine blood, has in recent years led to increased research in its possible use in food. Scientists in Russia and Chile determined that bovine blood is rich in iron and protein and that the proteins are assimilated by the human body twice as fast as chicken-egg proteins (Mosnews, 2007; Walter *et al*, 2001). Other studies have also confirmed the high iron and protein content of bovine blood and its safety for human consumption (Ofori and Hsieh, 2011). However, it's potential to reduce IDA among vulnerable populations in Africa has not been investigated. This study therefore gives some insight on how bovine blood can be processed in the local setting, and used to enrich young children's porridge, in order to reduce iron deficiency anaemia.

The use of animal blood as food or in food is a culture practiced by several communities in various parts of the world (Ofori and Hsieh, 2011; Kikafunda and Sserumaga, 2005). In Africa it is most common among pastoralist communities. In Kenya, cow's blood is often consumed as a mixture with milk, by the Maasai people. Other communities also consume blood, incorporated in various food preparations. An example is the *mutura* (African sausage), commonly prepared and consumed by the people of central Kenya, contains blood as one of the ingredients in the filling. *Mutura* is a meat emulsion, prepared from blood and offal's (mainly intestines and stomach of goat), cooked and stuffed in a section of the large intestine and then roasted. In Western Kenya blood mixed with milk and boiled (called *riga* in Nyanza) is a delicacy consumed with *kwon* (ugali) when bovine blood is available.

The objective of the rapid assessment conducted at the start of the study was to show bovine blood availability, acceptance and use in the targeted community. The rapid assessment also gave information on foods commonly consumed by young children; this guided the choice of the food to be enriched with bovine blood. Since in some communities or religious groups the use of bovine blood as food is prohibited, it was crucial to ensure acceptance in the community.

Iron Deficiency and Anaemia among Children in Kenya

According to the results of the 1999 Kenya national micronutrient survey, 60% of Kenya's population is iron deficient, with the largest burden of anaemia being borne by children and pregnant and lactating mothers. The study dealt with children 6 to 72 months old. The mean haemoglobin concentration in children was below the cutoff of 108g/L for anaemia adjusted for altitude. Among infants aged less than six months the mean Hb level was 9.63mg/L and 10.1mg/dl for those aged6 to 72 months. Haemoglobin levels were lowest in the lake basin and Western highlands sub regions, coastal areas and the semi-arid lowlands, where 99.5% of less than 6 months and 69% of 6-72 months were anaemic (Mwaniki *et al*, 2001).

Food for Feeding Children in Kenya

Most Kenyan children are introduced to semi-solid foods between the ages of four to twelve months. The Kenya micronutrient survey carried out in 1999, found that porridge (or gruel) is the most common semi-solid food in infant diets, and was consumed by 74% of the infants in the study. *Ugali* with milk or soup, was also fairly commonly consumed. Contrary to expectation, fish consumption was low among young children in the lake-basin and coastal regions. Consumption of meat and blood, which represent important sources of dietary iron were rarely reported (Mwaniki *et al*, 2001). Green vegetables are key components of the adult diet but information on their use by young children is lacking (Rotich, 2004).

Porridge is the most common complementary food and is predominantly prepared from maize flour. Other commonly used cereals and tubers are sorghum, millet and cassava flours. The common practice is the use of a mixture of maize flour with millet or sorghum and cassava or both, depending on availability and personal preferences and practices (Mwaniki *et al,* 2001). It is recommended that more than 90% of the iron requirements of young children be met by complementary foods, which should provide sufficient bio-available iron. Both breast and cow's milk, which are important sources of calcium and protein in these diets, are poor sources of iron. In most cases, studies indicate that it is almost impossible to meet the iron needs of young children from unfortified complementary foods (Agostoni *et al*, 2008).

Problem and Rationale for the Study

Iron deficiency anaemia is a major public health problem in Kenya with young children and pregnant mothers bearing the greatest burden of the associated negative health consequences. Food based approaches are seen to offer the most effective means of addressing the problem at its source, however the animal source foods like liver are rich in iron, are expensive thus have limited accessibility. The main challenge therefore lies identifying suitable, affordable and effective iron-rich foods. This study therefore investigated the potential of bovine blood, an underused, haem- iron rich source of food in many parts of the country, and the possibility of using it to reduce the anaemia gap among young children. Kisumu County was selected for the study due to the high prevalence rates of IDA and the fact that bovine blood is an accepted traditional food.

Overall Objective

The overall objective of the study was to assess the potential of bovine blood enriched porridge in improving iron status of anaemic children in Kisumu County, Kenya.

Specific Objectives

1. To establish the knowledge and practices related to iron deficiency anaemia in Kisumu County, Kenya.
2. To determine the knowledge, attitudes and use of bovine blood as food in Kisumu County, Kenya.
3. To develop a bovine blood enriched porridge flour for use in alleviating anaemia among young children.
4. To determine the effect of consumption of bovine blood enriched porridge on anaemia and iron status of anaemic children.

Study Area

The study was done in Kisumu County. The county was purposively selected due to the high prevalence of anemia as well as the established practice of incorporating bovine blood into food. Kisumu County lies in the south-western part of Kenya along the shores of Lake Victoria within longitude 34º 10E and 35º 20E and latitude 0º 20S and 0º 50S. Kisumu County has 6 sub - county's, namely; Kisumu North, Kisumu East, Kisumu West, Nyando, Muhoroni and Nyakach and borders Homa Bay County to the south, Kericho County to the east and south-east, Nandi County to the north and north-east, Vihiga County to the north and north-west and Siaya County to the west and north-west. Figure 1 shows the location of Kisumu County in Kenya (KNBS, 2012).

Kisumu East sub-county was selected to represent the others because it hosts a high proportion of the county's population. The sub-county contains two administrative divisions; Winam and Kadibo. Winam division is larger and more populated than Kadibo, and hosts the county and district headquarters. Kisumu City is located in Winam as well as the three largest informal settlements in the county; Manyatta, Nyalenda and Obunga, where over 40% of the county's population live. Kadibo Division on the other hand, represents the rural population in the county (GOK, 2009). Kisumu East Sub-County has a long shoreline along Lake Victoria and fishing is a key source of livelihood and food. Mean annual temperatures range from 20ºC to 30ºC, and the mean annual rainfall range is between 1,000 and 1,800mm. Rainfall reliability is low, resulting in essentially one planting season. The main food crops cultivated are maize, sorghum, millet, beans, green grams, kales, cow peas, groundnuts, tomatoes and sweet potatoes. The major cash crops are sugar-cane and rice. The main livestock kept are cattle, sheep, goats and poultry (GOK, 2002; CBS 1999; GOK, 2009).

Kisumu East sub-county with a population of 150,124 is the second most populous sub county after Kisumu Central sub-county. The number of children aged 0-5 years is 190,143. Unemployment is high, and 49% of the populations live below the poverty line

(KNBS, 2010). The infant mortality rate in the district is 123 per 1,000 and the under-five mortality rate is 220 per 1,000. The three most prevalent diseases in the sub county are malaria, pneumonia and anaemia. HIV/AIDs is a significant health problem and major cause of mortality in the entire county and the prevalence rate (11.2%) is higher than the national prevalence rate of about 6% (GOK, 2009; KDHS 2008/9).

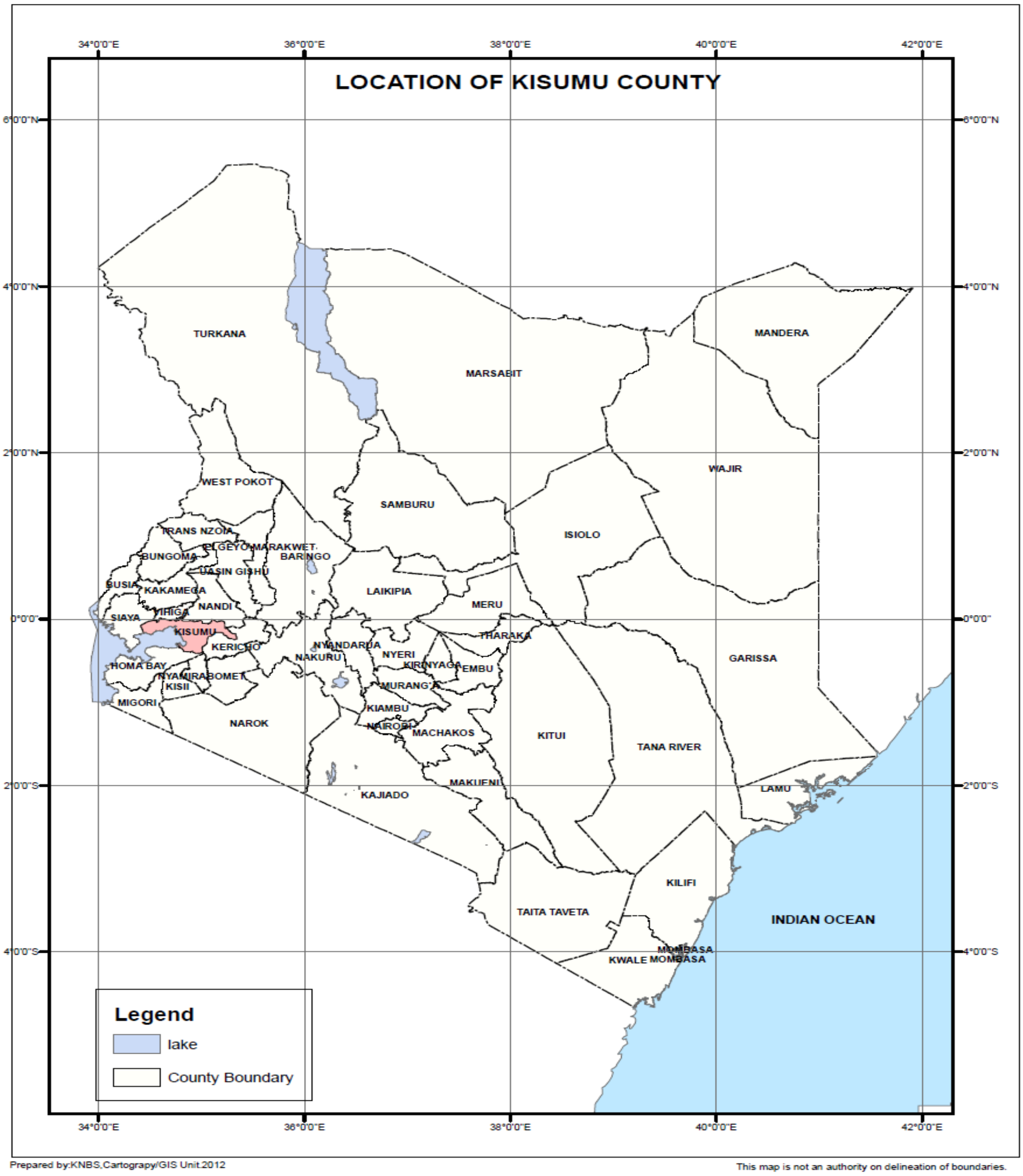

Figure 1: Location of Kisumu County in Kenya

Thesis Layout

The study was organized as four sub-studies, which for purposes of presenting the thesis are organized in six chapters as follows:

Chapter 1: A general introduction to the study describing the study background including the problem, rationale and objectives.

Chapter 2: **A** rapid assessment to establish existing knowledge and dietary management of iron deficiency in the study area. The study also determined foods commonly consumed by children, with a view to identifying a vehicle for iron enrichment.

Chapter 3: The rapid assessment also determined the knowledge, attitudes and use of bovine blood in the study area.

Chapter 4: Development of an acceptable bovine blood enriched porridge flour for use in the study.

Chapter 5: The intervention study, a randomized controlled trial, to evaluate the efficacy of the porridge in improving the iron status of anaemic children.

Chapter 6: General discussion on main findings, conclusions and recommendations.

CHAPTER 2

KNOWLEDGE AND PRACTICES RELATED TO IRON DEFICIENCY ANAEMIA IN KISUMU COUNTY KENYA

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Knowledge and practices related to iron deficiency anaemia in the Lake Victoria region of Kenya.

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ABSTRACT

Iron deficiency and anaemia are a public health problem in Kenya affecting about 60 % of young children. Previous studies identified inadequate dietary intake of iron as the main cause of the deficiency. Poverty, lack of knowledge and adoption of new eating habits are probable contributors to inadequate dietary intake. This study was a rapid assessment of the knowledge and practices of community members and caregivers of young children with regard to iron deficiency and anaemia in Kisumu County. Key informant interviews and focus group discussions were used to collect data from community groups and experts. The study showed there exists general knowledge of anaemia among the community although the community did not have a specific name for iron deficiency or anaemia, using only descriptors. Cereal porridges are the main foods used to feed young children and consumption of animal foods is low due to their prohibitive costs. There are no institutional attempts to enrich young children's porridge to improve iron content, because knowledge of the low iron content of the porridges is low. The results indicate that despite existing general awareness about anaemia and its dietary causes, formulations of iron rich foods for children is low.

INTRODUCTION

Iron deficiency is considered one of the world's top ten contributors to the global burden of disease and affects about 2 billion people in both the developed and developing countries, with developing countries bearing the greatest burden. Majority of the vulnerable population groups are women in the reproductive age and young children. Approximately 40-60% of the children in many developing countries are reported to suffer from iron deficiency anaemia (WHO, 2015; Mclean *et al*, 2007). The prevalence within the general population in Kenya is about 60%, with the majority coming from the Lake Victoria region, the Western highlands sub regions, coastal areas and the semi-arid lowlands where practically all infants (99.5%) were reported to be anaemic (Mwaniki *et al*, 2001).

Children who are iron deficient tend to be weak and tire more easily. They eat less, are easily irritable, have short attention spans, fall ill frequently and fail to grow normally both mentally and physically (Ruel, 2001).The major risk factor for Iron Deficiency Anaemia (IDA) is low dietary intake of iron. Populations subsisting on diets based on starchy staples are vulnerable because the iron is tightly bound to phytates or phenolic compounds and are therefore poorly absorbed. During periods of increased growth, altered metabolism and illness, iron requirements by the children are heightened and iron deficiency may evolve, even when adequate dietary intake exist (Ruel, 2001).

Awareness about the magnitude and health implications of iron deficiency has led to the commitment of governments and world organizations to focus on addressing the problem, by attempting to reduce the high prevalence rates, especially among vulnerable populations (De

Benoist, 2008). Food based approaches, also referred to as dietary modifications encompass a wide variety of interventions including supplementation, fortification, and dietary diversification (Kennedy *et al*. 2003; WHO/CDC, 2007). Foods that are selectively formulated to contain high levels of iron augment very well the public health institutions to combat IDA. However this may not be within the common knowledge of the communities and high prevalence of anaemia may linger due to inappropriate dietary formulations.

Studies have reported that knowledge and awareness about anaemia leads to practices that lower risk of developing anaemia (Soungainidis *et al*. 2012; Natalya *et al*. 2007; Galloway *et al.* 2002; Vereecken and Maes, 2010). Lack of proper knowledge about iron deficiency and anaemia may be the reason why practices related to solving the problem of anaemia remain inadequate in many regions of the world.

The objective of the study was to assess knowledge and practices related to iron deficiency and anaemia, as well as knowledge on iron rich foods and anaemia management among those living in the malaria endemic region of Kisumu County, with a view to designing a suitable dietary intervention.

METHODS

The study was carried out in Kisumu East sub-county of Kisumu County. This area was purposively selected because unemployment and poverty in the sub-county are widespread with 49% of the population living below the poverty line.The infant and the under-five mortality rate in the sub-county are 123 per 1,000 and 220 per 1,000 respectively. The three most common diseases in the district are malaria, pneumonia and anaemia (GOK, 2009). HIV/ AIDs is a significant health problem in sub-county with an overall prevalence rate of 18.7%, which is above the national rate of 6.1% (GOK, 2012).

Qualitative data was collected usingFocus Group Discussions (FGD's), Key Informant Interviews (KII's) and free listingduring the FGD's to supplement group discussions. FGD's were held with mothers (12) and fathers (9) of young children, senior citizens (12) and community health workers (12).Senior citizens in the FGD's were adults aged between 50 and 70 years and were included in the FGDs because they are perceived to possess traditional knowledge and practices, which the younger adult's may not have. Moreover, they also often have influence on child feeding and care practices as grandparents (Roberts and Pettigrew, 2010; Aubel, 2011). Each Community Health Worker (CHW) was in charge of 50-100 households for monitoring and referral to the provincial general hospital when necessary, they also take part in health and nutrition talks in the community. Mothers and fathers of young children as well as community health workers were the main sources of information on current knowledge and practices in the community. Consent to carry out the study was obtained from the of Kenyatta National hospital ethical committee and the Nyanza Provincial Nutrition office. Informed consent was also obtained from each participant.

The key informants included the Nyanza Provincial Nutritionist, the Kisumu East District Nutritionist, the District Clinical Health Officer, and the District Veterinary Health Officer. Discussions and interviews were conducted using structured interview and question guides.

Apart from the District Veterinary Health Officer whose questionnaire differed slightly, to provide information related to bovine blood and slaughter houses in the Region, the other three key informants gave information on anaemia situation and practices to control it.

All interviews and discussions were conducted by the researcher and one assistant. Information was audio-taped using digital recorders, and thereafter transcribed and uploaded in *Atlas-ti* in windows for coding and subsequent analysis.

RESULTS

Characteristics of Study Participants

Most mothers, fathers and senior citizens were involved in small scale income generating activities such as selling foods and second hand clothes, tailoring, mending shoes, welding, shop and bar attendants and transportation business such as bicycle-taxi(*boda-boda*). A few mothers were full-time housewives and some senior citizens were retirees.

Knowledge on Anaemia

All the focus group discussions participants were aware of anaemia and its basic effects in the body, but there was no specific name for anaemia in the local dialect; only descriptors used fathers and senior citizens described anaemia as ***'ng'at ma rembetin'*** meaning a person with little blood, or ***'rembe*** **orumo'** meaning blood is finished. In addition to these terms some mothers described anaemic persons as ***'worm infested people'***. In the community anaemia is mainly identified through symptoms like palmar and eye pallor as well as general body weakness. It was also reported that mothers of young children are advised in the clinic on how to check for anaemia while breastfeeding. They are advised to look at the face, look at the eyelids, the tongue, and the palms of their hands, and compare with their own. But as reported by key informants and CHW's, occasionally even the mother's palm is pale.

Many of the participants know that anemia is a form of malnutrition. Anaemia was also strongly associated with morbidity. All Key Informants (KI's) indicated that the anaemia prevalence among young children is high, and cases of severe anaemia are more common than mild or moderate cases by the time children are brought to hospital. According to the district public health officer it was estimated that out of every 100 children brought to hospital, 50 would be anaemic"

Knowledge on Causes of Anemia

Inadequate or monotonous diets and morbidity were indicated as the most important causes of anaemia in the community. Reasons given for the monotonous diets were high poverty levels, food insecurity and ignorance. The poverty levels, estimated at above 60% by the provincial nutritionist, were attributed to mothers having no or low income, thus depending on their husbands income from informal and often unstable jobs such as quarrying and bicycle taxi (*boda-boda*). Participants also indicated that there is little or no land for food production in the informal settlements, which has contributed to food insecurity and poverty.

Morbidity is perceived as a significant cause of anaemia. The most common illness was reported to be malaria, which sometimes went untreated, alongside other diseases such as

HIV/AIDS and worms. Worm infestation was perceived to contribute to loss of blood. Other conditions leading to blood loss and anaemia were indicated as illegal abortions by most participants and fibroids, as indicated by senior citizens.

Poor dietary practices were attributed to mothers' ignorance, food choices and attitude by most respondents. According to the CHW's:

"… mothers have a poor habit of giving one type of food, mostly carbohydrate, she uses the cheapest and simplest food available".

The senior citizens reported that young mothers tend to feed young children from the family meal, which often is less palatable to the child and contains less than the child's requirements of nutrients. According to the public health officer, some mothers especially those who run small scale business, leave the children in the morning and only come back when the children are tired and sleepy in the evening, they arriving with food to be cooked for the child. The child, who has fed on nothing throughout the day, eats very little and then goes to bed. Other causes of anaemia indicated by very few focus group members were poor child spacing, or child born to anaemic mother. A summary of all the causes of anaemia indicated is shown in Table 2.1, presented as those commonly indicated and those less commonly indicated.

Table 2.1 Knowledge on causes of Anaemia

Causes commonly reported	CHW's	Mothers	Fathers	Senior citizens	KII's
Inadequate / diets	√	√	√	√	√
Malaria	√	√	√	√	√
HIV	√	-	√	√	√
Blood loss conditions	-	√	√	√	-
Worms/diarrhea	-	√	√	-	√
Poor care practices	√	-	√	√	√
Sickle cell anaemia	√	-	-	√	-
Ignorance	√	-	√	-	-
Colds/flu	-	-	√	√	√
Less commonly reported					
Eating soil		√			
Sitting near hot fire		√			
Some drugs		√			
Stress		√			
Alcoholism			√		
Diet not balanced			√		
All diseases			√		
Chira				√	
Child born to anaemic mother				√	

Foods Consumed by Young Children

All the discussions and interviews identified porridge and Irish potatoes as the main foods consumed by all young children. According to the provincial nutritionist,

"Complementary feeding for young children starts with porridge and graduates to blended or mashed semi-solid foods".

Porridges for children are mainly composite in nature, with the most common being prepared from the flours of cereals, mainly sorghum, finger millet, maize and cassava. Other foods such as *Dagaa* or *Omena* (*Rastrineobolaargentea),* beans, groundnuts and soy beans in the form of flour are commonly added to enrich porridge flours. The flours are either purchased or prepared at home.

According to the District Nutritionist and Public Health Officer, the porridge is usually cooked for about ten minutes only. Inadequate cooking time was reported to be a possible cause of diarrhea common among children. Key informants and CHW's also indicated that most young children do not like these mixed porridges, especially the ones containing *omena(or dagaa)*and therefore they do not eat adequate amounts. This is manifested in the fact that they struggle to feed the children, sometimes with dire consequences as reported by the public health officer.

"When omena is used in the porridge, unabahatisha (it's a matter of trying your luck). The children have to be forced to take it, and sometimes they choke"

Irish and sweet potatoes were reported to be commonly consumed by young children as mashes. Some mothers enrich the mashes by adding soup from fish, meat or vegetable stews. *Ugali* (stiff gruel) green leafy vegetables, fish as well as bananas were indicated as the next most commonly consumed foods. Community members lamented about the high cost of fish, which made it difficult to access in adequate amounts. Other foods mentioned, though rarely consumed, include beans, fruits, green grams, commercial infant formula, milk, black tea, chips, juice and biscuits.

Knowledge on Foods for Anaemia Prevention

Green leafy vegetables and beans were reported by most groups as iron-rich and relatively affordable sources of dietary iron. Green leafy vegetables reported included *Corchorusolitorius* (apoth), *Amaranthusblitum* (ododo), *Cleome gynandra*(saga), *Cucurbita maxima* (pumpkin leaves) and spinach (*Spinaciaoleracea*). Other plant based foods like beetroots (red variety) were also mentioned. The participants did not seem to know much about the degree of bioavailability of iron from the foods. Both community members and KI's reported that foods like meat, fish and liver which are good for anemia are rare in the diets of young children since they are expensive and unaffordable. They indicated that meat is sometimes consumed once a week. In the past or traditionally animal blood has been used to treat those who lacked blood; this was indicated in all the group discussions. Senior citizens were more knowledgeable on the methods of preparation of blood for consumption. Mothers also reported that sodas may be rich in iron since they have observed on several occasions that people are rewarded with sodas after blood donation. Table 2.2 summarizes the foods reported as iron rich and that are suitable for blood improvement as reported by participants.

Table 2.2: Special Foods for Blood Improvement and anaemia support

Food item	CHW's	Mothers	Fathers	Senior citizens
Liver	√	√		√
Beans	√	√	√	√
Green leafy vegetables	√	√		√
Chicken	√			
Water melon		√		
Omena		√		
Soda		√		
Citrus fruits			√	
Animal blood		√	√	√
Wild fruits			√	
Beetroot			√	
Fish				√
Green grams				√
Eggs				√
Bovine offal's				√
Composite flour porridges				√
Milk				√

CHWs: Community Health Workers

Interventions for Anaemia

The results of the discussions and interviews indicated that there are no specific nutrition interventions which target anaemia in young children in the study area. The existing interventions are on the general control and management of diseases, such as malaria management, deworming and public health education.

The Provincial Nutritionist had this to say:

"For children we have no major interventions. The use of sprinkles, started but never picked up for the community, apart from the Nyando region. Then the use of multiple micronutrient swallow tablets also did not pick up".

The Provincial Nutritionist also indicated that children who are fed with fortified blended foods or Food By Prescription (FBP), appear to have lower incidences of anaemia. However the programme focuses on HIV positive children only. A major challenge expressed is the fact that the FBP tended to create a dependency syndrome, in addition to being donor driven and reaches only limited numbers. Specific interventions reported for anaemia are summarized in Table 2.3.

Table 2.3: Anaemia Interventions/Activities in Kisumu East Sub-County

Type of intervention	Community level	Clinical/ health centre level
Nutrition and public health education	√	√
Deworming	√	√
Mosquito net distribution	√	-
Referrals to hospitals by CHW’s	√	-
Monitoring at community level	√	-
Free malaria treatment for YC	-	√
Food by prescription	-	√
Haematinics /iron supplementation	-	√
Blood transfusion	-	√

YC: Young Children

DISCUSSION

The findings of this study, that there is knowledge of iron deficiency and anaemia in the community and that no local name exists for anaemia hence various conjugations of words are used to indicate low blood levels; are similar to findings of a survey conducted in eight developing countries in Africa, South East Asia, South and Central America and South Asia, which also found anaemia referred to as low, thin or no blood, among other terms. Anaemia was also largely associated with its symptoms such as skin pallor and body weakness in this study as in the Galloway study (Galloway *et al*, 2002).

The community has the knowledge about the main causes of iron deficiency and anaemia. They indicated this as insufficient dietary intake in quantity and quality, and malaria. Other publications and reports have similarly indicated these as important causes of anaemia in Kenya and other countries (Thurnham and Northrop, 2007; Galloway *et al,* 2002; Mwaniki *et* *al*, 2001). Apart from malaria other diseases have also contributed to the high incidence of anaemia as indicated by respondents. It must be noted that malaria prevalence in Kenya is highest in the Lake Victoria region, at 38% compared to the national level of about 5%, and this has important implications for all nutrition interventions, which must incorporate malaria control. The current trend of increasing malaria incidences, especially among young children is an issue of concern and scientists from the Kenya Medical Research Institute (KEMRI) are evaluating their data in a bid to establish the actual cause (KMIS, 2010; CDC/KEMRI, 2015).

Attribution of anaemia to causes such as sitting near a hot fire, stress and diseases that result from taboos are found not just in Kenya, but also in other developing countries. Working in the sun is perceived to sap ones physical strength and '*drink*' blood leading to tiredness, and eating lemons are believed to dry up blood (Galloway et al, 2002). These are not causes of anemia, though they are probably associated with feelings of weakness or fatigue, which are symptoms associated with anaemia.

Porridge predominantly from maize, sorghum and millet mixes was identified as the main food for young children in the study, as in other studies (Mwaniki et al. 2001, Lung'aho and Glahn, 2009). Despite health and nutrition education talks that discourage mothers from including certain mixtures in porridge flour, such as beans and groundnuts, the practice is rampant. These mixed flours increase the concentration of anti-nutrients in the diet, thus lowering the bioavailability of iron (Ndagire et al, 2015). Moreover, the porridge was reportedly often cooked for 10-15 minutes only, which does not allow for proper cooking of components in the porridge, and may cause diarrhea in children. Since porridge flours present an opportunity for iron and other nutrient enrichment, there needs to be clear knowledge and related practices on porridges. The practice of forced feeding of unpopular porridge mixes, sometimes leading to death, is completely unacceptable.

Most of the foods for feeding children are plant based. Plant based foods are known to be low in bioavailable iron due to their high content of phytates and at times, polyphenols and fibre which inhibit absorption (Allen and Casterline-Sable; 2001). Milk and fruits, which are important sources of nutrients for young growing children are noticeably limited or absent in the diets, while animal source foods like meat, were rarely mentioned. This is largely attributed to low income levels common among most families living in the study area. Other reports also indicate financial constraints as a major limitation to access of animal source foods (Drolet et al. 2012; Bwibo and Neumann, 2003). With the inhibitory cost of animal source foods, it is important for families of vulnerable children to access alternative cheaper foods to ensure sufficient intake of iron, such as bovine blood.

Most participants had no knowledge about anti nutritional factors or their role in iron absorption. However, the fact that citrus and wild fruits were mentioned by fathers of young children indicates that there is at least some knowledge of the importance of vitamin C in promoting iron nutriture. Beetroots, though not rich in iron, were perceived to be rich in iron because of the red colour, which tends to be associated with presence of iron. The practice of giving sodas to blood donors has also led to some misconceptions that soda must be good for boosting blood; this misconception was also noted in the study in several developing countries (Galloway et al, 2002).

There were no specific interventions targeting nutrition of children regarding anaemia, and only mentioned curative interventions. Some nutrition education is given to mothers when they go to the clinic; but this education is based on general nutrition and childcare. In this study mothers were implicated in contributing to anaemia through laxity in childcare. This may reflect the extent of changes in socio-cultural practices, where mothers, who are important in childcare, are involved in income generation throughout the day and have limited time for their households and children. This makes the children vulnerable and the main victims of malnutrition. The role of fathers is a factor that is not clearly addressed.

CONCLUSIONS AND RECOMMENDATIONS

Conclusions

There is awareness on iron deficiency and anaemia in the community. Iron-rich foods are locally available, though expensive. Children are fed mainly with cereal gruels with little addition of iron rich foods.

Recommendations

The iron status of children needs improvement. There are locally available iron-rich foods, however, research is required to determine how these foods can be effectively incorporated in their diets. The results indicate porridges may have the highest potential for fortification with iron to alleviate iron deficiency and anaemia in children.

CHAPTER 3

KNOWLEDGE ATTITUDES AND USE OF BOVINE BLOOD AS FOOD IN KISUMU COUNTY KENYA

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Knowledge, attitudes and use of bovine blood as food in the Lake Victoria region of Kenya

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ABSTRACT

Anaemia is a leading cause of morbidity and mortality in Kenya. Around the Lake Victoria region anaemia is a leading cause of death. The main underlying causes of the anaemia are inadequate dietary intake of iron, and the prevalent holoendemic malaria. A potential, readily available source of iron is animal blood, which is rich in haem iron. Animal blood is part of many traditional diets in Kenya. However, blood from abattoirs is underutilized. This study was designed to determine the knowledge, attitudes and practices on the use of bovine blood as food, with a view to using it to enrich diets for children. Qualitative methods that included key informant interviews, focus group discussions, free listing and observations were used to collect the information on utilization in local food preparations. The focus groups included mothers and fathers of the young children, senior citizens aged 55 years and above and community health workers. The key informants included the Provincial Nutritionist, District Nutritionist, District Clinical Health Officer and District Veterinary Health Officer. Results showed that the community has a long tradition of use of bovine blood as food and methods of preparation are well known, although there were a few negative cases, which were based on religious convictions. Five main methods of food preparation which incorporate blood were identified. Two of the methods combined bovine blood with cow's milk, two methods involved cooking blood with onions and tomatoes and the fifth method involved use of blood in a mixture with tripe to prepare a sausage-type product. There were a few cases reported where mothers give raw blood to supplement prescribed foods. Results conclude that bovine blood is well-accepted and incorporated in food using several traditional methods and could therefore be used as a component in developing iron-rich food to alleviate iron deficiency.

INTRODUCTION

In Kenya iron deficiency remains a serious public health concern. Results of the national micronutrient survey in 1999 revealed that iron deficiency is widespread in Kenya, with the moderate and severe forms being more common than mild forms of the deficiency (Mwaniki *et al,* 2001). According to the study approximately60% of Kenya's population was iron deficient, with the largest prevalence being among children and pregnant and lactating mothers.

Iron deficiency is caused mainly by inadequate dietary intake of iron and losses related to parasitic infestation and recurrent disease infections. Most of the iron consumed by Kenyans is from non-haem, plant sources. Apart from anaemia, iron deficiency also has long-term negative effects on children's cognitive development.

Iron deficiency interventions targeting young children are recommended as they have the potential to improve iron status, and thus enable children to develop optimally and live healthy productive lives. Food based strategies, which include supplementation; fortification and dietary diversification have received particular attention due to their sustainability. These approaches, also referred to as dietary diversification and modifications, aim at increasing production and therefore improve on availability and access to food high in iron through agricultural production, social marketing for behavior change, processing and preparation of foods to increase bioavailability, food combinations and bio-fortification(Ruel, 2001; Kennedy et al,2003; Allen, 2003; Allen and Gillespie, 2003) Experience has shown that although each one of these strategies contributes to the control of micronutrient deficiencies, no single strategy can do the job alone (UNICEF and MI, 2008; Zimmermann, 2007).

Dietary iron can be obtained from plant and animal foods, but evidence indicates that iron from animal sources manifests better absorption compared with plant sources (Allen 2003; Bwibo and Neumann, 2003). Unfortunately, the most vulnerable and malnourished groups cannot afford sufficient amounts of the animal source foods (Ruel, 2001).

Bovine blood is rich in haem iron as well as protein, calcium and phosphorus. The high nutrient content and the desirable functional properties of bovine blood, has in the recent past led to increased interest in its possible use in food formulations. Furthermore most of the blood from public and private abattoirs goes to waste and is discarded. Food scientists from the Russia's Voronezh State Technological Academy established that bovine blood is rich in iron, and that the proteins are assimilated by the human body twice as fast as those of chicken-egg (Mosnews,2004). Other studies, such as the study by Ofori and Hsieh (2011), have also confirmed these high iron and protein contents (Walter *et al* 2001; Kikafunda and Sserumga 2005). Use of animal blood for food is a culture that is practiced by several communities in various parts of the World (Ofori and Hsieh , 2011; Ofori and Hsieh 2012; Kikafunda and Sserumaga, 2005) and in Africa, it is most common among pastoral communities.

In Kenya, cow's blood is often consumed alone or as a mixture with milk, by the Maasai people. Other communities also consume blood, incorporated in various food preparations. An example is the *mutura* (African sausage), commonly prepared and consumed by the people of Central Kenya. *Mutura* contains blood as one of the ingredients in the sausage filling. The other ingredients include assorted offals, such as intestines and stomach (of goat) mixed with onions and pepper. These are cooked together and stuffed in a section of the large intestine as casing. The ‘sausage’ is then roasted or grilled for eating. Food avoidances worldwide most frequently relate to animal source foods (Hartog *et al* 2006). In Kenya, some communities or people shun blood as food, on the basis of religious convictions or religious taboos.

This study was therefore designed to assess the knowledge, attitude to and use of bovine blood as human food, with a view to using it to enrich food with iron in an area of Kenya that is characterized by a high prevalence of iron deficiency, especially in children.

METHODS

Key informant interviews and focus group discussions were held in Kisumu East District, now referred to as Kisumu East Sub-County, which is located in the Lake Victoria region of Kenya. Observations were done in slaughter houses located in Kisumu and the environs of Nairobi (Dagoretti and Athi River). Qualitative techniques used to collect data included Key Informant Interviews, Focus Group Discussions and observations. Approval for the study was granted by the Ethical Committee of the Kenyatta National Hospital and the University of Nairobi, and the Nyanza Provincial Nutrition Office. Informed and signed consent was obtained from all KI and FGD participants.

Key Informant Interviews (KII): Four key informants were interviewed consisting of the Nyanza Provincial Nutritionist, the Kisumu East District Nutritionist, the District Clinical Health Officer, and the District Veterinary Officer. A standard interview tool was used to collect the information. The questions for the first three key informants were similar while the one for the veterinary officer was designed to capture data on bovine blood availability and hygiene.

Focus Group Discussions (FGD): Four focus group discussions were held in Manyatta sub-location of Kisumu East sub-county. Manyatta is the second largest informal settlement in the County, with a high population of young children. Their views were assumed to be representative of the views of the region. Structured interview guides in English and translated into the local dialect, *Dholuo* were used to collect information from the focus groups on bovine blood as food, knowledge, availability, uses and challenges. Characteristics of participants are summarized in Table 3.1

Table 3.1: Characteristics of FGD Participants

Group	N	Age range (yrs)	Occupation	Percent
Fathers	9	25-42	Informal/self employed	78
			Employed	22
Mothers	12	17-30	Informal/self employed	75
			Employed	17
			Housewife	8
Senior citizens	12	55-75	Informal/self employed	66
Males 58%			Housewives	17
Females 42%			Retired	17
CHW's	11	28-45	Community health work	11
Males 27 %				
Females 73 %				

Observations: Observations were made during visits to slaughter houses in Athi River (Kenya Meat Commission), Dagoretti Corner and Kisumu slaughter houses.

Data analysis: The data were recorded, transcribed and uploaded in *Atlas ti* software for coding and subsequent analysis. Information obtained from the key person interviews was collated using methodological triangulation with that derived from the focus group discussions to describe the emerging themes.

RESULTS

Knowledge of Bovine Blood as Food

All participants indicated that bovine blood is generally accepted and consumed in the community; however its consumption has decreased greatly over time. There was also a general consensus that bovine blood consumption is higher in rural areas than in Kisumu city. Bovine blood is a cherished cultural food, which was prepared for significant gatherings where there was an animal slaughtered. Animal blood was also used as traditional treatment for anaemia or to improve blood status, especially among mothers' post-partum and young anaemic children. Some families were reported to still use this method to treat their children. According to the District Clinical Health Officer of Kisumu East Sub County hospital:

"Cow's blood! They fetch. Even from the ward. They run to the slaughter house and go fetch it. The time I used to do ward rounds in ward two, paediatric ward, they actually used to come with it raw! And the child is given the way it is"

According to the senior citizens and the veterinary officer, excess bovine blood, which was not utilized after slaughter, was traditionally preserved for later use. Fresh blood was left to settle and the resulting serum drained off. The clotted blood was then spread out in small lumps on an iron sheet *(mabati)* to dry in the sun. It was dried until it became hard. Then it was placed in a gourd, the gourd tightly closed and stored. This form it would remain preserved for long periods for later use. Participants also indicated that chicken, sheep and goat blood are also consumed in the community. Chicken blood is commonly added to chicken stew during cooking, after home slaughter. It coagulates in the soup and is often served to young children. Goat and sheep blood are collected during slaughter and prepared like fried bovine blood.

Attitudes Related to Bovine Blood Use

The predominant attitude to bovine blood use as food was positive and respondents indicated that it is a valued and tasty traditional dish. However, religious prohibitions and allergic reactions to bovine blood consumption were indicated by some respondents. Religious prohibitions in the area mainly affect members of the Muslim faith, Seventh Day Adventist (SDA) and an independent African church (*Akorino*). However, there is reportedly a low prevalence of members of these faiths in the study area.

Availability and Use of Bovine Blood as Food

Bovine blood availability: Bovine blood is available in Kisumu East Sub-County from Mamboleo and Rabuor slaughter houses. Another two slaughter houses in Otonglo and Kiboswa are relatively close in bordering sub-counties. Senior citizens who were more conversant with bovine blood reported that:

"We get it from the slaughter house in Otonglo, or any other slaughter houses near, like Rabuor or Mamboleo"

The District Veterinary Officer in Kisumu estimated that an average of 150 - 200 litres of bovine blood is discharged daily from each of the slaughter houses, and approximately 80-90% is discarded and the remainder of 10-20% is collected and used by community members. Observations made during slaughter house visits also indicated that bovine blood was most frequently collected by mothers of young sick children, women who have recently given birth, bar or food stall employees, livestock farmers (for animal feed) and a few individuals involved in scientific research. Blood from goats, sheep and chicken slaughtered in Mamboleo all goes to waste. The veterinary officer indicated that home slaughter of cows during gatherings is a common practice though there is no data on actual numbers slaughtered. It is a significant source of bovine blood which according to the veterinary officer, rarely goes to waste in the region, as most of the blood is collected and used for human consumption. The interviews also determined that bovine blood collected from slaughter houses in Kisumu is usually at no cost at all, or at low cost.

The District Veterinary Officer's view is quoted below:

"We can't say there is a cost as such, as the owners of these animals do not count blood as part of their profit. It is a product which, I believe, if somebody starts, maybe a factory is when they will wake up and know that there is a cost. The boys who collect it for the mama's who go there, maybe they get ten or twenty bob, it's not something you can say has a fixed cost. It is quite cheap"

Observations and informal discussions with the chief veterinary officer at the Kenya Meat Commission (KMC) indicated that the largest meat plant in Kenya, the Athi River slaughter house disposes of approximately 600 litres of bovine blood daily, all of which goes to waste except when there are orders from animal feed manufacturers. In Dagoretti, a conglomeration of slaughter houses, where most of the meat consumed in Kenya's capital, Nairobi, comes from; the scenario is similar to that in Athi River. Most of the bovine blood goes to waste.

Challenges related to blood use: According to the Veterinary Officer the main health challenges of cattle in the area are tick borne diseases, due to inadequate spraying of livestock. Cases of anthrax in the locality are not common and there had not been any incidence in five years (2007-2012). Animals at the slaughter house are procedurally checked by qualified inspectors to ensure good health status (ante-mortem) before slaughter.

Information on how blood from slaughter houses in the area is disposed of was not readily available. In Mamboleo slaughter house, Kisumu, the blood is drained into blood tanks, and then later disposed of, but precisely how, was not indicated. From group discussions in this study and earlier personal communications, the indication is that disposal of blood from slaughter houses is a major challenge, especially in the bigger slaughter houses, where environmental pollution was evident.

Loss of skills in bovine blood preservation and preparation among mothers of young children has contributed to reduced consumption according to fathers of young children, senior citizens and key informants. Most senior citizens indicated that they sometimes prepare bovine blood dishes, for their own consumption. However the response given by fathers of young children when asked who cooks the blood at home was

"My mother at home. My wife does not know how to cook blood"

Practices related to bovine blood preparation are described are summarized in Table 3.2

Table 3.2: Dishes Prepared Using Bovine Blood

Local name	Ingredients	Method	Accompaniment
***Riga* (or *origa*)** with fresh blood	-Fresh bovine blood -Fresh bovine milk -Ghee or *mor dhiang* (optional)	- Allow blood to stand till it clots. - Use *osinde*(a local grass) or millet stalks to mix blood. This results in smaller lumps. - Add fresh milk and stir - Boil while stirring till cooked. It will be dark in colour and resemble porridge in consistency - Ghee may be added if desired -Consumed alone or with accompaniment.	- Sweet potatoes - *Ugali - Cassava
***Riga* with dried blood**	-Dried bovine blood. - Fresh cow's milk. -Ghee (optional)	-Grind dried blood using grinding stone. - add fresh milk, while stirring and boil till ready. -Serve alone or with accompaniment	
Atururu	-Fresh blood - bean ash - fresh milk - cooking fat	- boil fresh blood in shallow pan while stirring continuously. - add a little ash - add fat and continue stirring till ready - Serve hot with accompaniment	
Fried bovine blood	-Fresh bovine blood -Onions. - Tomatoes - Cooking fat	-Heat fat in pan - add chopped onions, tomatoes and allow to cook. - pour in blood and turn till it resembles liver - Serve hot with accompaniment.	Ugali
Mutura (African sausage)	-Section of Intestines. -Chopped offals (fresh or precooked) - fresh bovine blood - salt	- Chop offals for filling and mix - Add salt if desired - Add fresh bovine blood and mix - Stuff intestines with offal mix - roast over hot charcoal fire till cooked - Consumed alone or with accompaniment	- Ugali

**Ugali is a stiff maize porridge (It may also be prepared from sorghum, millet or cassava)*

Origa (or *riga*) and *atururu* are traditional dishes prepared using bovine blood. Other dishes are prepared by frying onions and tomatoes in oil, then adding blood. These are modified dishes, in line with modern cooking. *Mutura* was reported to be quite popular especially among men, and is prepared and sold mainly as a street food and in bars. Preparation of *mutura* has been largely borrowed from Central Kenya.

DISCUSSION

The first and important finding is that animal blood is recognized and accepted as a food that boosts the amount of blood an individual has and can be used to treat anaemia (Andago *et al*, 2015). Senior citizens appeared more knowledgeable about bovine blood use compared to other groups, with the Community Health Workers (CHW's) appearing to be least knowledgeable. It may also be that CHWs felt their professional role was not compatible with supporting a traditional practice; or they may also be out of touch with traditional practices, having spent most of their growing years in educational institutions. The extensive knowledge regarding use of animal blood among senior citizens is an indication that there was a well-established traditional knowledge and skills related to bovine blood use as food. Although senior citizens may not know the actual content of iron in bovine blood, research indicates that the iron content of raw blood is indeed high at 18.8 mg per 100g of the edible portion(Sehmi, 1993); while in cooked and dried powder form the iron content increases to 195.46 mg/100g (Kikafunda and Sserumaga,2005). There are no indications that iron content decreases during cooking, except in preparations where milk is added (Sehmi, 1993), this is due to the binding effect of calcium from milk on iron. Interestingly this interaction may to some extent explain why there are no known cases of iron overload in communities consuming blood with milk on a daily basis, like the Maasai of Kenya.

A second important finding was that most of the bovine blood produced in slaughterhouses in the country is discarded. Discarding animal blood from the slaughter houses is the prevalent practice in several countries and that the main concerns are around environmental impact (Mosnews, 2004; Walter *et al* 2007; Ofori and Hsieh, 2011; Ofori and Hsieh 2012; Kikafunda and Sserumaga, 2005). Inappropriate disposal leads to blood coagulating in drains where it putrefies, causing bad odours and sanitary and environmental problems (Ofori and Hsieh, 2012; FAO, 2015; Koech *et al.*, 2012). In some countries these concerns have led to research and development of interventions which give value to bovine blood through food quality improvement and anaemia reduction interventions. Walter *et al.*(2001) reported a significant reduction in anaemia prevalence among school children, after bovine blood fortified cookies were served for the national school/lunch breakfast programme in Chile(Walter *et al.* 2001). This demonstrates that in regions where bovine blood is accepted as food, it is possible to design an intervention to reduce anaemia among children (Walter *et al*, 2001). Ofori and Hsieh indicate in their review on bovine blood products, that bovine blood and derived products are used not only as additives, in meat and non-meat industries, but also in the production of iron supplements for anaemia control (Ofori and Hsieh, 2012).

In this relatively rapid assessment we were able to identify five different recipes that use animal blood. This is a clear demonstration of the widespread acceptance and integration of bovine blood as food in this community. This finding is consistent with the results of the Kenya National Micronutrient Survey, which lists bovine blood as one of the semi-solid foods introduced in infant diets in Kenya, with an average consumption rate above the national average in the Lake Victoria region (Mwaniki *et al*, 2001). These traditional recipes maybe an important platform for marketing animal blood, as a rich and easily accessible source of haem-iron at household level, or as a cottage industry. Acceptance of bovine blood as food has also been found in other regions of the world, as manifested by the existence of dishes and recipes which incorporate blood in cooking, ranging from various black puddings and sausages in Europe and North America to blood tofu in China (Mosnews, 2004; Ofori and Hsieh, 2011; Ofori and Hsieh, 2012; Kikafunda and Sserumaga, 2005).

Among young mothers, knowledge on preparation methods was found to be weak and the practice of giving hospitalized children uncooked blood to treat anaemia could be detrimental. Although comparative documented literature on use of raw bovine blood to treat anaemia is not available, drinking raw blood is known to be accepted in many cultures. Thus this practice may be largely based on advice from older mothers and grandmothers, if this was one of the traditional ways that anaemia was addressed.

Challenges related to bovine blood consumption were found to be few despite religious prohibitions. Some participants indicated that they consumed bovine blood, because they were accustomed to taking it as young children. They believe it is nutritious and liked the taste. The information gathered from talking to a cross section of population and the existence of traditional methods for preparing and preserving bovine blood is a major strength which increases the likelihood of a well-designed anaemia intervention using bovine blood being accepted in the community.

CONCLUSIONS AND RECOMMENDATIONS

Conclusions.

The study shows that the majority of the respondents were knowledgeable on the use of bovine blood as food and methods of preparation for consumption exist, even though knowledge and use have reduced considerably over the years hence this cultural information may not be available for future generations, unless preserved and practiced. Any negative attitude towards consumption of blood was low and mainly based on religious convictions. Since bovine blood is rich in iron and most of it goes to waste, an important opportunity is presented for nutritional and iron improvement in children's diet.

Recommendations

Investigations on bovine blood and how it can be effectively incorporated in diets to reduce anaemia is required is important. The findings of study have been localized to population in a specific region. Iron deficiency anaemia is prevalent throughout Kenya. Since there is great population and ethnic diversity within Kenya, further studies are required to determine acceptability bovine blood use in infant diets.

CHAPTER 4

DEVELOPMENT OF A BOVINE BLOOD ENRICHED PORRIDGE FLOUR FOR ALLEVIATION OF ANAEMIAAMONG YOUNG CHILDREN IN KENYA.

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Development of a bovine blood enriched porridge flour for alleviation of anaemia among young children in Kenya.

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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 of 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 powder, 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 value, sensory acceptability and shelf life stability. Sensory acceptability was assessed using a laboratory panel in a 7- point hedonic rating scale for the attributes of colour, taste, mouth-feel and overall acceptability. Shelf life stability was evaluated by storing the products in gunny bags, polythene bags, Kraft paper and plastic jars each at 25ºC and 35ºC. The most acceptable mix contained flours of sorghum, finger millet, cassava, and blood powder in the ratios of 6:14:10:0.5 and provided 54% of the recommended daily intake of iron for 1-3 year old children, when consumed as a cup (250 mls) 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 concluded that acceptable and shelf-stable porridge mix can be formulated from local cereals and cassava flour with iron enrichment from bovine blood powder.

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 prevalence is even higher 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.

MATERIALS AND METHODS

Composite Flour Ingredients and Preparation

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

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, quickly flashed 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 to await processing and analysis.

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 cereal flours. The milled cassava was stored in similar containers and temperatures as the other cereal flours.

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 pass through 150µ sieve and stored in sealed zip-lock polythene paper bags, prior to further analysis.

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 guide formulation of the composite flours.

Hydrogen Cyanide

The Hydrogen Cyanide (HCN) content was analysed according to the Association of the Official Analytical Chemists methods (AOAC, 1984). Approximately 20g of cassava flour were mixed with 200ml 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.

Proximate Composition

Moisture, crude protein, crude lipid, crude fibre and total ash were analysed using AOAC methods (AOAC, 1980). Soluble carbohydrates were calculated by difference. The moisture content of cereal flours and blood powder were determined by drying at 105^0C in an air oven to constant weight. Total energy was calculated from fat, carbohydrate and protein contents using Atwater’s conversion factors of 7.0, 4.2 and 4.2 respectively. 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$\equiv$ 0.0282g oleic acid).

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).

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 250ml 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 determined using Nutrisurvey.

Porridge Preparation and Sensory Evaluation

Porridge was prepared from each of the 5 (CF1-CF5) 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 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.

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).

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 different 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 methods (AOAC 1980). Shelf life 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.

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 $P \leq 0.05$. Microbiological counts were transformed into log number and analysis of variance (ANOVA) performed on log count.

RESULTS AND DISCUSSION

Hydrogen Cyanide Content of Cassava Flour

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

Proximate Composition of the Flour Ingredients

The results of proximate composition and energy analyses of the composite flour ingredients are shown in Table 4.1. 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% moisture content recommended for optimum storage of cereal flours (KBS, 2003). Bovine blood powder had a moisture content of 8.5% which is comparable to that reported by Kikafunda and Sserumaga of 6.85 % (Kikafunda and Sserumaga, 2005). The blood powder had a higher protein content at 80g/100DM compared to the cereal and cassava flours whose protein content ranged from 8.5g/100DM (in maize meal) to11.81g/100DM (in millet flour).

The protein content of the bovine blood powder was higher than the protein content of bovine liver of 17mg/100g (Sehmi, 1993), which is 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 blood powder (4.64g) was lower than the carbohydrate content of 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 densities and digestibility, as opposed to high dietary fibre content which increases bulk and satiety while reducing digestibility (MIYCN working group, 2009).

Table 4.1: 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]	8.1±0.1[a]	4.6±0.3[d]	2.3±0.0[c]	1.3±0.0[a]	77.8	379
Millet flour	7.0±0.1[a]	11.8±0.6[b]	3.5±0.2[c]	2.8±0.0[d]	5.2±0.1[c]	72.9	358
Cassava flour	7.0±0.3[a]	8.9±0.3[a]	1.1±0.1[a]	1.5±0.0[b]	2.3±0.0[b]	80.2	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±0.0 [b]	64.7	331
Blood powder	8.5±0.0[b]	80.1±0.0[c]	2.5±0.3[b]	0.2±0.0[a]	3.9±0.0[d]	4.6	362

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

Table 4.2: 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	Trace	Trace	4.1±0.09	0.52±0.0	1.6±0.1	trace
Millet flour	207.2±4.8	102.3±14.4	8.6±0.8	9.5±0.1	0.53±0.0	0.9±0.0	trace
Cassava flour	484.7±63.8	82.9±4.6	Trace	5.4±0.0	trace	0.8±0.1	trace
Maize flour	330.0±31.1	Trace	Trace	1.5±0.0	0.50±0.0	2.5±0.1	trace
Bovine blood	210.0±10.5	Trace	Trace	121.0±5.6	trace	trace	trace

Trace = insignificant amounts

Mineral Content 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 4.2. The iron content of the bovine blood powder at 121.0±5.6mg/100g was 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 have 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.

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 4.3. 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 4.4. 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 4.3: 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 1-3 years (%)	RDA 4-6 years (%)
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

Iron RDA for 1-3 years= 7mg/day Iron RDA for 4-5 years is 10 mg/day

Table 4.4: 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

CHO: Carbohydrates *Nutrisurvey data has no RDA values for these nutrients

Selection of the Most Preferred Composite Flour

The results of sensory evaluation of the porridge from the five composites are shown in Table 4.5. All the porridges scored more than 4.0 out of a maximum of 7.0 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 4.5: Sensory Attribute Mean Score of the Composite Flours

Sample	Colour	Taste	Mouth feel	General acceptability
CF1	4.8 ± 1.3^{ab}	5.0 ± 1.2^{ab}	5.2 ± 1.4^{ab}	5.1 ± 1.2^{a}
CF2	4.7 ± 1.3^{ab}	4.7 ± 1.5^{ab}	4.8 ± 1.5^{ab}	4.8 ± 1.4^{a}
CF3	5.6 ± 1.4^{b}	5.3 ± 1.5^{b}	5.4 ± 1.3^{b}	5.5 ± 1.3^{a}
CF4	4.1 ± 1.7^{a}	4.0 ± 1.6^{a}	4.0 ± 1.4^{a}	4.5 ± 1.2^{a}
CF5	4.5 ± 2.0^{ab}	4.8 ± 1.6^{ab}	4.5 ± 1.5^{ab}	4.6 ± 1.6^{a}

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 for all composite flours.

CF 3 had the highest scores compared to 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.

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

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 (Figure 4.1). 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.

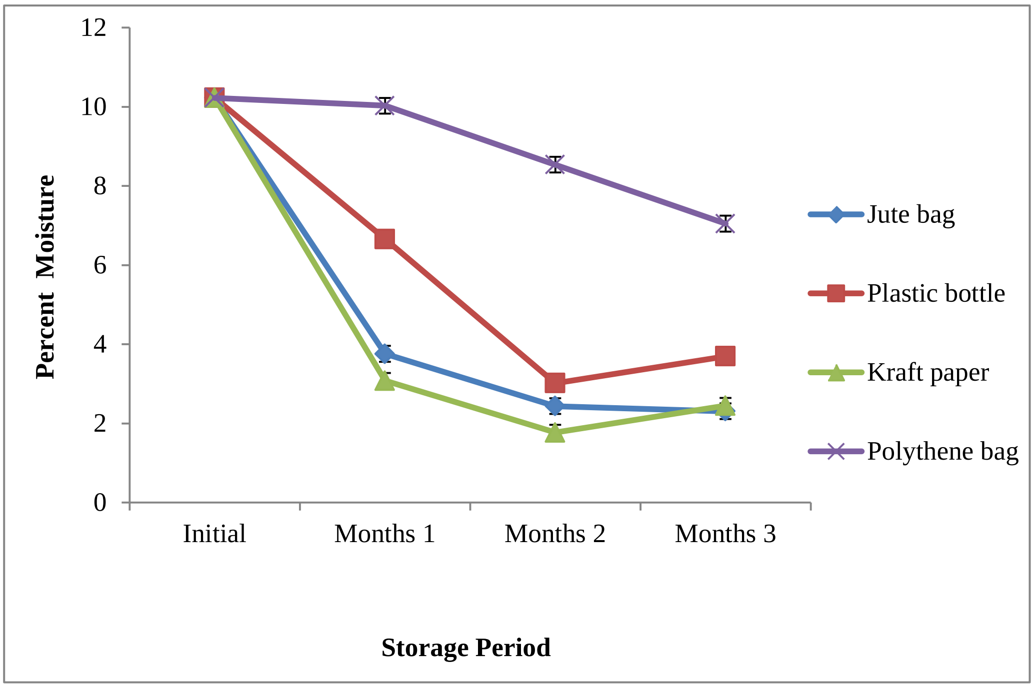

Figure 4.1: Moisture Content of Selected Composite Flour During Three Months of Storage in Different Packaging Materials

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 4.2. The composite flour showed a rapid and significant increase in free fatty acid during first 2 months of storage in all packaging materials ($p < 0.01$). 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. 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.

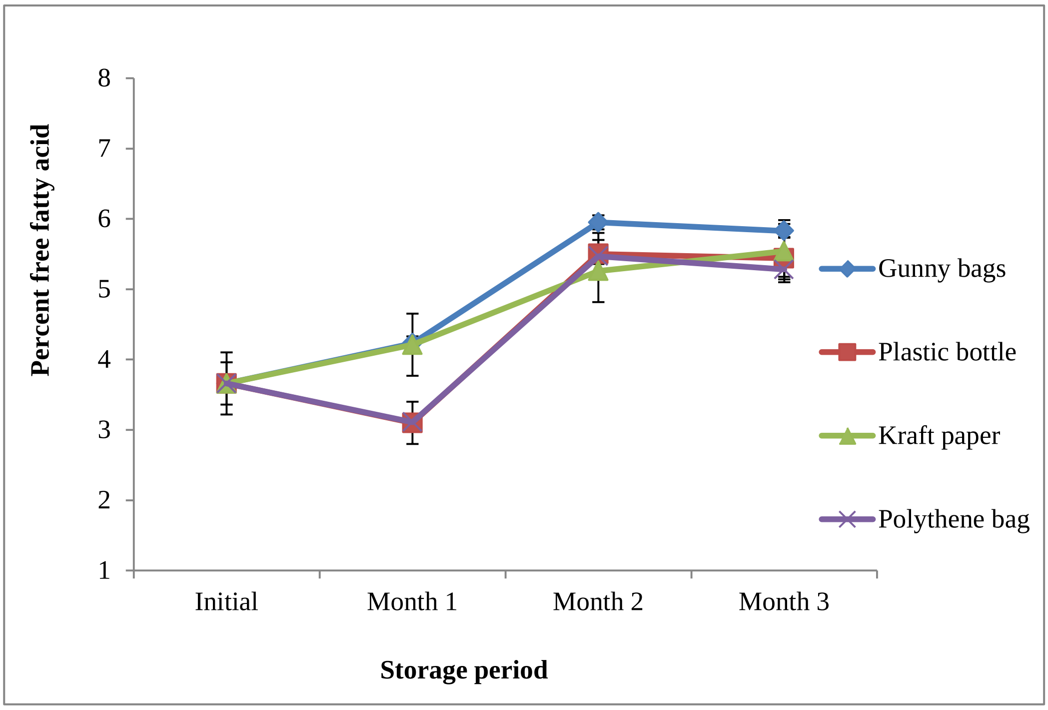

Figure 4.2: Free Fatty Acid of Selected Composite Flour during Three Months of Storage in Different Packaging Materials

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.

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 5cfu/g, log 1cfu/g and log 3cfu/g of composite flour for total aerobic count, total coliform count and yeast and moulds respectively (KBS, 2003). This was attributed to the slow sun drying rate (2-3 days), which could allow 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 4.3. The initial total viable count in the composite flour was log 6.65 cfu/g of the flour sample. These were above the recommended Kenyan standards level of 5 cfu/g for composite flour. There was apparent decline in Total Viable Counts (TVC) in all packaging materials during storage at 25°C. At 35°C there was an initial decline for the first two months, followed by an increase in the plastic bottle and gunny bag. During the third month TVC deceased further in kraft paper. 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.

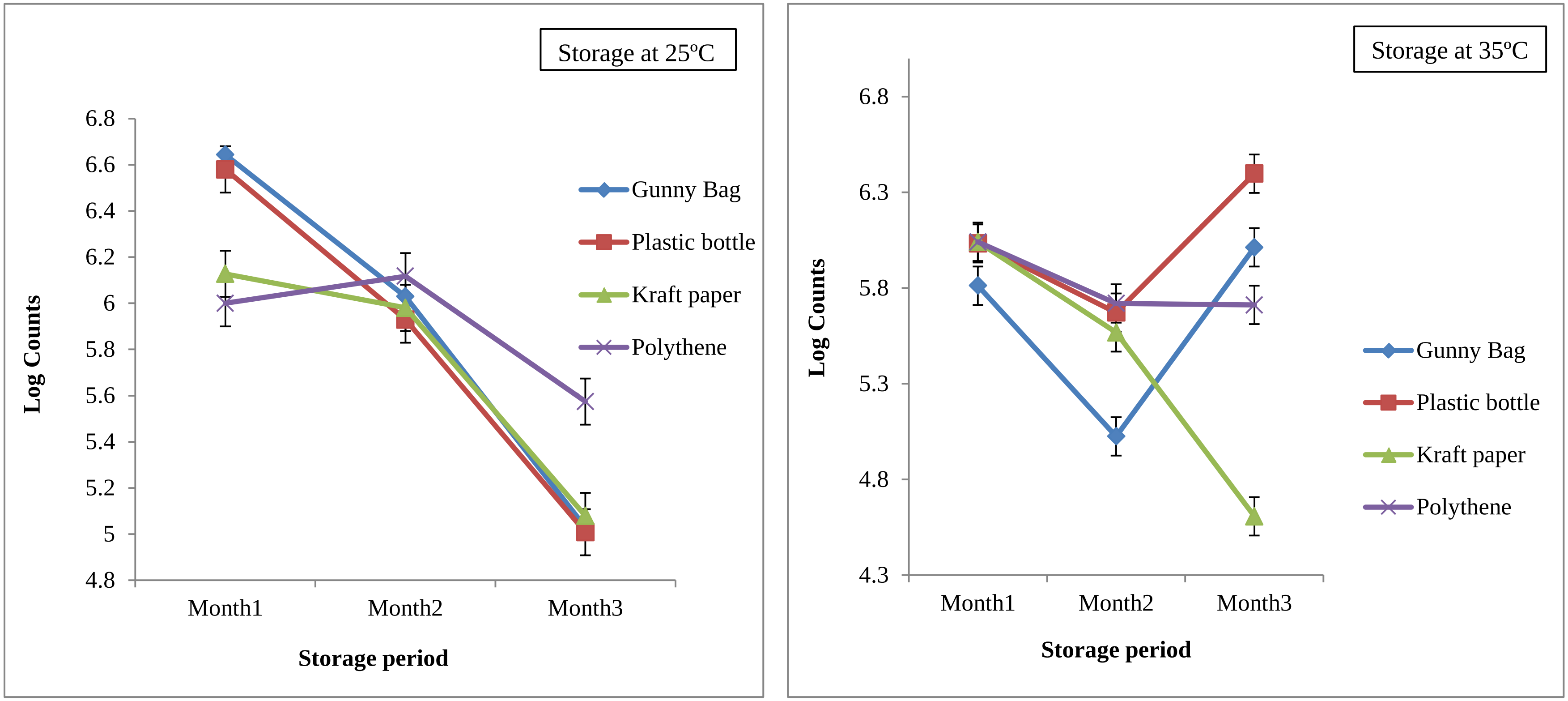

Figure 4.3: Total Viable Counts for Most Preferred Composite Flour During Three Months of Storage in Different Packaging Materials

Changes in total coliform count of the composite flour: The results for total coli forms counts are shown in Figure 4.4; the initial total coli forms count was log 4.32 cfu/g. Which were above the recommended Kenyan standards levels of log 1cfu/g gram of composite flour (KBS, 2003). Changes recorded in total coliform count in all packaging materials during the first two months of storage were not significant at 25°C and 35°C. The counts decreased by approximately log. 0.5 cfu/g, during the third month of storage at 25°C and 35°C, which was also not significant.

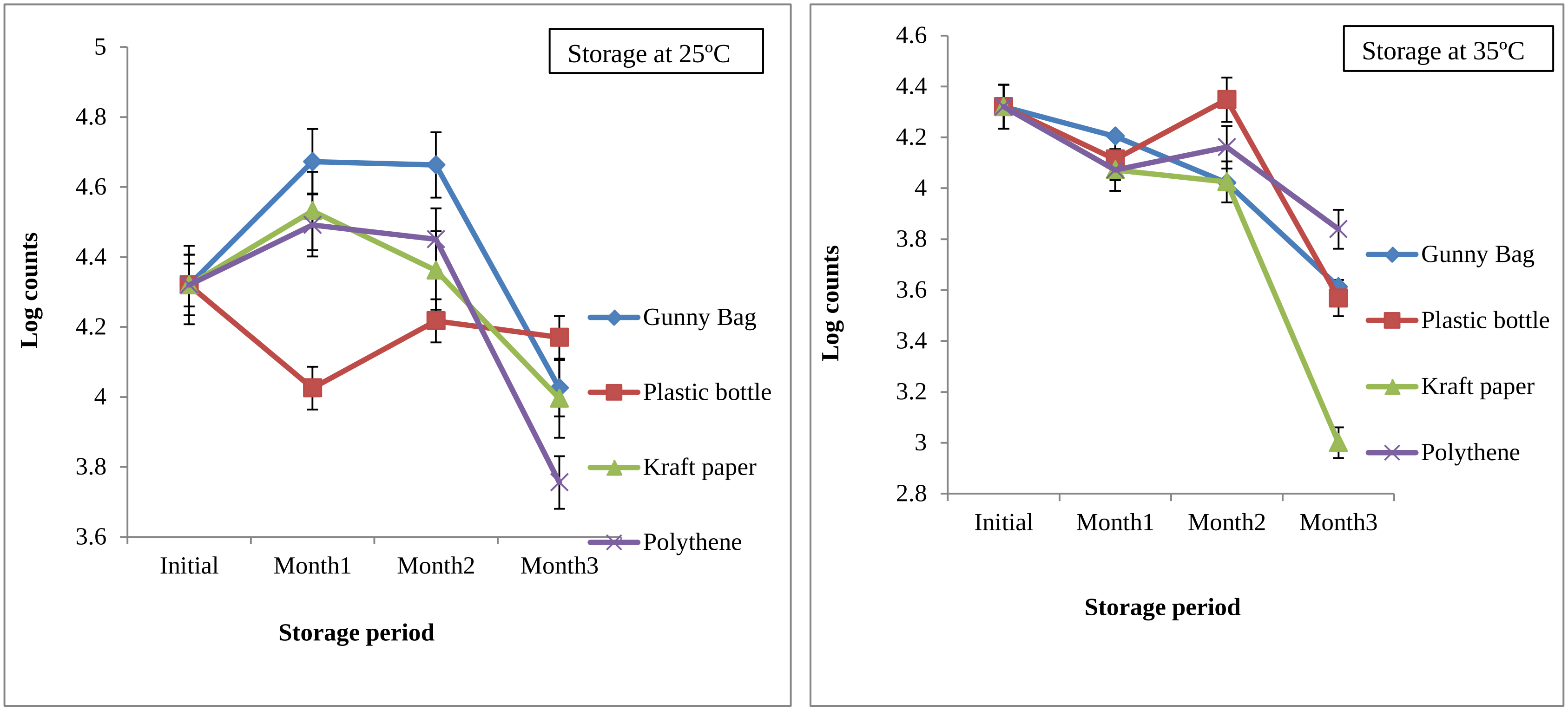

Figure 4.4: Total Coliform Counts Most Preferred Composite Flour during Three Months of Storage in Different Packaging Materials

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 and these counts were above the recommended Kenyan standards levels of log 3 cfu/g of composite flour, though the difference was not significant (p> 0.05). 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 these 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.

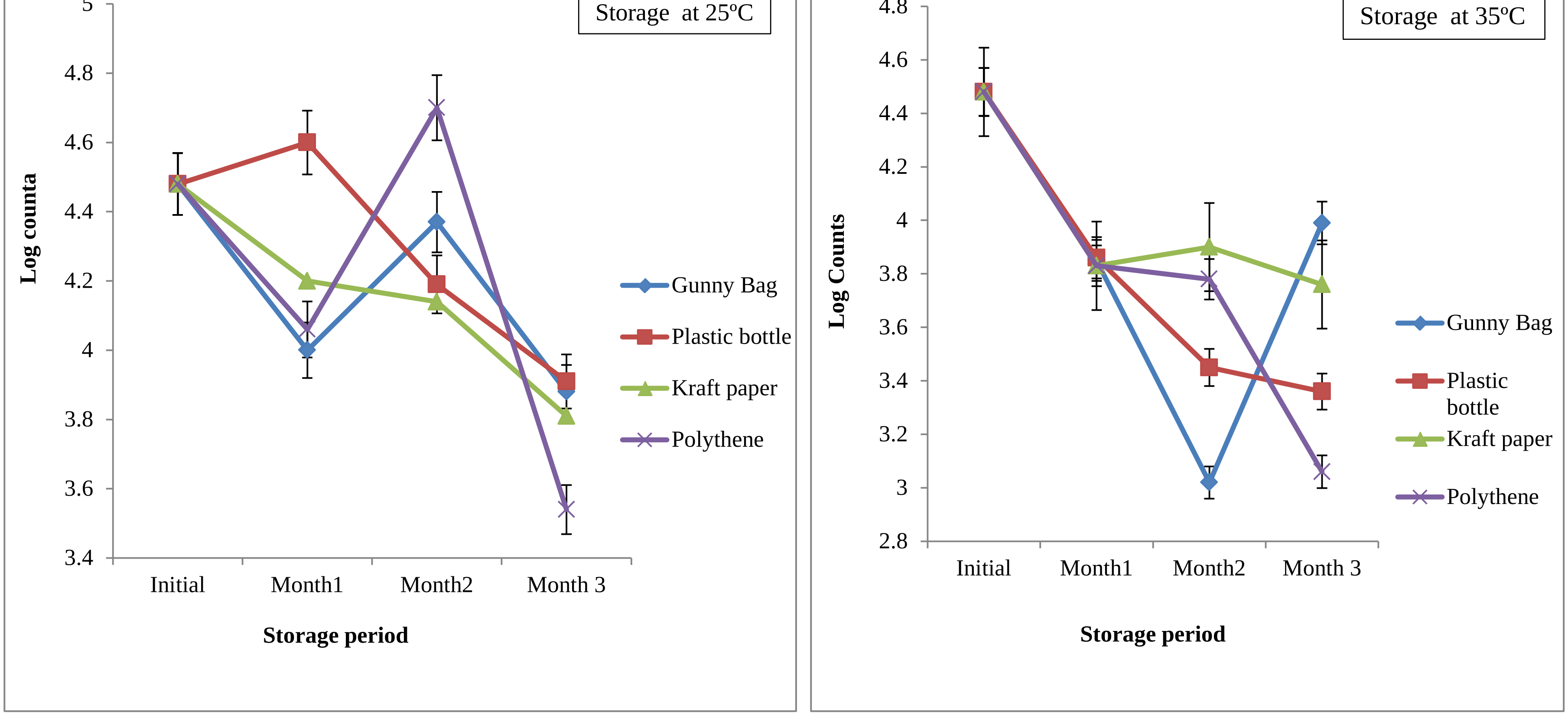

Figure 4.5: Yeast and Mould Counts of Most Preferred Composite Flour During Three Months of Storage in Different Packaging Materials

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. 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. Composite flours enriched with bovine blood must be handled using maximum hygiene to improve safety levels. Proper cooking is also essential to destroy any microorganisms. The dark colour of bovine blood powder may be a deterrent and is an important factor to be considered where it is incorporated in foods such as porridge mixes, suitable foods must thus be carefully selected.

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 are needed and work on improving shelf stability in composite flour mixes.

CHAPTER 5

FEEDING ANAEMIC CHILDREN IN A MALARIA ENDEMIC REGION OF KENYA WITH PORRIDGE ENRICHED WITH IRON FROM BOVINE BLOOD TO IMPROVE IRON STATUS

ABSTRACT

Iron deficiency and anaemia are public health problems of great concern in Kenya. The Lake Victoria basin region has one of the highest prevalence rates of anaemia, and young children are among the most affected. There are indications that dietary enrichment of the foods commonly consumed by the vulnerable groups is most effective and sustainable in improving iron status. In Kenya bovine blood is incorporated in diets by many communities. The blood is a good source of high quality heme iron, but it is usually left to go to waste in many large abattoirs. This study was therefore designed to assess the possibility of improving iron status of anaemic children from a malaria endemic region by feeding them with porridge enriched with iron from bovine blood. The porridge was formulated to contain sorghum, millet, cassava and the blood flours in the ratios of 8:8:8:1. A total of 270 pre-school aged children, selected from 5 Sub-locations of the study area had their haemoglobin, serum ferritin and soluble transferring saturation assessed. The children were also screened for malaria. Anaemic children with haemoglobin of 5.0 to 10.9 mg/dl were recruited for the study.Out of these, 102 children were selected and randomly allocated to two groups, the intervention and placebo group.Guardians of the study children were interviewed for socio demographic characteristics of the households and morbidity patterns of the children. Dietary intake of iron was assessed using 24-hour recall, on a sub-sample of 46 children. Food consumption frequency was determined on the total sample of children and used to calculate the Dietary Diversity Scores based on eight food groups. During feeding trials, the intervention group received 300ml each of the enriched porridge, while the placebo group received the same amount of the porridge without enrichment daily, at the feeding centres. Feeding was carried out for six weeks, after which the children's haemoglobin and iron status were re-assessed.

The socio-demographic characteristics of children in both intervention groups were not significantly different from each other ($p>0.05$) in terms of household size (4.7) and mean age of the mothers (27 years). The majority of the mothers had primary level education. Malaria was the most frequent illness reported (76.5%), followed by gastrointestinal tract (51%) and acute respiratory infections (47%). During screening, 10.7% of the children tested positive for malaria, which was treated before commencement of the study. Overall 44(88.9%) of the children were receiving less than the RDA for iron. The intervention and placebo groups had serrum ferritin levels of 58.71(±50.71)µg/L and 36.69 (±55.78)µg/L respectively, while the soluble transferrin receptor levels were 9.43 (±4.28) and 11.26 (±5.27)µg/L respectively. The levels in each pair were not significantly different from each other ($p> 0.05$). After the six weeks intervention there was a significant ($p= 0.001$) increase of haemoglobin of 1.161 mg/dl in the intervention group with 38% becoming non-anemic, compared to an increase of 0.111mg/dl in the placebo where 14.6% became non-anemic. The mean change in serum ferritin after intervention was not significant ($p= 0.080$) between the placebo than the intervention groups. However, the soluble transferrin receptor levels were significantly higher in the placebo and intervention group ($p>0.05$). The results show that bovine blood enriched porridge can be effectively used to improve the iron status of anemic children.

INTRODUCTION

In Kenya there is a high prevalence of iron deficiency and anaemia with young children bearing the largest burden. A national micronutrient survey showed that young children living in the Lake Victoria basin region have among the highest prevalence rates, with over 90% of children aged 2-72 months suffering from anemia (Mwaniki *et al*, 2001).

A major cause of nutritional iron deficiency is insufficient dietary intake and poor bioavailability in diets (Kennedy *et al*, 2003). Frequent exposure to inflammation and infection increases the risk of developing anaemia in infants and young children. Parasitic infections, diarrhoea, vomiting, fever, malaria and helminth infection, which are quite prevalent in developing countries also directly or indirectly contribute to anemia in children (Thurnham and Northrop, 2007). Due to increased growth and during illness, additional iron is required by young children. Anaemia often develops under these conditions due to iron deficiency, and is exacerbated when both these conditions occur together.

Food based strategies, also referred to as dietary modifications have received strong support as sustainable means of meeting the nutritional needs of populations. Although iron can be obtained from plant and animal foods, evidence from several studies, indicate a strong positive association between animal food intakes and iron status. However, the most vulnerable and malnourished groups often cannot afford sufficient amounts of these foods (Allen, 2003; IFPRI, 2001).

Blood obtained from animals slaughtered for meat is accepted as food in many communities of Kenya. Many pastoralist in the African region, such as the Maasai of Kenya and Tanzania, consume fresh bovine blood regularly (Ferraro, 2001; Oiye *et al*, 2009). Other communities in Central and Western Kenya also prepare and consume bovine blood. However documented literature is limited, though blood is considered as waste especially in the commercial abattoirs. If properly harnessed therefore, it constitutes a cheap source of nutrients such as protein and haem iron (Sehmi, 1993).

Iron deficiency interventions targeting young children are important as they have the potential to help improve iron status thereby enabling them to develop optimally and live healthier, productive lives. Children living in the informal settlements of Kisumu County were perceived to suffer from anaemia mainly due to inadequate dietary intake. This has already been reported in a publication from an earlier part of this study (Andago *et al*, 2015). The publication also confirmed acceptance of bovine blood as food in the region. This study was thus designed to assess the effect on iron and anaemia status of feeding young anaemic children with porridge enriched with iron from bovine blood, a new approach which has not been tried before.

METHODOLOGY

Study Design

The study was a longitudinal (feeding trials) and analytical (blood screening for iron status) study. The intervention was a randomized-controlled feeding trial which was partially blinded, so that those involved in the feeding could not differentiate between the enriched and placebo porridges.

Study Site

This study was based in the Winam Division of Kisumu East sub-county in Kisumu County. Kisumu East Sub-county was purposively selected due to the high prevalence of anaemia, endemic malaria and the local culture of acceptance of bovine blood as food. Winam division was purposively selected due to its central position in relation to abattoirs.

Recruitment into the Study

Screening for anaemia: Mothers of young children residing in Winam division, mainly from the informal settlements of Manyatta, Nyalenda, Migosi, Nyawita and Obunga sublocations were sensitized for the study with the help of community health workers. The mothers brought their children to a central place for screening (Kolping Centre, Nyawita). The children who met the inclusion criteria for age (12 to 60 months), with no chronic or underlying illnesses and who had resided in the area for at least six months and whose parent or guardian gave verbal and written consent were screened for anaemia status and malaria. Children were also clinically examined by a paediatritian, to ensure they met the inclusion criteria before collection of blood samples. All children whose haemoglobin levels fell within the range of 5.0-10.9mg/dl were eligible for the study. For households who presented more than one child only one child was accepted for the study. A total of 270 children were screened over a period of five days.

Sample Size Determination

The mean Hb obtained during screening (9.176mg/dl ±1.6) was used to determine the sample size for the intervention phase. Using the formula which is recommended for such interventions by Magnani, a total sample size of 102 (51children per intervention group) was adequate to detect an improved haemoglobin concentration of 1.25g/dl (±18.0), assuming a power of 80% and an attrition rate of 10 % (Magnani, 1999).

$$n = \frac{[(Z_{\propto}+Z_{\beta})^{2}\times(sd1^{2}+sd2^{2})]}{(x_{2}-x_{1})^{2}}D$$

Where:

Z_{α} = 1.645 for α at 95% confidence interval

Z_{β}= 0.840 for β at 80% confidence interval

x_1 = sample mean at baseline 9.176 with a standard deviation sd_1 of 1.6

x_2 = sample mean at endline expected to give an increase in mean Hb of 1.25g/dl with a standard deviation $sd_2$of 1.8

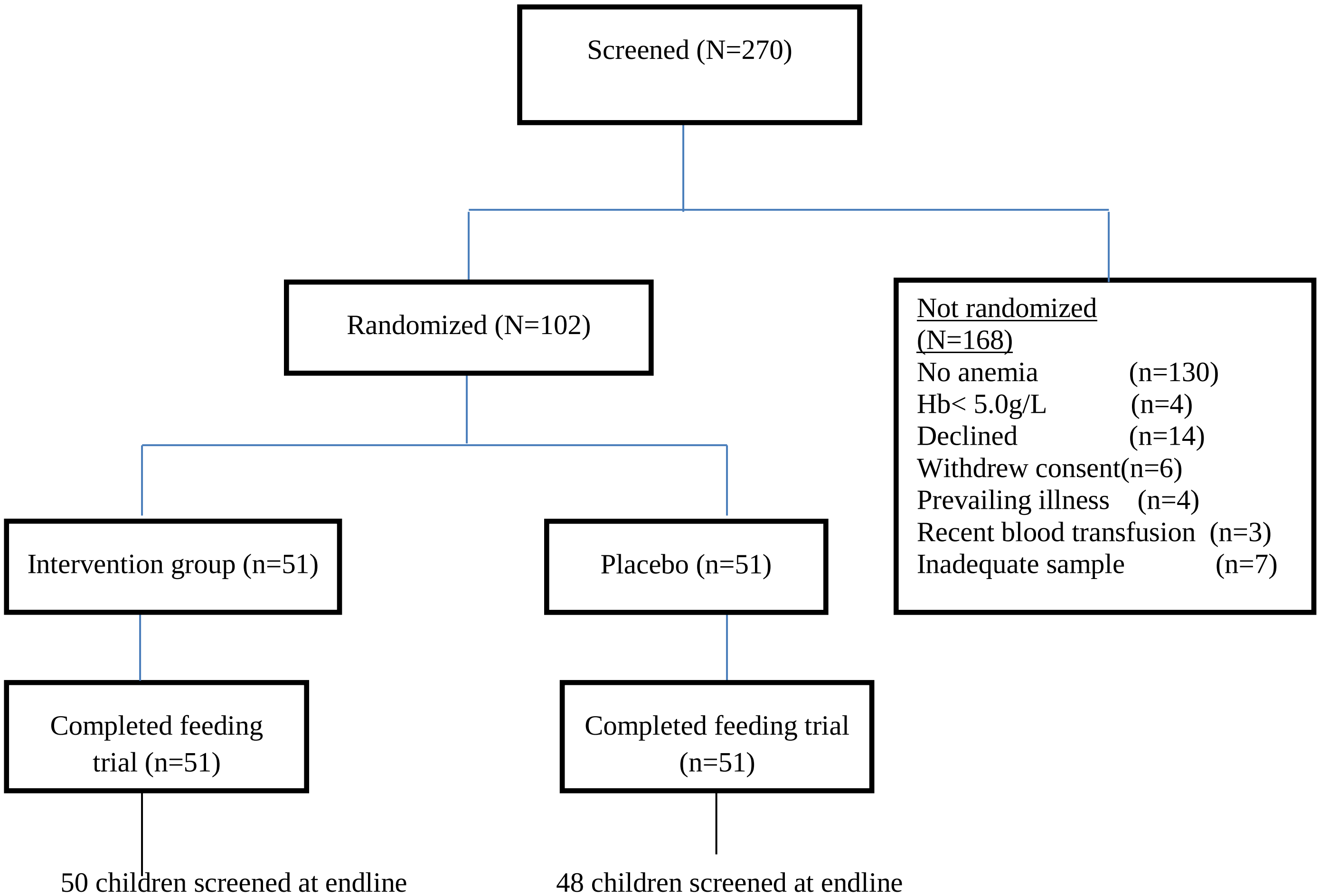

Figure 5.1: Sampling procedure for study groups

Data Collection

Household survey: A pre-tested questionnaire was used to collect background information of all the children. The main respondents were mothers or principal caretakers. The questionnaire was structured to collect data on socio-economic and demographic characteristics, morbidity status, dietary intake, nutrition status, food production, water and sanitation

Socio-demographic characteristics: Data was collected on household composition, size and ages and educational status of parents.

Morbidity status: A fourteen-day recall was used to assess child morbidity. Diseases assessed included cough, diarrhea, malaria, fever, pneumonia, skin diseases, worms and tiredness among others. Immunization was assessed based on child's clinic immunization card. During the survey all children who had not been dewormed for more than 3 months were dewormed with a dose of albendazole (GlaxoSmithkline, Nairobi, Kenya). Children aged 12 to 24 months received 200mg albendazole while children aged 25-60 months received 400mg albendazole.

Dietary intake: Food intake was assessed based on food frequency, 24-hour recall and breast-feeding and weaning questions.

The Food Frequency Questionnaire (FFQ): The FFQ consisted of a list of foods and an associated set of frequency-of-use response categories. The list of foods focused on iron rich foods as well as other foods that promote iron absorption and was sufficiently extensive to enable estimation of dietary diversity on the seven food groups for children as recommended by FAO. (Gibson, 2005, FAO, 2014).

24-hour dietary recall: This was conducted in a sub sample of 46 households to provide an estimate of habitual dietary intake of the children (Gibson and Ferguson, 2008). Mothers or primary caretakers of the children were the main respondents. During the 24 hour recall subjects were first asked to mention all the foods and beverages they had eaten during the preceding 24-hours (from the time they woke up on the previous day to the time they woke up again on the day of interview). Then they were asked to describe the foods and beverages consumed, including ingredients and preparation methods. The quantities of all foods, beverages and ingredients were estimated either in weight, household units (volume determined by water content), or in monetary value. The proportion of what was eaten by the child was determined based on the volume eaten and the total volume of the dish prepared. This proportion was used to calculate amount of ingredients consumed. For cooked dishes prepared outside the home, mainly food stalls, recipes and amounts of ingredients were obtained and amounts consumed determined. Conversion factors from household measures and monetary values to weight equivalent were determined (Van‘tRiet *et al*., 2002; Gewa *et al*., 2007).

Anthropometric Measurements

Weight, height and Mid Upper Arm Circumference (MUAC) measurements for all children were done. Data on the age of the child was also recorded. Weight was measured to the nearest 0.1 kg using a Salter spring scale (UNICEF) or a digital bathroom scale (Secam). Children were weighed in light clothing and without shoes. Height of children less than 2 years was measured to the nearest 0.1cm using recumbent length, while standing height was used for children above two years with a wooden measuring board as recommended by WHO, 1995 (Gibson, 2005). MUAC measurements were taken using a flexible non-stretch tape made of fiberglass, using standard procedures. All measurements were done twice and the mean value used in the analyses. Where the difference between the two measures was higher than 0.5 cm, a third measure was taken and the mean of the two closest values used in the analyses. Age of the children was estimated from their birth certificates, clinic cards or baptism cards which the parents or guardians produced. Weight and height measurements were converted to weight-for- age, weight-for-height and height-for-age indexes, expressed as *z*-scores by using

WHO cut-off points. Children were classified as stunted and underweight if their respective *z*-scores were < 2 SDs below the reference age and sex median.

Preparation of the Porridge Flour

Sorghum, millet and cassava were obtained from a local open air market in Kisumu (Kibuye market) and cleaned, milled, weighed and packaged in one kilogram portions, ready for mixing. Bovine blood was collected from Otonglo slaughter house in Kisumu and allowed to stand for 15 minutes, after which the serum was drained. The coagulated blood was cut into cubes and boiled while stirring continuously to break the clots. After cooking the blood was spread out in trays lined with aluminium foil and dried in a tent solar drier, until the blood completely dry before being milled. The flours were then mixed using ratios for sorghum: millet: cassava: bovine blood of 8:8:8:1. The aim was for each child to consume 1 cup daily, (approximately 300mls) of the cooked porridge. Each cup of the intervention porridge (enriched with bovine blood) was made from 25g of composite flour, while for the placebo (not enriched) group each cup had 25.5g of composite flour.

All porridge was cooked in a central place each morning. Each porridge type (bovine blood enriched or placebo) was cooked separately in a big saucepan. Using one and a half kilograms of the already mixed enriched porridge flour (enough for 60 cups of porridge) and a total of four and a half litres of water the porridge was mixed and cooked. Sugar was added to the porridge which was then allowed to boil briefly before removing from the fire. The same quantities and methods were used to prepare porridge using the un-enriched flour. hot porridge was then poured into ten litre jerry cans, which were labeled according to the colour code (pink for bovine blood enriched porridge and green for placebo) and feeding centre for ease of distribution.

Feeding Trials

On the first day of feeding each child was randomly allocated to one feeding group, either the intervention group (bovine blood) or the placebo group. Each group contained 51 children and children remained in the same group till the end of the intervention. Enriched porridge was served in blue plastic cups with handles, while the un-enriched porridge was served in similar green cups. Mothers or guardians came with the children and assisted the children to feed when necessary. The quantity consumed by each child was then recorded by the research assistants. After the first day children were directed to one of five allocated feeding centres. Each centre was less than five minutes' walk from the children's houses. Children were fed for a total of 30 days over a period of six weeks for five days each week, from Monday to Friday, during the morning hours from eight to ten o'clock. Consumption and attendance were monitored and recorded through daily records. Two public holidays fell within the feeding period and for these days mothers were given flour rations of the intervention porridges, which they were to use to prepare and feed the children at home. During endline biochemical assessment, two children from the intervention group and one from the placebo group did not present themselves for screening, although they had completed the feeding trials. At the end of the six weeks of feeding children were tested again for anaemia and iron status

Analytical Methods

Blood samples were taken from each child at the beginning and end of intervention and analyzed for haemoglobin (Hb), serum ferritin and soluble transferrin receptor as indicators of iron status. At baseline, blood samples were also analyzed for malaria parasites. Two medical laboratory technologists collected 5ml non-fasting blood samples via venipuncture using the vacutainer blood collection system. Each blood sample was divided and into two portions, one portion was stored in heparinized tubes to be analyzed later at the Lancet laboratories for serum ferritin and soluble transferrin receptor, while the other portion was immediately analyzed for haemoglobin content and malaria.

Malaria test: Children were tested for presence or absence of malaria soon after blood collection, using giemsa staining procedure. Preparation of working giemsa stain was done using giemsa buffer (40.0 mls), giemsa stain stock (1.0 mls) and 5% triton (2 drops). Using thick blood films, standard staining procedure was applied and the stained slides examined for presence or absence of malaria microscopically using Olympus microscope (CX21FSI-Japan). Children who tested positive for malaria were treated before inclusion in the study.

Determination of haemoglobin: A 5ml non-fasting blood sample from venepuncture was taken from each child by laboratory technicians using BD contact activated lancets (BD Microtainer). The blood sample was collected in BD pink microtainers with EDTA and flow tops connection, immediately stored at $4^{o}C$ and shielded from light. haemoglobin analyses was done using battery powered HemoCue machine (HemoCue HB 201: Sweden).

Serum ferritin and soluble transferrin receptor: The rest of the blood was stored in cool boxes and transported to Lancet Laboratories in Kisumu for analysis of Serum Ferritin (SF) and Soluble Transferrin Receptor (sTfR). Plasma concentrations of SF and sTfR were analysed using fully automated Cobas Integra analysers (Roche diagnostics, Mannheim, Germany) at the Lancet laboratories, Nairobi. Children who had Hb levels greater than 50 to < 110g/L, serum ferritin concentrations < 10µg/L and sTfR values > 8.5mg/L were described as having both anaemia and iron deficiency.

Data Analysis

Data coding, entry, and analysis was done using IBM SPSS Statistics package, version 22 (© Copyright IBM Corporation 1989, 2011) and Nutrisurvey software. Statistical tests included descriptive statistics and comparisons between key variables. Categorical data was reported as percentages and P values below 0.05 were considered statistically significant.

Nutritional anthropometric indices were determined using WHO child growth standards. Stunting, underweight and wasting were defined by Z scores below -2SD for Weight for Age, Height for Age and Weight for Height respectively. Middle upper arm circumference (MUAC) was determined using cut-off point of 12.5cm.

Data from food consumption was analyzed using nutrisurvey software (24 hour recall) while food frequency data categorized into food groups for young children as recommended by WFP, and analyzed according to intervention group and dietary diversity. The proportion of children consuming below the RDA for iron and other nutrients was also determined. Social demographic characteristics of the two groups and the percentages of subjects with iron below the cut off levels will be compared by chi-square test. The independent sample t-test (two tailed) will be used to compare the group means of iron status indicators. Changes in means of iron status indicators within groups will be assessed using paired sample t-test. Analysis of variance (ANOVA) will be used to compare means of iron status indices between the two groups. The covariance's will include demographic and socioeconomic factors such as age, religion and parents education level.

Ethical Considerations

The study was approved by the Ethics Committee of Kenyatta National Hospital and the University of Nairobi, Department of Food Science, Nutrition and Technology. Permission was also obtained from the Nyanza provincial administration and the Ministry of public health and Sanitation. Written informed consent was obtained from the caregiver of each child.

RESULTS

Baseline Screening

Out of 270 children screened for anaemia 53.6% (144.7) children were male and 46.4% (125.3) were female. Out of these 140 children were anaemic (Hb<11.0g/dl) giving an anaemia prevalence of 51.9%. The mean hemoglobin level was 9.176 (±1.6) mg/dl. At 95% confidence level, there was no significant difference in the mean hemoglobin levels for boys and girls ($p>0.05$). In terms of anaemia category, moderate anaemia was the highest among the children (53.9%), followed by mild anaemia (35%) and severe anaemia (10.8%). When children in the study, were categorized into those below and above three years of age there was no significant difference between age and anaemia ($P=0.132$) or sex of child and category of anaemia ($P=0.501$), thus the chances of being anaemic at 12 months or 60 months for all children are equal.

Socio-demographic and Socio-economic Characteristics

Characteristics of children were compared according to their feeding group to determine the extent to which the intervention or placebo groups were similar or different. The results indicate that in terms of socio- demographic and socio-economic characteristics children in the two feeding groups were not significantly different from each other (p> 0.005).The data is presented in Table 5.1.

Table 5.1: Household Socio-demographic and Socio-economic Characteristics of Children by Feeding Group

	Intervention group	Placebo group	Mean	P value
Household characteristics				
Household size	4.7(±2.05)	4.6(±1.77)	4.7	0.7
Male headed HH (%)	82.4	86.3	84.3	0.6
Age of HHH (yrs)	32.8(±10.5)	32.6(±8.4)	32.7(±9.4)	0.9
Age of caregiver (yrs)	27.0(±7.01)	27.0(±9.15)	27.04(±8.11)	0.9
Married HHH (%)	80.4	86.3	83.3	0.4
Education HHH(%)				
Primary	60	41.7	51	0.2
Secondary	38	46.1	44.3	
Tertiary	2	6.3	4.1	
Education caregivers(%)				
Primary	60.8	64.7	62.7	0.5
Secondary	39.2	33.3	36.3	
Tertiary	0.0	2.0	1.0	
Religion (%)				
Christian	84.3	85.3	85.3	0.52
Others	15.7	14.7	14.7	
Economic characteristics				
Source of income(%)				
Salary	9.8	9.8	9.8	1
Others	90.2	90.2	90.2	1
HH income(Ksh/m)	9000 (6000,12000)	7000(5000, 10000)	8000(5050,11625)	0.7
Food expenditure (sh/m)	5,515	6,110	5,600	0.3
Own farm (%)	29.4	28	28.7	0.9
Farm size(acres)	1.5	1.4	1.4	0.9
Own livestock (%)	37.3	25.5	31.4	0.2

HH-household. HHH: house-hold head. Ksh/m- Kenya shillings per month. Median:25thpercentile,75th percentile

Food Consumption

Food frequency: The food frequency data indicated that cereal based foods are the most commonly consumed foods by young children with most children taking cereal porridges at least two or more times every day. Consumption of animal source foods was reportedly low. Milk was the most commonly consumed animal source food by most children, but the frequencies were low and 12.7% of children indicated non-consumption. Fat consumption appears adequate as it is mainly used during preparation of foods. A high proportion of children (>70%) take black tea (tea with no milk) and this has important implications for iron nutriture. Consumption of fruits appears high as the season for mangoes was beginning at the time of the study while, consumption of Dark Green Leafy Vegetables (DGLV) was reportedly low. The results indicate no significant difference in consumption by food frequency in both the intervention groups ($p>0.05$). Table 5.2 analyses frequency of intake by the two intervention groups, based on seven food groups recommended for young children by FAO

Table 5.2: Food Frequency (%) Based on Weekly Consumption (N=102)

Food group		Intervention group	Placebo Group	Total	p value
Cereals		52.3	47.7	100	0.3
Legumes		68.6	72.5	70.6	0.7
DGLVs		19.6	23.5	21.6	0.6
Fruits		74.5	82.4	78.4	0.3
Milk		84.3	92.2	88.2	0.2
Meat		35.3	31.4	33.4	0.7
Fats		76.5	64.7	70.6	0.2
Beverages		64.7	52.9	58.8	0.2
Dietary diversity score	High (> 6)	25.5	33.3	63.7	0.7
	Moderate (4-6)	66.7	60.8	63.7	
	Low (< 4)	7.8	5.9	6.9	

Dietary Diversity: In terms of dietary diversity most children (63.7%) consumed four to six food groups weekly, which is rated as moderate. Only 6.9% of children in both intervention groups consumed less than four food groups on a weekly basis. With a mean diversity score of 5.7 (SD±1.46) for the active intervention group and 5.6(SD±1.46) for the placebo group, there was no significant difference in dietary diversity between the two groups (P=0.840). Results of dietary diversity are shown in Table 4

Dietary recall: Results of the 24-hour dietary recall, based on a subsample of 46 children, indicated inadequate nutrient intake in both feeding groups for practically all nutrients. Inadequate intakes varied from 38.8% inadequate intake of protein to 100% inadequate intake of calcium. Almost all children were also not able to meet their daily iron and folic acid requirements from the foods they consumed. However, there were no significant differences between feeding group and intake for all nutrients (p> 0.05). Results of the 24-hour dietary recall are presented in Table 5.3 according to age category (1-3 years and 4-5 years) and feeding group.

Table 5.3: Nutrient Intake of Children (n=46)

Nutrient RDA	Mean intake	Median (25th & 75th percentile	Inadequate intake (%)
Protein	25.1(±13.7)[1]	22.9[1] (15.0, 32.4)	20.0[1]
13g*/19**g	23.6(±18.8)[2]	19.7[2] (13.3, 24.1)	18.8[2]
CHO	144.7(±84.4)[1]	132.2[1] (76.1, 197.4)	56.7[1]
130g (1-5 years)	133.3(±77.6)[2]	130.3[2] (78.3, 156.3)	37.5[2]
Fat	17.2(±7.6)[1]	16.9[1] (11.2, 22.9)	-
ND	17.7(±11.5)[2]	14.7[2] (11.5, 19.4)	-
Vitamin C	24.2(±20.6)[1]	19.1[1] (7.1, 41.7)	53.3[1]
15*/25**mg	30.2(±35.6)[2]	35.6[2] (7.4, 46.2)	31.3[2]
Vitamin A	2074.0 (±7892.7)[1]	380.6 (188.2, 1050.7)	58.3[1]
300*/400**µg	1015.8 (±2410.2)[2]	362.2[2] (163.2, 950.0)	53.8[2]
Iron	4.4(±2.5)[1]	4.4[1] (2.9, 5.5)	83.3[1]
7*/10**mg	4.1(±2.7)[2]	3.3[2] (2.2, 4.9)	100[2]
Folic acid	70.3(±36.6)[1]	70.6[1] (42.9, 92.9)	96.7[1]
150*/200**µg	68.2(±41.2)[2]	57.3[2] (36.5, 89.5)	100[2]
Calcium	163.4(±92.8)[1]	154.3[1] (74.5, 240.1)	100[1]
700*/1000**mg	176.6(±116.4)[2]	139.4[2] (107.8, 209.4)	100[2]
Zinc	4.89(±3.0)[1]	4.8[1] (2.2, 6.6)	36.7[1]
3*/5**mg	4.44(±3.3)[2]	3.6[2] (2.3, 5.6)	56.3[2]

RDA Source: FNB 2001: *1-3 years/ **4-5years; ND: Not Determined. Intervention group[1]Placebo group[2]

Morbidity Among Children

Results of the 14 day morbidity recall indicated that 76% of the children had been sick in the previous two week period before the survey, with most of them having two or more illnesses or symptoms of illness. Baseline screening for malaria (N=270) indicated that 10.7% of the children had malaria. However, malaria was reported (n=102) as the most common illness experienced by 80% of the children.

Other conditions reported as common were acute respiratory infections (ARI) such as coughs, gastro-intestinal infections such as diarrhea and worms and various skin problems. However there were no significant differences in morbidity status among children on the two intervention groups ($p>0.05$) as indicated in Table 5.4.

Haemoglobin, Iron and Nutrition Status of Children.

The haemoglobin and serum ferritin concentrations, though not significant, were slightly lower in the placebo group (9.04g/dl and 58.71 µg/L respectively) compared to the intervention group (9.34g/dl and 36.69 µg/L respectively). The soluble transferrin concentration in the placebo group was 11.26 mg/L compared to 9.43 mg/L in the intervention group. The difference was not significant ($p=0.06$) though almost significant. The proportion of children with iron deficiency anaemia was higher in the placebo group (43.1%) compared to the intervention group (27.5%).

The results on nutrition status indicate that the height for age, weight for age and weight for height z scores are all within the normal range for children in both feeding groups. The data, especially height for age, was slightly skewed towards the negative. However there were no significant differences in nutrition indicators for the two groups ($p>0.05$). The proportion of children who are stunted at 27.5% was higher than in the intervention group compared to the placebo, while the proportion of those who were wasted and underweight was higher in the placebo group. These differences were, however, not significant ($p>0.05$) and the results indicate that the randomization process was successful as both the intervention and placebo arms were comparable for the variables that were measured including haemoglobin, iron and nutrition status. Table 5.4 presents the baseline characteristics of the children.

Table 5.4: Baseline Characteristics of Children by Feeding Group

	Intervention group n=51	Control group n=51	Mean N=102	p value
Sex ratio(M/F) %	47.1/52.9	37.3/62.7	42.2/57.8	0.3
Age of children (m)	29.3(±12.5)	33.0(±13.4)	31.2±13.0	0.2
Nutritional status				
Height-for-age	-1.3±2.4	-1.05±1.8	1.17±2.1	0.5
Weight-for-age	-0.3±1.7	-0.7±1.6	-0.6±1.6	0.3
Weight-for-height	0.46±2.1	-0.24±2.1	0.12±2.1	0.1
Malnutrition				
Stunting (%)	27.5	21.6	24.5	0.5
Wasting (%)	9.8	11.8	10.8	0.8
Underweight (%)	13.7	19.6	16.7	0.4
Hb/Iron status				
Haemoglobin (mg/dl)	9.34±1.6	9.04±1.6	9.19±1.6	0.3
Serum Ferritin(μg/L)	58.71±50.2	36.69±55.8	47.70±52.9	0.1
sTfR (mg/L)	9.43±4.3	11.26±5.3	10.3±4.9	0.058
ID (%)	26.5 (14.2,38.9)	34 (20.5,47.6)	30.2 (21.0,39.4)	0.4
IDA (%)	27.5	43.1	35.3	0.1
Morbidity, water, sanitation				
Malaria (%)	76.5	84.3	80	0.3
ARI (%)	47.1	62.7	54.9	0.1
GIT infections(%)	51	45.1	48	0.6
Skin disease(%)	37.3	41.2	39.2	0.7
HH water/day (L)	86.4	78.1	82.2	0.3
Treat drinking water %	62.7	47.1	54.7	0.1
Use pit latrine%	96.1	91.8	94	0.4

sTfR-soluble transferring receptor   ARI-acute respiratory infections L-Litres
GIT- gastrointestinal tract

Randomized Controlled Trial

Ascertainment of outcome: The intervention and control group consumed similar volumes of the study porridge, 6,565 mls and 7,119 mls respectively (p=0.1). At the end of the study the mean Hb in the intervention group was 10.5mg/dl while that in the control group was 9.15mg/dl. This increase in the intervention group was significant (p=0.000). The mean change (Δ) in Hb concentration in the intervention arm was 1.2mg/dl compared to 0.2mg/dlin the control arm (p=0.008). 38% of the children from the intervention group had normal Hb levels compared to 15% in the placebo group.

At the end of the study there was a non-significant difference in the mean SF concentration in the intervention group, compared to the control arm, of 40.79 and 35.45µg/L respectively (p=0.578). The mean change in SF was -17.9 and -3.32µg/Lin the intervention and control group respectively, however this change was not significant (p=0.252). Although there was no improvement in iron stores after intervention, when all factors are considered, the higher values at baseline may have been due to mild infection or inflammation. The SF values obtained indicate that the iron stores are above the prescribed cut offs for deficiency of 10-30 µg/L (Biesalski and Erhardt, 2007).

At the end of the study there was a significant difference in the mean Soluble Transferrin Receptor (sTfR) concentration in the intervention group compared to the control group of 9.2 and 11.6mg/L respectively (p=0.021). The mean change within the group was -2.0 and 0.6mg/L for the intervention and control arms which was not significant (p=0.469).

At the start of the intervention all the children (100%) were anaemic. After intervention the prevalence of anaemia reduced to 62% and 85.4% in the intervention and control groups respectively and this reduction was significant (p=0.009).

At the end of the study the prevalence of Iron Deficiency (ID) was significantly lower (p=0.013) in the intervention group (8%) compared to the placebo group (27.1%). There was also a reduction in the prevalence iron deficiency anaemia from 27.5% to 12.9% in the intervention group and from 43.1 to 29.3%in the placebo group. This reduction was significant within each group when compared before and after intervention(p=0.02), but insignificant when comparing the intervention and placebo groups. The overall prevalence of iron deficiency anaemia reduced from 35.3% at the baseline to 22.2% after intervention. The results are presented in Table 5.5

Table 5.5: Effect of Bovine Blood Enriched Porridge on Iron Status of Children (N=98)

	Intervention group (n=50)	**Control group (n=48)**	**P value**
Total mean consumption porridge (mls)	6565 (±2012.1)	7119 (±1493.7)	0.13
Hb concentration (mg/dl)	10.5 (±1.05)	9.15 (±1.4)	0.00
Serum ferritin concentration (µg/L)	40.79 (±37.53)	33.45 (±50.23)	0.58
sTfR concentration (mg/L)	9.2 (±3.8)	11.6 (±6.4)	0.02
Mean (Δ) in Hb conc.	1.2 (±1.7)	0.2 (±2.0)	0.01
Mean (Δ) in SF conc.	-17.9 (59.7)	-3.23 (±64.3)	0.25
Mean (Δ) in sTfR conc.	-0.2 (±5.0)	0.6 (±6.0)	0.47
Anaemia (%)	62.0	85.4	0.01
Iron deficiency (%)	8.2 (0.5,15.5)	27.3 (14.5,39.7)	0.01
Iron Deficiency Anaemia (%)	12.9(3.0,21.0)	29.3(16.3,42.0)	0.10

The risk of anaemia, iron deficiency and iron deficiency anaemia attributable to the exposure to the adverse situation of being in the control group and therefore not benefiting from the supplementation was 23.4%, 19.1% and 16.4% respectively. At the end of the feeding period, compared to the control group, children in the intervention group had a 127%, 239% and 37% reduction in the prevalence of iron deficiency anaemia, iron deficiency and anaemia respectively.

DISCUSSION

In this study it was important that the feeding groups should not be significantly different, so as to control for possible confounding factors, which is important in areas of high transmission of plasmodium falciparum malaria, and other morbidities common in the study area

(Brittenham, 2007). Background characteristics of children in both feeding groups were comparable at baseline, as there were no significant differences for age, sex, nutritional status, haemoglobin concentration and serum ferritin concentration ($P>0.05$). The sTfR results at baseline indicated a slight significance between the groups ($p=0.058$), which could be attributed to asymptomatic inflammation which might have been higher in this group, but was not physically evident at baseline.

Malaria, an illness positively associated with anaemia was the most commonly reported illness in the two week morbidity assessment. In this study, malaria prevalence was 10.7%, which is comparable to the estimated prevalence rate of malaria in the region of 12% (Kenya Malaria Indicator Survey, 2010). Children with malaria were not excluded from the study based on malaria status. They were included if they met the other criteria, after being treated for malaria. The other common illnesses children experienced two weeks before the survey were acute respiratory infections and gastrointestinal tract related symptoms such as vomiting and diarrhea. These findings are similar to results of the recent Kenya Demographic and Health Survey (KDHS, 2015) which found these conditions to be among the most common in Kenyan children. All these conditions have important implications as they compromise both nutrition and iron status.

Findings from several studies indicate that the diets of young children's in many countries, including Kenya, fails to provide adequate iron and that starchy cereals are the main foods consumed (Mwaniki *et al*, 2001; Macharia-Mutie, 2012; Dewey,2007). These findings are similar to the results of this study which also found cereal based foods, and especially cereal porridges to be commonly consumed by young children. The results of the food frequency and 24-hour dietary recall both indicate that consumption of animal source foods such as meat and liver, known to be rich sources of iron, was low. This can be largely attributed to the prohibitive cost of these foods, hence households of vulnerable children are limited in accessing them (IFPRI, 2001). Milk intake is important for all young children and provides critical nutrients for child growth and development, especially calcium. However in this study, none of the children met their daily requirement for calcium. This is of particular concern for mothers of young children as calcium is critical in the development of strong bones and teeth. Thus in addition to inadequate iron and folic acid intake the results indicate widespread nutrient deficiencies for children which require attention.

Out of the three nutritional indicators assessed, all the children were found to be within the normal distribution. However it was noted that the distribution was skewed towards the negative especially for stunting and wasting, with height for age having a mean of -1.2 z-score for all children. The prevalence of stunting in the study children of 24.5%, though lower than the Kenya national stunting rate of 35% (KDHS, 2015) falls within the WHO medium category prevalence rate (20-29%) while wasting prevalence in the children of 10.8% falls within the WHO category of high prevalence (10-14.9%). This information is important and shows that the children require targeting for nutrition intervention (WHO, 1995).

After six weeks of consuming porridge there were haemoglobin changes in both groups, with the improvement in the intervention group being highly significant (p=0.00) compared to the placebo group. This finding supports other findings which have likewise found both the iron content and bioavailability of bovine blood high enough to improve the iron quality of diets (Allen, 2003; Mosnews, 2007; Walter *et al*, 2001; Ofori and Hsieh, 2011;Kikafunda and Sserumaga, 2007). The status of 52% of the children changed from anaemic to non anaemic category in six weeks. The majority were from the intervention group which realized a mean Hb improvement of 1.161g/dL compared to the placebo group's improvement of 0.111g/dL.

Conclusion

The results indicate that bovine blood enriched porridge can be used to improve iron status of anaemic children in malaria endemic zones. With proper morbidity control, iron status can improve in a relatively short period of time. It must also be noted that even just giving porridge prepared from the local cereals and cassava led to a slight improvement iron status.

Recommendations

Nutrition interventions using bovine blood should be developed and implemented for young children living in malaria endemic regions of Kenya, to reduce the prevalence of iron deficiency and anaemia.

CHAPTER 6

GENERAL DISCUSSION AND RECOMMENDATIONS

Background

This study aimed at improving the iron status of anaemic children by finding a way to utilize bovine blood in a randomized trial. The study results are intended to reduce the prevalence of anaemia and iron deficiency, especially among vulnerable population groups in malaria endemic regions who do not have alternative sustainable sources of intervention. The ability to generalize the findings depends on understanding existing and traditional consumption patterns so as to enable effective incorporation of bovine blood in the most acceptable way. Hence the preparatory stages of the study focused on finding out if bovine blood is acceptable and preparation of the porridge product before conducting the feeding trials.

Methodological Considerations

The use of bovine blood as food is practiced in several communities in Kenya. The prevalence of anaemia among young children in Kenya is highest in the Lake Victoria basin and coastal region of Kenya. The Lake Victoria basin region was selected as it represents a region where bovine blood is accepted as food (Mwaniki *et al*, 2001; Andago *et al*, 2015), unlike the Coastal region where a higher proportion of the population are Muslims, and are thus prohibited from consuming bovine blood.

Information on the use of bovine blood as a food or to reduce anaemia in Kenya is limited and mostly based on the culture of the Maasai people who usually consume fresh bovine blood.

Since not much has been recorded from other regions, a rapid assessment was done at the onset of the study, using key informants and focus group discussions to establish existing practices, acceptability and consumption. The study also gave information on foods consumed by young children, which could be selected for enrichment to improve iron status. Most of the information given in the FGD's on anaemia and food consumption was confirmed by the KII's and was considered reliable enough to be used in development of the porridge product, since it was important that the product be based on existing practices to make it acceptable to the targeted community.

The dark colour of bovine blood is often considered a deterrent to its use in foods, as well as safety concerns, since blood is known to be a rich media for bacterial growth (Walter et al,2001). Hence it was important to ensure acceptable parameters are obtained to meet standards, through rigorous laboratory procedures. These objectives were achieved by conducting the necessary analyses and evaluations using appropriate procedures, ensuring that acceptable standards of hygiene and sensory acceptability were met. Methods for porridge flour preparation in the field, for the feeding intervention, used the same methods applied during product development for sorghum, millet and cassava. For bovine blood all the methods were the same except for drying of cooked blood which was done using a tent solar dryer in the field as opposed to oven drying in the laboratory. The blood powder from the field was analyzed for iron and nutrient content to ensure similarity with laboratory quantities.

Baseline screening aimed at identifying anaemic children aged 12-60 months with no underlying health conditions, as the intervention aimed at assessing the effect of consumption of bovine blood enriched porridge on iron status. To minimize the chances of children with underlying health conditions being included, a pediatrician from Kisumu East District hospital, clinically assessed all children after screening. Children with malaria or other conditions were referred to the hospital for treatment. The study criteria of enrolling children aged 12 to 60 months were suitable as all the children were already familiar with drinking porridge as it is a common weaning food. This could be part of the reason why most children in the intervention consumed their daily porridge rations without any problem. Anaemic children were identified using haemoglobin cut-off points (Hb< 11.0mg/dl), which is widely accepted as a common, inexpensive and quick method in such studies. However Hb is not considered sufficiently sensitive or specific for iron deficiency hence blood samples were further tested for Serum Ferritin (SF) and Soluble Transferrin Receptor (sTfR). Ferritin was used to determine iron stores (<10.0µ/L iron deficiency) and sTfR was used to determine if iron deficiency (> 8.5mg/L) occurred alone or due to anaemia of chronic disease (Biesalski and Erhardt, 2007; WHO, 2011).

The household survey aimed at establishing background characteristics of the children by collecting data on demographic and socio-economic dimensions of households. The study was done in the peri urban area of Kisumu and it was important to establish that the background characteristics of children in the study were not different. In addition, many studies have indicated a relationship between socio-economic factors and anaemia (UNICEF/UNU/WHO; 2001), these factors once determined, often offer important considerations for designing effective anaemia interventions.

Low dietary intake of iron is often indicated as the main cause of iron deficiency and anaemia (Underwood, 2001; Gibson R. 2005; Mannar, 2007), hence it was important to establish the extent to which iron requirements are met by the diet. The food frequency questionnaire, which is based on recall, gave a general picture while the 24 hour dietary recall, though based on a representative sample, gave a much clearer picture, clearly bringing out the extent of inadequate intakes of iron rich foods.

During the feeding intervention children were randomly allocated to groups, by being given a pink or a green card alternately, as they walked in on the first day of feeding, the child's number and feeding group was then recorded and they stayed in these groups till the end of the intervention. Data at baseline also indicated no significant difference between the two feeding groups in all aspects, that is age, sex, nutritional status and iron and anaemia status, except for soluble transferrin receptor where there was some significant difference, though the association is not strong (p= 0.058).

Main Findings

Results of the study on knowledge and practices related to iron deficiency and anaemia are presented in chapter two. The main finding is that there is knowledge on iron deficiency and anaemia in the community, the knowledge is largely based on symptoms and causes. Anaemia is also mainly identified through symptoms such as pallor and weakness. These findings are similar to findings of studies done in other regions which have likewise observed that the condition of anaemia described as *'blood is over'* or *'blood is not enough'* and other similar terms is quite common, and that these terms are largely semantic constructions associated with the ill health of anaemia (Utzinger J. *et al*, 2013; Galloway *et al*, 2002; Kouadio *et al*, 2013). Studies have also found that anaemia is mainly identified through physical symptoms, such as pallor and weakness. This implies that by the time symptoms are observed for possible action, it is most likely at the moderate to severe stage, sometimes even requiring blood transfusion. Identifying anaemia at the earlier stages of mild and moderate could result in more timely and less intrusive interventions. Biochemical analyses are highly recommended to determine subclinical anaemia, though they are often prohibitive due to cost and required technical expertise.

Responses from the community indicate that they have knowledge on the major causes of anaemia which were based on insufficient dietary intake, in both quantity and quality, especially of animal source foods; illnesses leading to blood loss, either directly or indirectly and certain environmental conditions. These findings are not different from findings of studies in other countries, which have likewise indicated similar knowledge (Kouadio *et al*, 2013; Galloway *et al*, 2002). However, knowledge at the community level has not translated to reduced anaemia prevalence, mainly due to the multifactorial nature of the causes of anaemia, especially the low socio economic status of vulnerable populations. There were however a few misconceptions related to causes and iron rich foods. For example heat from the hot sun or fire is believed to cause anaemia by ‘sapping’ one’s energy or blood; and certain foods like soda’s (coke and fanta) and beetroot are seen as iron rich. Even lemons in this study were seen as drying up blood, yet lemons can actually improve iron absorption being citric in nature. These misconceptions which have also been documented in other studies, shows that there is a need to improve the community understanding of anaemia causes and iron rich foods. knowledge of antinutritional factors was poor, accompanied by practices like consumption of black tea. These practices can be addressed to improve iron status.

In chapter two and three, animal blood is clearly indicated as an iron rich food as well as a known traditional remedy for anaemia. Therefore the study established that bovine blood is a cherished and well accepted food, with existing traditional methods of preparation and preservation. It was however noted that senior citizens were more knowledgeable about bovine blood compared to young mothers or even health workers. This is consistent with the erosion or loss of traditional knowledge and practices related to food consumption, over time which has been noted in many developing countries often leading to negative nutritional consequences, among vulnerable populations. This was also noted by Oruka who commented in his publication (Oruka, 1997)that:

"Indigenous Africa has a cluster of serious and simple techniques for creating and preserving foods, and most of these are becomming completely unknown to to the so called modern African..."

Available literature indicates that bovine blood use in Kenya is most common among the Maasai people. The implications of this study are that bovine blood maybe more widely accepted than is actually documented in many communities in Kenya, this information together with the fact that most blood goes to waste during slaughter, presents an opportunity for the development of suitable food products to alleviate anaemia.

There is also a growing interest in the use of bovine blood, one of the major waste products from the meat industry, to improve on quality of food products. Scientists from the Voronezh

Technological Academy in Russia, in a breakthrough innovation, came up with methods of manufacturing yoghurt and cream among other products for human consumption using bovine blood. (Mosnews, 2004). In Chile bovine blood has been used in the national school breakfast programme to reduce anaemia among young children. However there is so far no literature indicating bovine blood use in food production or anaemia alleviation in Africa, despite the potential. The publication by Kikafunda and Sserumaga, (2005), investigated the possibility of developing a bovine blood powder for anaemia.

The findings of this study show, as no other study in Kenya has shown, that bovine blood can be successfully used to reduce anaemia and iron deficiency in malaria endemic regions since children in the intervention group had a 127%, 239% and 37% reduction in the prevalence of iron deficiency anaemia, iron deficiency and anaemia respectively, compared to the placebo group.

Results of this study also confirm the results of other studies, on the inadequate dietary intake of iron and other nutrients by young children at this important stage of growth and development. This might be the reason why there was an improvement in the placebo group, with the one cup of porridge per day, implying that even a simple nutrition intervention can lead to improved iron status.

Strengths and Implications of the Study

The study had several strengths the main one being that randomisation was successful as the children in both groups were comparable, hence there were no significant difference in the baseline characteristics of children in both feeding groups. This made it possible to truly assess the effect of the enriched porridge on iron status. All the products used to make the porridge were based on local foods, hence the porridge can easily be prepared even at household level, using appropriate technologies. Where bovine blood is an accepted food, using it in porridge would help to build on existing traditional knowledge, which is otherwise likely to be forgotten over time. The fact that bovine blood is so widely available, and largely goes to waste has implications for value addition through industrial development and local income generation possibilities. The results of this study are thus a strong basis fo further research and innovation andpolicy guidelines, leading to significant contribution in the management of the iron deficiency and anaemia in Kenya.

Conclusion

The main objective of the study to improve iron status among anaemic children in a malaria endemic region was achieved as 52.6 % had improved status after six weeks.

Recommendation

The study findings provide important information for addressing the serious problem of anaemia and iron deficiency among young children in malaria endemic regions in Kenya. The findings should be considered for future research and interventions aiming to reduce the high prevalence rates of iron deficiency.

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Appendix 1: Key Informant Interview Question Guide.

Name of Key in formant____
Designation ____
Interviewer's name____
Date of Interview ____
Time ____________
Place ______________________________

General introduction of researcher/recorder. Introduce topic. Inform of intention to record.

Anaemia

1. What are the main nutritional deficiencies of young children in the area?
2. What are the common childhood diseases in the area?
3. What is the anaemia situation in the area? (mild,moderate. Severe)
4. Is there a local name for anaemia?
5. What are the main causes of anaemia in the region?
6. Who is most affected?
7. What is being done about anaemia in the area?
8. Have these contributed to reducing anaemia in the area?
9. Are parents of young children in the area aware about the anaemia problem?
10. Do parents of young children know about the causes of anaemia?
11. Are mothers aware about the impact of antinutritional factors in food (iron binders)? (milk, black tea)
12. What do parents of young children do to prevent/control anaemia?
13. Are there any specific interventions? (nutrition education? Supplementary food programmes)

Food consumption habits

1. What foods are commonly consumed by young children in the region?
2. Are there any foods which are culturally encouraged or prohibited for children?
3. Which porridge ingredients are commonly used for young children?
4. Where do mothers obtain information on porridge flour for children?
5. What is the trend with the consumption of iron rich foods? (meat, chicken, liver etc)
6. What are the trends in consumption of fruits? (oranges, lemons---)
7. Are vegetables commonly consumed by children? Which ones?
8. Do mothers commonly add foods to enrich porrige (e.g. lemon, sugar, milk)?
9. How is the porridge prepared ?
10. Are there any special foods used by those who are anaemic? Which foods?
11. Are there any foods children are not allowed to eat and why

Bovine blood consumption

1. What is the communities' attitude towards use of bovine blood in food?
2. Is bovine blood consumed by children?
3. How is it consumed (fresh/cooked/methods)?
4. What challenges might be experienced with the bovine blood study?

Appendix 2: Focus Group Discussion Guide

Date of discussion	Marital status and/or age
District	Level of education
Location	Occupation
Sub-location	Number of children
Village	Number of children under 5

Moderator's name ______________________ Recorder's Name ________________________

General introduction of researcher/team member (s)

Topic	Discussion
Introduction	Facilitator, recorder's names.
Topic of interview	I am a student from the UoN-I will be conducting research in your community, and would like to discuss with you about the knowledge, attitudes and practices related to IDA and foods consumed.
No right or wrong answers- your opinions	This is not a test and there are no right or wrong answers to any of the questions. I just want to find about iron deficiency, foods consumed in the community and your opinions on this.
Length of time of discussion	The discussion will take about one hour
Talking to one another	As we are discussing many things about ourselves, it is important we do not all talk at once, so that we can hear one another's point of view.
Explain note taking and tape-recording. Confidentiality.	--(Name of note taker) will write down some things that we talk about, so that we can remember them later. We would also like to use a tape recorder if that is ok with you. We are the only ones who will know your names.
Check understanding. Clarification if needed	Let us check, if we have all understood what we are going to do. Can anyone repeat for us? If anyone needs clarification, you may ask questions now before we continue.

Topic	Discussion	Probes
Introduction	Please introduce yourself and as you do, tell us something about yourself your family etc.	
Knowledge	- What is the role of iron in the body? - What are the common childhood illnesses in the community? -What do you understand by the term anaemia? -Which foods do you know which contribute to building/improving blood? -What is the cost of these foods? -Are there other causes of this problem (anaemia)? -Which groups in the community are most affected by anaemia? Why? -How is this condition addressed in the community? - Source of knowledge for iron, anaemia, child feeding?	- Plant/animal sources - Local name for anaemia? - Symptoms of anaemia - Short/long term impact on child - Diseases/conditions affecting iron status - Vulnerable groups - Supplementation/fortification
Practices	-list all foods consumed bychildren aged 1-5 in this community? - which is the most desired complementary food(s)? - Method of preparation of preferred food -Are there special foods for anaemic children? -Where do you obtain these foods ? -Are there some households not able to obtain these foods? Why? -At what age do children start taking porridge? -Which flour(s) are used to prepare porridge? - What influences choice of porridge flour? - how much porridge is consumed by children aged 1-5 years? -Are there foods children are not allowed to eat in the community? Why? - -Where is bovine blood for consumption obtained in the community?	- Self production/purchase - Preparation method when resources adequate/limited. - Culture limiting consumption of iron rich foods/bovine blood. - Factors affecting absorption and nutrients of iron and nutritional quality e.g. tea, citrus fruits, DGLV's, sugar, milk. - Preparation of bovine blood dishes. - Porridge quantity consumed at ages 1,2,3,4 and 5 years.

	-Who prepares the food? -How are these foods prepared/consumed	
Attitudes	- In your opinion which porridge flour do children like most? - Which porridge flour(s) would mothers use if they had a choice? -Is bovine blood an acceptable food for adults/children in the community? -do you think it can contribute to reducing anaemia in children.	

Appendix 3: Sensory Evaluation Form

Form number_ ______________________

SENSORY ANALYSIS (AT THE COLLEGE OF AGRICULTURE AND VETERINARY SCIENCES)

Study ID: _________________________ Date:_________________________

Instructions

- Read the instructions before the exercise
- Grade the products according to the codes given for colour, taste, mouth feel and general acceptability attributes
- Rinse your mouth after examining each product

Sample	**Colour**	**Taste**	**Mouth Feel**	**General Acceptability**	**Comments**
201					
303					
304					
204					
205					
206					

1 = dislike very much
2 = dislike moderately
3 = dislike slightly
4 = neither like or dislike
5 = like slightly
6 = like moderately
7 = like very much

Appendix 4: Household Questionnaire.

Improving the iron status of children in the Lake Victoria Basin region, using porridge flour enriched with bovine blood.

Please fill in the spaces with the requested information or tick or circle where applicable

Date of interview____________ Questionnaire No._______________ HH number_________

Name of interviewer ____

Name of respondant ____

Location __________ Sub-location ________________ Village__________________

Section A: DEMOGRAPHY

Q1. List all HH members in the table below and information corresponding to each member by using the appropriate code

Serial number	Name	Sex	Age	Relationship to HHH	Marital status	Religion	Level of education	Occupation
1								
2								
3								

Sex: 1. Male 2. Female 3. Other

Relation to HHH	Marital status	Religion	Education	Occupation
1.HHH 2. Spouse 3. Son 4. Daughter 5. Brother 6. Sister 7. Grandchild 8.Parent of HHH 9. Niece/nephew 9.Other (specify)	1. Married 2. Single 3. Divorced 4. Widowed 5.Other (specify)	1. Catholic 2. Protestant 3. SDA 3. Muslim 4. ATR 5. other (specify)	1.None 2. underage 3.Pre-school 4.Primary incomplete 5.Primary complete. 6.Secondary incomplete 7.Secondary complete. 8.College 9.University 10.Other ______	1.Salaried employee 2. Housewife 3.Farmer 4.Self employed 5.Casual labourer 6. Student 7. Unemployed 8.Retired(on pension) 9. Other (specify)

Section B: SOCIO-ECONOMIC STATUS

1. Is the house you live in?
   1. Your own 2. Rented 3. Other ___________________________
2. **(If rented)** How much do you pay per month? Kshs ____________________

3**.** What is the HH's main source of income?
   1. Salaried/wage employment
   2. Trade
   3. Crop sales
   4. Animal & animal product sales
   5. Casual labour
   6. Food aid/gifts
   7. Other ____

4. What is your total household income per month?
   1. Less than 2,000

2. 2. 2000-5000
3. 3. 5000-10,000
4. 10,000-20,000
5. 20,000 and above

5. Approximately how much money do you spend on HH food per month? __________

6. **Observe**: Materials used to construct main house.

a) Roof : 1. Grass thatch 2. Iron sheets 3. Tiles 4. Other __________________

b) Wall: 1.Plaster 2. Brick/block/stone 3. Wood 4. Iron sheets 5. Mud
6. Other______________________________

c) Floor: 1. Cement 2. Brick/tiles/stone3. Mud 4. Wood
5. Other_______________________________________

7. What is the main source of energy for lighting?
1. Tin lamp (nyangile) 2. Hurricane lamp2. Pressure lamp 3. Gas 4. Electricity
5. Solar 6. Other_________________________________

8. What is the main source of energy for cooking?
1.Wood 2. Charcoal3. Paraffin 4. Gas 5. Electricity 6. Other_________________

Section C: Food production and utilization

9. Do you have your own farm? (1. Yes 2.No)
10. **(If yes)** What is the size in acres_____________________
11. How much land do you use for food production?____________________
12. Do you rent land? (1. Yes 2.No)
13. How much do you pay per acre of land? ___________________________
14. What crops do you grow on your farm?

	Cereals	Fruits	Vegetables	Others
1.				
2.				
3.				
4.				

15. What do you do with your farm produce?
1. Home consumption 2. Sale3. Home consumption and sale 4.Other__________________

16. Do you own any livestock? (1. Yes 2.No) **If yes** indicate which ones below.

Animal	Number	Use 1=HC 2=HC &S 3=O
1=cows		
2=goats		

3=sheep		
4=pigs		
5=rabbits		
6=chicken,ducks,turkey		
7= other		

HC- Home consumption S-Sale O- Other

Section D: Water and sanitation

17. What is your main source of drinking water?

1.Tap 2. River 3. Borehole 4. Wells 5. Rain 6. Pond

7. Other___________________________

18. How much water do you use per day? (in litres)_____________________________

19. Do you treat your drinking water? (1.Yes 2.No) **If Yes** how?

____

20. Distance to water source. ____________ Km ______________minutes.

21.Do you have a toilet? (1. Yes 2. No)

22. **If Yes,** What type of toilet? 1. Pit 2. Water closet (WC) 3. Other

____

23. **If No**. What happens? ____

24. How do you dispose of refuse from your compound?

1. Compost pit 2. Burn 3. Bury 4.Public/private disposal

5. Other _______________________________

25. Observe the following during interview and fill appropriately.

Observation	**Yes=1 No=2**
Compound clean	
Food covered	
Utensils clean	
Utensils covered	
Presence of human waste in compound	
Presence of toilet in compound	

Section E: Morbidity and Immunization.

26. What are the most common illnesses among children in the community?

1. Anaemia (Tiredness)
2. Coughing
3. Diarrhoea
4. Fever

5. Malaria
6.Malnutrition
7. Measles
8. Pneumonia
9. Scabies
10. Worms
11. Any Other________________________________

(*Please circle the appropriate number)

27. Has your child suffered from any of the above illnesses in the past two weeks? 1. Yes 2. No

If yes which one(s)____

28. Does your child have a clinic card? 1. Yes 2. No
29. Verify with clinic card if child fully immunized.
1. Fully immunized for age 2. Not fully immunized for age 3. Not immunized

30. Has your child ever received iron supplements or had blood transfusion? (1. Yes 2. No)

31. Are there any foods you withhold when your child is sick? (1. Yes 2. No)

32. If Yes, indicate the following.

Sickness	Food with-held	Reason

Section F: Food consumption

32. Is your child still breastfeeding? **1. Yes 2. No**

33. **If yes**. how often in one day? 1, 2, 3, 4, 5, 6= more than 5 times a day.

34. **If no**. At what age did your child stop breastfeeding?___________________________

35. At what age did you start giving your child other fluids/foods _____________________

How many times do you feed your child in one day? (meals and snacks combined)
1. Two to three times 2. Four to five times 3. Less than twice a day 3. More than 5 times

36. Please indicate according to the table, if and how your child consumes porridge.

Food/dish	1. Yes 2. No	Quantity consumed by child per day	Ingredients used (and amounts)	Preparation method
Porridge				

37. Are there any special foods you give your child at certain times? (Yes=1. No=2).

38. **If yes** which ones?____

39. Are there any foods your child does not consume or is forbidden to consume (Yes=1. No=2)

40. Which food(s) and for what reason? ____

Food frequency questionnaire

Household number ________________________

Child ID_______________________

Indicate the number of times your child consumes the following foods.

Food	Frequency of consumption (number of times food eaten by child)					
	Food code	**Daily**	**Weekly**	**Monthly**	**Occasionally**	**Never**
Porridge	01					
Beef	02					
Liver	03					
Goat meat	04					
Sheep meat	05					
Bovine blood	06					
Chicken	07					
Beans	08					
Fish	09					
Omena (dagaa)	10					
Milk (cow)	11					
Tea with milk	12					
Tea with no milk	13					
Cocoa	14					
Fats/oils	15					
Oranges	16					
Lemons	17					
Mangoes	18					
Pineapple	19					
Guavas	20					
Dark green leafy veges	21					
Other	22					

*For each food indicate only one frequency e.g.. Tea 2 times a day (2× in day column) or meat once monthly (1× in month column)

24 hour dietary recall.

Child ID:____________________________ Sex: M / F

Date of Birth:_____________________________ Age:______________(months)

24 hour recall intake record sheet

Dish				**Ingredients used in preparation**					**Child consumption**			**FINAL EXPRESSION**		
Time	Description of Dish	Total amount (ml)	Dish code	Description of ingredients	Amt used (ml)	Unit	Size	Ingrd code	Amt Served to child (ml)	Plate Waste	Amount consumed	Weight of ingredients:		
												g	g waste	g E.P.

Appendix 5: Anthropometric Data

Child ID____________________________

Date of weighing______/___________/_________

Age of child______________________ (months)   Sex (M/F)_______________________

Child ID	**DOB**	**Oedema (Y/N)**	**MUAC**			**Weight**			**Height**			
			1st	2nd	Av	1st	2nd	Av	1st	2nd	Av	