QUALITY OF FERMENTED PORRIDGE AND ITS EFFECT ON NUTRITIONAL STATUS OF MALNOURISHED CHILDREN ATTENDING MATERNAL CHILD HEALTH CLINIC IN THIKA DISTRICT HOSPITAL

A DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN APPLIED HUMAN NUTRITION OF THE UNIVERSITY OF NAIROBI

ANN WATETUTHUITA BSc. (FST)



SEPTEMBER 2010

DECLARATION

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

Date BASSAPAMBER 2010

Ann Watetu Thuita

DECLARATION BY SUPERVISORS

This dissertation has been submitted with our approval as university supervisors.

Date 13th Sept 2010

Prof. Jasper. K. Imungi, PHD Department of Food Science, Nutrition and Technology

Sharpance

Prof. S.K. Mbugua, PHD Department of Food Science, Nutrition and Technology

Date. 13/9/2010

DEDICATION

I dedicate this dissertation to the Lord and Saviour, Jesus Christ. In the potter's hand, He makes something out of nothing. Thank you, Father, for your faithfulness and mercy.

To my awesome husband Samuel Wambugu Ndiritu, my sons Simon Ndiritu and Joseph Thuita Wambugu and my great parents Joseph Thuita Kibacio and Jecinta Thuita, thank you for your support, love and joy during the period of my studies.

ACKNOWLEDGEMENT

I am deeply grateful to my supervisors Prof. Jasper. K. Imungi and Prof. S. K. Mbugua, who unreservedly offered unrivalled guidance without which this study would have been a veritable fiasco.

I also express my gratitude to Dr. Mbugua (Former DMOH Thika district), Mr Nduati (Medical Laboratory Technician; MLT, Thika District Hospital; TDH), Pascaline Wamiti (N.O III), Caroline Mwangi(DHRIO Thika District) for the great support during the entire data collection period. I would also wish to acknowledge Mr. P. Muthami and the entire staff of the Pilot Plant, Department of Food Science Nutrition and Technology, University of Nairobi for there assistance during the formulation of the flours. In addition I would also wish to acknowledge the support of Ms. Jane Njenga, Chief Technologist and entire laboratory staff Department of Food Science Nutrition and Technology, University of Nairobi for their help during the analyses of the formulated flours. I offer my tribute to the mothers of children who participated in the study, their generous participation that enabled my research to sail through smoothly.

Special appreciation goes to my husband Mr. Samuel Wambugu Ndiritu and my parents Joseph Thuita Kibacio and Jacinta Gathoni Thuita for their forbearance, inspiration and assistance throughout the period of writing the dissertation..

Finally, I am greatly indebted to all those people who have contributed in one way or another, consciously or unconsciously, towards the completion of this work.

i٧

TABLE OF CONTENT

DECLAR	ATION	. ii
DEDIC	CATION	iii
ABBREV	IATIONS/ ACCRONYMS	. x
OPERAT	IONAL DEFINITIONS	xii
ABSTRA	.CT	άv
CHAPTE	R ONE	1
INTROD	UCTION	1
1.0	Background information	1
1.1	Problem Statement	3
1.2	Study Justification	3
1.3	Objectives of the Study	4
1.3.1	Main objective	4
1.3.2	Specific objectives	4
1.4	Study Hypothesis	5
CHAPTE	CR TWO	6
LITERA	FURE REVIEW	6
2.0	Introduction	6
2.1	Protein Energy Malnutrition (PEM)	6
2.2	Micronutrient Malnutrition	7
2.2.1	Vitamin A deficiency	7
2.2.2	Iron	8
2.2.2.1	Dietary source, absorption and metabolism	8
2.2.2.2	Iron deficiency	9
2.2.3	Zinc deficiency	10
2.2.4	Strategies to reduce micronutrient deficiencies	.11
2.3	Infant and Young Child Feeding in Developing Countries	.13
2.4	Ingredients used in Complementary Foods Formulation	14
2.4.1	Finger millet	14
2.4.1.1	Anti-nutrients and toxic components in cereals	15
2.4.2	Amaranths	16
2.5	Fermentation in Increasing Nutrient Availability	.21
2.6	Standards for Complementary Foods	.22
2.7	Dietary intake	.23
2.8	Biochemical Assessment of Nutrient Intake	.24
CHAPTE	ER THREE	.25
STUDY	SETTING AND STUDY METHODOLOGY	25
3.0	Study Setting; Thika District Profile	25
3.1	METHODOLOGY	.28
3.1.1	Study Design	.28
3.1.2	Study population and sampling frame	28
3.1.3	Sample size determination	.30
3.1.4	Sampling procedure	.30

	3.1.5	Inclusion and exclusion criteria	31
	3.1.6	Screening and screening procedure	31
	3.1.7	Sensitization of the parent for consent and recruitment of study children	33
	3.1.8	Allocation of study group	33
	3.2	Formulation of the Flours	33
	3.2.1	Non fermented Amaranths-finger millet- pumpkin porridge flou	ır
	process	sing	34
	3.2.2	Fermented amaranths-finger millet- pumpkin porridge flour processing	34
	3.3	Chemical and Microbial analysis	35
	3.3.1	Determination of proximate composition of the porridge flours	35
	3.3.1.1	Determination of moisture	35
	3312	Determination of crude protein	38
	3313	Determination of Crude Linids	38
	3314	Determination of crude fiber	39
	3315	Determination of total ash	39
	3316	Determination of soluble carbohydrates	40
	332	Determination of total Energy	40
	333	Determination of nH	40
	334	Determination of pre-	40
	335	Determination of iron and zinc	40
	336	Microbial analysis	41
	3.3.0	Sensory evaluation of the porridges	42
	3.4	Baseline Data Collection	42
	3.4.1	Social demographic demographic charcteristics of household members	42
	3.4.2	Anthronometric measurement (Weight and height measurements)	43
	3 4 3	A ge determination	43
	3.4.4	Blood sample collection	43
	3 4 5	Determination of total serum protein serum zinc and serum retinol	44
	3/151	Serum protein determination	44
	3 1 5 7	Serum zinc determination	45
	3.4.5.2	Serum retinol determination	45
	2.5	Food consumption patterns and Dietary Intake	46
	3.5	Deworming and Malaria Prevention	46
	3.0	Easting trials	46
	2.7	Deta collection and management	17
	J.O LIADTE		47
	HAP1E		49
ĸ	A	Drawingste composition of the normidae flours	49
	4.1	Proximate composition of the portage flours	47
	4.2	Microbial Quality of the Flours	51
	4.5	Acceptability Score of the Porridge Samples	54
	4.4	Baseline characteristic of Household of the study children	55
	4.4.1	Social demographic characteristics of the nouseholds of the study children	122
	4.4.2	Mother's profile	38
	4.4.3	Morbialty status of study children	01
	4.5	Dietary intake	05

	4.6	Dietary intake of study children	.64
	4.6.1	Dietary adequacy of energy, protein, iron, zinc and vitamin A as RDA	.68
	4.7	Knowledge of mothers on fermentation	.73
	4.8	Morbidity experience of study children during feeding	.73
	4.9	Consumption and sharing of the porridge	.74
	4.10	Nutritional status of the study children	.75
	4.11	Micronutrient Status	.81
	4.11.1	Haemoglobin levels	.81
	4.11.2	Change in total serum protein	.81
	4.11.3	Changes in total serum zinc	.82
	4.11.4	Change in serum retinol	.83
CI	HAPTE	ER FIVE	.86
C	ONCLU	JSIONS AND RECOMMENDATIONS	.86
	5.1	CONCLUSIONS	.86
	5.2	RECOMENDATIONS	.86
R	eferenc	es	.88

LIST OF TABLES

Table 2.1 Proximate and mineral content of finger millet, Amaranth and Pumpkin20
Table 3.1: Demographic profile of Thika District 26
Table 3.2 Top five diseases in Thika district 27
Table 3.3: Recommended dietary Allowance (RDA) of the children aged 1-3yrs34
Table 4.1 Proximate composition, iron, zinc and beta carotene content of the
fermented and non fermented finger millet amaranth pumpkin porridge flour per 100
gm
Table 4.2 Microbial quality of the flours and pH
Table 4.3 Mean rating for sensory characteristics of the porridge samples55
Table 4.4 Distribution of household by size 58
Table 4.5 Occupation of the mothers 59
Table 4.6 Education status of the mothers 59
Table 4.7 Correlation between social demographic characteristic and nutrition status
of children
Table 4.8: Marital status of the mothers 61
Table 4.9 Type of illness suffered by study children
Table 4.10 Other characteristic of study children 62
Table 4.11 Proportion of children who had received Vitamin A supplementation less
than 6 months prior to the study
Table 4.12 Food consumption frequency
Table 4.13 Dietary intake of study children before and after supplementation with
porridge
Table 4.14 Dietary intake of study children without porridge supplementation71
Table 4.15 Dietary intake of study children with porridge supplementation
Table 4.16 Knowledge of mothers on fermentation 73
Table 4.17 Morbidity experience during feeding for the two groups of children74
Table 4.18 Sharing of porridge flour by children 74
Table 4.19 Prevalence of underweight before feeding and after feeding of study
group .77
Table 4.20 Prevalence of stunting before feeding and after feeding of study group 77
Table 4.21 Prevalence of wasting before feeding and after feeding of study group 78
Table 4.22 Change in anthropometry during the intervention
Table 4.23 Mean Z score of study children at baseline and post intervention
Table 4.24 Nutrition status distribution by age at baseline and post intervention80
Table 4.25 Haemoglobin levels of study children 81
Table 4.26 Mean change of total protein (g/dl) of the study children 82
Table 4.27 Change in serum zinc
Table 4.28 Change of serum retinol (umoll 1) of the study children 85

LIST OF FIGURE

Figure 3.1 Study Design	29
Figure 3.2 Sampling scheme	32
Figure 3.3 Processing of Non Fermented amaranth-finger millet-pumpkin po	orridge
flou	36
Figure 3.4 Processing of Fermented amaranth-finger millet-pumpkin porridge f	flour.37
Figure 4.1 Education level of household the members of the study children	56
Figure 4.2 Occupation status of household members of the study children	57

APPENDICES

Appendix 1: Data collection tools	
Appendix 2: Laboratory request form	
Appendix 3 Training Curriculum	
Appendix 4: Informed Consent Form	

ABBREVIATIONS/ ACCRONYMS

AAS	Atomic Absorption Spectophotometry
AOAC	Association of Analytical Chemistry
AOP	Annual Operation Plan
BOSTID	Board off Science and Technology for International
	Development
CAC/GL	Codex Alimentarius Commission
CBS	Central Bureau of Statistic
СНО	Carbohydrates
CKDAP	Central Kenya Dry Area Project
DDMS	District Director of Medical Services
DHRIO	District Health Record and Information Officer
DMOH	District Medical Officer of Health
DNO	District Nutrition Officer
FAFMPP	Fermented amaranth finger millet pumpkin porridge
FAO	Food Agricultural Organization
GMP	Growth monitoring and promotion'
HCL	Hydrochloric acid
IDA	Iron Deficiency Anaemia
IMCI	Integrated Management of Child Illnesses
INACG	International Nutritional Anaemia Consultative group
KDHS	Kenya Demographic Healthy Survey
КОН	Potassium hydroxide
LLITN	Long lasting insectside treated net
MCH	Maternal child health
MLT	Medical Laboratory Technologist
MI	Micronutrient Initiative
МОН	Ministry of Health
NAOH	Sodium hydroxide
NHCS	National Health Centre for Statistics

NAOH	Sodium hydroxide
NHCS	National Health Centre for Statistics
NFPF	Non Fermented porridge flour
NO	Nutrition Officer
PEM	Protein Energy Malnutrition
RDA	Recommended Daily Allowance
TDH	Thika District Hospital
UNCEF	United Nation Children Emergency Fund
USAID	United State Agency for International Development
VAD	Vitamin A Deficiency
VBRA	violet red bile agar
WHO	World Health Organization

OPERATIONAL DEFINITIONS

Anaemia- Manifestation of nutritional deficiencies and or other diseases that accelerate haemolysis of red blood cells or interfere with haemoglobin production or frank blood loss

Anthropometry - Physical measurements of weight, length /height, mid upper arm circumference, head circumference.

ENA-Nutrisurvey - A series of microcomputer packages used to assess nutritional status such as height for age, weight for height and weight for age

Erythropoiesis – Depletion of red blood cells of iron

FPF group – refers to those children whose meal was supplemented with fermented flour porridge

Food security – Access by all people at all times to adequate and quality food for active healthy life.

Hematocrit – The percentage of total blood volume occupied by red blood cells.

Household – Refers to one person who lives alone or a group of persons, related or unrelated who share food or make common provisions for food and other essentials for living.

Hypozincaemia – Low zinc serum levels.

Iron deficiency - Depleted body iron stores without regard to the degree of depletion or to the presence of anaemia

Iron deficiency anaemia - Severe depletion of iron stores that results in low haemoglobin and small, pale, red blood cells.

Malnutrition – Condition of impaired development or function caused by either a long term deficiency or an excess in energy and /or nutrient intake, the latter representing a state of over-nutrition.

NFPF Group - Refers to those children whose meal was supplemented with non fermented porridge flour during the study

Nutritional status – This refers to stunting underweight and wasting for the purposes of this study.

Underweight - Weight-for-age is a composite index of height-for-age and weight-forheight and, thus, does not distinguish between acute malnutrition (wasting) and chronic malnutrition (stunting).

Wasting - Weight-for-height measures body mass in relation to body length and describes current nutritional status.

Stunting – Deficit in linear growth achieved pre-and post natal. This indicates long term cumulative effect of inadequate nutrition

Z-Score or standard deviation - The deviation of anthropometric value(s) for an individual child from the median value of the reference population. National Centre for Health statistics of the United States of America NHCS /World health Organization (WHO), divided by the standard deviation of the reference population.

ABSTRACT

A study was carried out to evaluate the effect of feeding malnourished children with fermented amaranth-finger millet- pumpkin composite porridge (FP) on their nutritional, vitamin A, zinc and iron status. The feeding study was done in comparative trials with non-fermented porridge (NFP). The children aged 7-24 months were sampled from those attending Maternal Child Health clinics (MCH) at Thika District Hospital (TDH). A total of 61 children whose weight for age Z-score was between -3.00 to -2.00 SD were recruited. The children were randomly allocated to two groups: children for fermented porridge flour (FPF group) and children for non- fermented porridge flour (NFPF group). The two porridges were prepared as flours and the ration for the children supplied to the mothers with instructions to prepare and feed the children in their homes. The feeding period lasted six weeks. The nutritional, protein, vitamin A, iron and zinc status of the children were determined at the start and end of the study. The dietary intake by the children and the morbidity of the children were also assessed at the two points. Laboratory analysis of the nutritional and the microbiological quality of the flours were carried out.

Result showed the energy content of NFPF to be 365Kcal/100g, slightly lower than that of FPF at 382Kcal/100g. The protein contents of the flours were 8.8g/100g for NFP and 9.2g/100g for FPF and this contributed to 58% and 61% of the RDA for the children. Beta-carotene content for NFPF was 146 µg/100g; significantly lower than that of the FPF at 180 µg/100g.

The 24-hour dietary recall showed that the study children met the Recommended Dietary Allowance (RDA) for energy, protein, iron and zinc after supplementation of their diets with the porridge flours. The RDA for protein and iron were significantly higher after supplementation with FPF than after supplementation with NFPF.

The study children were undernourished at the start but after intervention the proportion of those undernourished reduced significantly (P<0.05). The underweight,

wasting and stunting of the FPF group reduced from 50.8% to 4.9%, 40.8% to 16.4%and 45.8% to 1.6% respectively while those supplemented with NFPF reduced from 48.8% to 31.8%, 42.6% to31.1% and 44.2% to12.9% respectively. The change in weight was significantly higher (p<0.05) for FPF group than for NFPF group. However, the change in height was not significant between the groups.

There was no significance difference in nutritional status of the children between the two groups before feeding. However there was a significance difference (p<0.05) in nutritional status after feeding with FPF group having a mean weight for age Z-score of 0.71, height for age Z-score of -0.52 and weight for height Z-score of 1.30 compared to mean weight for age Z-score of -1.31, height for age Z-score of -1.44 and weight for height Z-score of -0.79 for the NFPF group.

There was no significant difference in mean haemoglobin levels at a mean of 9.53 g/dl and 9.47g/dl of the children in FPF and NFPF groups respectively at start of feeding. However, there was a significant difference in mean haemoglobin levels (11.60g/dl and 10.79/dl) between FPF and NFPF groups after feeding. There was no significant difference (p>0.05) in the mean total serum protein concentration (7.15 g/dl and 7.1 g/dl) for FPF and NFPF group at start between the groups. There was, however, a significant difference (p<0.05) in the mean total serum protein (7.46 g/dl and 7.28 g/dl) for FPF and NFPF group respectively between the study groups after end of study. There was no significant difference (p>0.05) between the mean zinc levels (66.74 μ g/dl and 65.75 μ g/dl) of FPF and NFPF groups respectively at start of study. However, after feeding there was a significant difference (p<0.05) between the mean zinc levels (71.64 μ g/dl and 69.23 μ g/dl) of the groups.

There was no significant difference between the increases in the serum retinol of the groups post intervention. Before feeding, 18.2% of children in FPF and 10.9% in NFPF had normal levels of serum retinol. After the intervention 34.5% of children in FPF and 27.3% of NFPF children had normal levels of vitamin A, but the changes

were not significantly different between the groups.

The study concludes that fermented composite porridge flour (FPF) improved the nutritional, protein, vitamin A iron and zinc status of the children, better than the unfermented composite porridge flour (NFPF).

CHAPTER ONE INTRODUCTION

1.0 Background information

Nutrition status is the result of complex interaction between food consumption and the overall status of health and care practices (Disilvestro, *et al.*, 2004). Malnutrition is a nutrition disorder or condition resulting from faulty or inadequate diet intake. According to WHO Global Database on child growth, covering 87% of under 5-years old in developing countries, 80% of the children affected by protein energy malnutrition (PEM) live in Asia, mainly southern Asia, 15% in Africa and 5% in Latin America (Disilvestro *et al.*, 2004).

Poor nutrition status is one of the most important health and welfare problems facing Kenya today and afflicts mainly women and children. Malnutrition leads to serious consequence on the physical and mental development in children (CBS, 2004). Protein energy malnutrition (PEM), iron deficiency anemia (IDA), vitamin A deficiency (VAD), and Zinc deficiency are among the major forms of malnutrition in Kenya (MOH, 1999; UNCEF, 1999). PEM is the most critical in children under five years and is usually coupled with infections that result in lots of disability and high motility (FSAU, 2003). However the consequence of micronutrient deficiencies especially that of iron, zinc, vitamin A and iodine in these children as well as pregnant mothers can not be underestimated.

In Kenya child mortality rates and malnutrition remain high in spite of the Government's initiative to provide quality health care. About 30% of under five children suffer from stunting, 6% from wasting and 20% are underweight. The prevalence of these problems is most critical in rural areas, drought stricken areas, and poor urban households (CBS, 2004).

To alleviate the problem of PEM and micronutrient malnutrition in infants and young children, the focus should be on use of locally formulated complementary foods, because these are affordable and culturally and socially acceptable. Foods formulated from local cereals and vegetables as a complement to breast milk can provide the deficit in dietary energy, protein and the micronutrient needed to meet increasing physiological requirements during growth and development (Tomkins and Watson, 1993).

Traditional technologies such as fermentation and malting have been used mainly to improve palatability of foods. However, these technologies serve to hydrolyse starch to simple sugars and reduce levels of anti-nutrient, thereby boosting the nutritional adequacy of foods. (Oniango, *et al.*, 2002). Pseudo cereals like amaranth with protein of high quality (Railey, 1999) can also be added to boost of cereal based formulation.

In Kenya supplementation programs to combat VAD in children 6-59 months already exist but the coverage is still very low with Thika district recording 27 %. MOH, 2009. These programs are short term, funded mainly by donors such as United Nation Emergency Fund (UNCEF) and Micronutrient Initiative (MI) and are unsustainable. Long term sustainable interventions are therefore required. Sustainable interventions are best based on fortification and food diversification. Fortification of oil and margarine with Vitamin A in Kenya is already underway. but this benefit mainly population in the urban areas. Consumption of vitamin A rich vegetable and fruits such as dark green leafy vegetables, pumpkins, yellow flesh fruits would reach wider segment of the population.

Unmodified gruels from maize and other cereals are the most commonly used as complementary foods for children. These gruels are usually bulky with low caloric density. The gruels are also sometimes high in anti- nutrient that may limit micronutrient bioavailability; thereby leading to deficiencies.

2

1.1 Problem Statement

In the developing countries more so those of the Sub-Saharan Africa growth of children begins to falter with the initiation of complementary feeding around the age of 7 months. This continues as the child grows and finally leads to protein energy malnutrition (PEM) and micronutrient deficiencies. This has been attributed due to use of complementary food based mainly on the predominantly cereal and starchy roots. These foods are bulky with low nutrient density. Children weaned on these porridges which are thick and pasty find it difficult to consume enough to satisfy dietary nutrient requirements. This problem is aggravated by attempts to dilute the porridges to level of consistency tolerable to the children.

Existence of anti-nutrient such as tannins and phytates may also severely limit bioavailability of some nutrients. Further cereal and cereal products are often limiting in some micronutrient. The protein quality of the foods is often low because it lacks some essential amino acids. Finally, these foods are often lacking in vitamins such as vitamin A. The problem can be alleviated by using processing and modification technologies such as hydrolysis, fermentation and germination.

1.2 Study Justification

Malnutrition (protein energy malnutrition and micronutrient deficiency) remains high in children. The prevalence of these problems in Kenya is most critical in the rural areas which often experience drought period resulting in crop failure and where most of the poor households live (CBS, 2004). Use of local food products like cereals, vegetables, fruits and legumes in fabrication of complementary foods is a feasible and sustainable approach to malnutrition in children but most of these foods especially cereals and legumes are high in anti-nutrients that bind nutrient thus making them unavailable after consumption..

Fermentation increases the nutrient density of the porridges by breaking down the

starch (Oniango, et al., 2002). Fermentation also hydrolyzes anti-nutrients such as phytates rendering micronutrient such as iron, zinc and vitamin A available and increases protein quality. Cereals are limiting in amino acids lysine. Addition of amaranth which is rich in lysine and methionine improves protein quality (Sehmi, 1993; Karen 1999). The cereals and amaranths, however, are poor in vitamin A which is a micronutrient of public health concern. The vitamin A content is improved by addition of pumpkin which is rich in pro-vitamin A, carotenoids (Sehmi 1993).

Thika District Hospital being a referral hospital attends to all the population of Thika District and its environment. It records a high rate of malnutrition with 53 admission of under five per month due to severe malnutrition and 27% cases of moderate malnutrition registered at MCH clinic monthly from the Thika District under five years children attending GMP with the most affected being children under the age of 2years. Due to this reason Thika District hospital was chosen as the study site for the intervention.

1.3 Objectives of the Study

1.3.1 Main objective

To assess the effect of lactic acid fermented composite porridge from amaranths, finger millet and pumpkin in improving the nutritional status of moderately malnourished children aged 7-24 months.

1.3.2 Specific objectives

- 1. To determine socio-demographic and morbidity status of children aged 7-24 months.
- 2. To assess the dietary intake of children.
- 3. To formulate acceptable fermented and non-fermented porridge flours with sufficient supplemental protein, energy and micronutrients.

- 4. To determine the proximate composition, vitamin A as beta-carotene, iron and zinc contents of the porridge flours.
- To assess the nutritional, vitamin A, iron and zinc status of the children aged
 7-24 months before and after feeding with the porridge flours.

1.4 Study Hypothesis

Malnourished children fed on fermented porridge improve their nutritional status better than those fed on non fermented porridge flour.

5

CHAPTER TWO LITERATURE REVIEW

2.0 Introduction

Nutrition is a key concern for the fulfillment of socially and economically sustainable development as well as health of individuals. Adequate nutrition and health during the first year of life is fundamental to the attainment of millennium development goal (MDG) for child survival and prevention of malnutrition. Poor nutrition during the first year of life has both an immediate and long term consequence. Immediate consequence include significant morbidity and mortality and delayed physical and mental development and long term consequence which include impaired intellectual performance, reduced work capacity and reproductive capacity as well as increased chronic diseases (Tomkins and Watson 1993).

Protein energy malnutrition (PEM), iron deficiency anemia (IDA), vitamin A deficiency (VAD) and Zinc deficiency are currently the forms of malnutrition of concern globally (UNCEF 1999; MOH 1999; Tina *et al.*, 2007). Up to 55 % of deaths of children in developing world are attributed to under nutrition (Disilvestro, *et al.*, 2004). Under nutrition is the most common form of malnutrition among poor people in developing and developed countries. Currently about 4 million African children under five years of age who die annually are undernourished (Disilvestro, *et al.*, 2004).

2.1 Protein Energy Malnutrition (PEM)

Protein energy malnutrition (PEM) is a form of malnutrition caused by intake of diets which are extremely deficient in energy or protein, generally accompanied by an illness (Disilvestro, *et al.*, 2004 and Tina *et al.*, 2007)). PEM is characterized by growth failure both in weight and height accompanied by thick weak and wasting muscles; oedema which result from accumulation of fluid in the tissue causing them

to be soft and spongy ; skin change in colour , drying, peeling and the eventual formation of ulcers that heal slowly and therefore act as point of entry for other ineffective organism; the hair becomes dry and sparse and may lose its pigment ; loss of appetite, diarrhoea resulting to severe dehydration and loss of sodium and potassium; enlarged liver, anaemia and increased susceptibility to infection (Guthrie and Piccano, 1995). PEM also has a depressing effect on immune system leading to lowered immunity, but the effect to immunity can occur even with mild levels of malnutrition (Tomkins and Watson, 1993).

2.2 Micronutrient Malnutrition

Vitamins and mineral are essential for human health and development (Disilvestro *et al.*, 2004). These nutrients are important for chemical processes that ensure survival, growth and functioning of vital human systems. High rates of micronutrient malnutrition are observed in the world (Disilvestro *et al.*, 2004). Vitamin A deficiency (VAD) accounts for 9% of child deaths, iron deficiency causes half of anemia and zinc deficiency accounts for 5.5% of child deaths (Tina *et al.*, 2007).

Available global data estimates that 25% of pre school children suffer from VAD while an estimated 20% is at risk of zinc deficiency (Tina *et al.*, 2007). Deficiencies of vitamin A, zinc, and iron contribute to infant and child mortality and for those who survive the deficiencies cause disabilities mainly resulting from illness with delayed recovery (MOH, 1999; Tina *et al.*, 2007). VAD causes blindness of up to 250,000 to 500,000 preschool children in the World each year (Tina *et al.*, 2007).

2.2.1 Vitamin A deficiency

Vitamin A refers to preformed retinoid found in animal foods. In plant food, the vitamin occurs as a large number of provitamin A carotenoids which hydrolyze to yield bodies retinoid in animal. Vitamin A is required for vision, growth and development of many types of tissue and for immunity. Also lack of adequate vitamin A in the diet leads to vitamin A deficiency (VAD). VAD leads to night

7

blindness. Mucus forming cell also deteriorates and are no longer able to synthesis mucus, the essential lubricant used throughout the body. The eye especially the cornea, is adversely affected by the loss of mucus, which keeps the eye surface moist and washes away dirt and other particles that settle on the eye. Deterioration of the eye leads to bacterial invasion. Conjunctiva xerosis and bitot spots appear as first symptoms of VAD. Xeropthalmia leads to irreversible blindness. VAD also produce skin changes referred to as follicular hyperkeratosis. It also leads to poor growth (Disilvestro *et al.*, 2004). Also severe fat- mal-absorption such as incase of diarrhoea and pancreatic insufficiency leads to VAD.

VAD in children is most common often during the post weaning period. In Kenya vitamin A deficiency is at 14.7% for acute VAD and 6.12 % for moderate VAD (MOH 1999). In Kenya vitamin A supplementation as capsules is administered to children between 6- 59 months every six month to combat VAD although not all children are reached. Finding a suitable food to improve intake is required for a long lasting solution to combat VAD.

2.2.2 Iron

2.2.2.1 Dietary source, absorption and metabolism

The process of iron absorption can be divided into three stages namely:

- 1. Iron intake,
- 2. Intra enterocyte transport,
- 3. Storage and extra enterocyte transfer

During the intestinal phase of digestion, iron binds into specific mucosal membrane sites, it is internalized and then either retained by mucosal cell or transported to basolateral membrane where it is bound to transferrin in the plasma pool. The process of iron absorption is controlled by intraluminar mucosal and somatic factors. A multitude of intraluminar factors affect amount of iron available for absorption as either inhibitors or promoters. The mucosal factors influencing iron absorption include the amount of mucosal surfaces and intestinal mortality, while somatic factors include erythropoesis and hypoxia (O'Dell and Sunde, 1997)

No absorption of iron occurs in the mouth, esophagus, or stomach. However the stomach contributes hydrochloric acid (HCL) which helps dislodging the iron bound to protein by denaturing protein and also helps in solubilization of iron and reduction from ferric to ferrous state. This reduction is necessary as majority of iron in the diet is found in relatively insoluble ferric form (non heme) and is poorly absorbed (O'Dell and Sunde, 1997). Also gastric pepsin liberates some non heme iron from digestive through protease activity. The combined action of gastric acid and pepsin, account for slightly less than one half of the release of conjugated iron, and the reduction of one third of total dietary ferric iron. Majority of the iron absorption takes place in the duodenum and upper jejunum (O'Dell and Sunde, 1997). Once iron has been absorbed it is transported through blood bound to transferrin. The iron ion transported by transferrin can be released to the cell for manufacture of iron containing enzyme and other proteins, released to bone marrow for use in haemoglobin synthesis or deposited within the ferritn and haemosiderin, the iron storage site of the body (Guthrie and Piccano, 1995).

If a cell is in need of iron, it produces more transferrin receptors to enable collection of more iron from the circulating supply. If the amount of iron absorbed from the diet and that obtained from breakdown of red blood cell is insufficient to meet body needs, iron is removed from storage site and transported to the site of demand bound to transferrin molecule. Iron is stored in the body in the form of transferrin, in the bone marrow, liver, spleen and kidney. Symptoms of iron deficiency occur only when the body store is depleted (Guthrie and Piccano, 1995).

2.2.2.2 Iron deficiency

Iron plays an important role in many parts of the body including immune function, cognitive development, temperature regulation, energy metabolism and work performance.

Iron deficiency and iron deficiency anemia (IDA) are common world wide and affect estimated one billion people in developing and developed countries (Disilvestro *et al.*, 2004). Iron deficiency in early childhood has a significant negative effect on child's physical and intellectual development (Mannar, 2007). Iron deficiency is one of the most important causes of anemia. In developing countries anemia is mostly exacerbated by malaria and other parasitic infections (Tina *et al.*, 2007). Other factors that contribute to risk of childhood anemia are consumption of staples with low iron bioavailability such as cereals. In addition diarrhoea and maternal anemia may also be a contributing factor (MOH, 1999).

If iron deficiency is prolonged in early infancy it impairs the cognitive development (Tina *et al.*, 2007). In severe iron deficiency, there is not enough to make all hemoglobin needed. This level of IDA can be detected by measuring haemotocrit levels. Haemotocrit measures the percent of blood volume occupied by red blood cells. Values of 34- 37 % suggest IDA. Blood haemoglobin can also be used and values less than 10 to 11g/dl also suggests IDA.

Anemia impairs energy production as aerobic respiration cannot occur without oxygen. Signs of lack of sufficient energy include fatigue and difficulties in mental concentration. Anemia also impairs immune function (Disilvestro *et a.*, 2004). Globally the prevalence of anemia in preschool children is 47.4% (Benoist *et al*; 2007). In Kenya the prevalence of moderate and severe anemia among children are 19.2% and 54.2% respectively (MOH 1999).

2.2.3 Zinc deficiency

Zinc has been identified as essential component of metalloenzymes, polyribosomes, cell membranes and cellular function and plays a central role in cellular growth, tissue differentiation, protein synthesis and immune function.

The main source of zinc is animal products. The daily requirement of zinc may be difficult to meet in population who consume staple foods having high phytate content that reduce zinc bioavailability (Bruno *et al.*, 2005; Grases *et al.*, 2001). Deficiency of zinc was first demonstrated in animal models, followed by evidence in human which showed that zinc deficiency seriously limits growth in infants and children and decreases resistance to infection (Bruno *et al.*, 2005).

The extent of zinc deficiency worldwide is not well documented. Infant and young children are the most vulnerable to zinc deficiency. The health consequences of severe zinc deficiency are easily recognized and include dermatitis, growth retardation, delayed sexual maturity, and recurrent infections. Mild and moderate deficiencies are difficult to diagnose, because signs and symptoms, such as increased susceptibility to infection and reduced growth rates, are also seen in many other nutrient disorders and common child hood infection.

Zinc deficiency is among the most important causes of morbidity in developing countries. The risk of zinc deficiency is considered to be of public health concern when the prevalence of low serum zinc concentration is as greater than 20%, when prevalence of inadequate diet intake is greater that 25% and when prevalence of low height or length for age is greater than 20%. In such a case an intervention to improve population zinc status is advised (Bruno *et al.*, 2005).

2.2.4 Strategies to reduce micronutrient deficiencies

Strategies to reduce micronutrient deficiencies include supplementation, fortification, dietary diversification, fermentation and nutrition education among others. Strategies like supplementation and fortification are expensive and not sustainable. Therefore sustainable solutions need to be sought to prevent micronutrient deficiencies. Such solutions include food diversification, food fortification, nutrition education and advocacy and food preparation method that retain and lead to better nutrients release; these include fermentation, malting and sprouting.

Food diversification involves increase in production and consumption of foods rich in micronutrient. Promoting food production and increased consumption at community level for example through home gardening programs and encouraging a diet diversified to include natural and fortified micronutrient rich foods would be feasible. Also key to this strategy is changing people's dietary choices and practices. Food diversification requires planner to choose the most feasible and acceptable behaviour to promote and overcome identified barriers to new ideas and support of positive practice towards enhanced nutrition. This can be popularized through natural campaign, media, community awareness, use of extension agent and religious leaders. Dietary diversification can improve household food security and overall quality of diet as well as address multiple nutrition deficiency if well implemented, however it require aggressive sensitization of the community due to social cultural as well as economic constrain (USAID, 2000).

Supplementation involves giving of mineral or vitamins in form of concentrated form (tablet or capsules) to reduce deficiency in a population. Vitamin A supplementation is increasingly integrated in immunization programmes. In Kenya the ministry of health (MOH) in collaboration with UNCEF and MI have introduced child, mother health and nutrition weeks in the month of June and November to address child health including vitamin A deficiency, immunization, growth monitoring and deworming among others. This is a medium or a long term strategy to reduce deficiencies in children but the coverage still remains low indicating that not all population especially the vulnerable are reached.

Food fortification refers to addition of one or more nutrient to a food. The main objective of food fortification is to increase the level of consumption of the added nutrient to improve nutrition status of a given population. The primary role of food fortification is prevention of deficiency thereby avoiding the occurrence of disorder that lead to human suffering and social economic disadvantages. In development of a food fortification programme there is need for determination of prevalence of a deficiency, selection of appropriate intervention, calculation of dietary intake level of micronutrient, and daily consumption of food vehicle selected or level of fortificant to be added ands even technology development. It is also an expensive process as it requires promotion campaigns to improve consumer acceptance, development of legislation and regulation for mandatory compliance. It also requires selection of good vehicle and fortificant. The vehicle chosen should reach most of the population, shouldn't react with fortificant, should not react with fortificant and should not interfere with bioavailability of nutrient in the body. Also the fortificant should have good bioavailability during normal shelf life of the fortified food, have affordable cost and acceptable for the intervention to be feasible (Lotfi *et al.*, 1996))

Fermentation, malting and sprouting are biotechnological processes that can also be utilized to enhance product nutrition. They reduce anti-nutrient thus making nutrient available (Bruno *et al.*, 2007; Grases *et al.*, 2001; Oniango *et al.*, 2002). Fermentation also breaks down some of the fibre in the cereals and increases iron absorption (Meitzner and Price, 1996, Oniango., *et al* 2002). This is a cheap traditionally practiced and can be utilized to improve on micronutrient bioavailability of cereals used in making complementary foods for young children. It also leads to increased nutrient density of the food, as well as increased protein quality hence serving as one way of combating macro and micro nutrient deficiencies.

2.3 Infant and Young Child Feeding in Developing Countries

In developing world the problem of PEM exists among infant and children who are mostly complemented on cereal foods (Hofuande and Underwood, 1987; Oniango et al., 2002). These cereal products when cooked have low caloric density and they are bulky and are always fed in inadequate amounts. Weaning period is critical stage in child growth that often leads to PEM due to inadequate caloric intake. The cereals are mainly prepared as porridge diluted with water due to its bulkiness for the child to be able to swallow. This dilution often reduces the caloric density of the porridge

making it difficult to meet the recommended daily intake (Meitzner and Price, 1996).

Cereals are often used as complementary feeds and their demand is growing. The cereals include finger millet, maize, and wheat (Sehmi, 1993). They are high in antinutrient such as phytates and tannins that bind micronutrient like iron, zinc (Oniango et al., 2002). They also lack the essential amino acid lysine. There is need for combining the cereal with legumes or other seeds such as amaranths in order to improve protein quality (Rathode, 1991). Amaranth seeds have a fairly high protein content of good quality and rich in lysine. When used to supplement cereals they upgrade the overall protein quality of the diet (Rathod, 1991; Railey, 1999). It is also high in protein (15-18%) and contain respectable amount of lysine and methionine, two essential amino acid that are not frequently found in cereals (Railey, 1999).

In Kenya traditional preparation methods of cereal involve fermentation, malting and germination (Oniango et al., 2002). They are the most commonly used traditional methods of food processing (Oniango et al., 2002, Hemalatha et al., 2007). Fermentation reduces the phytic acid content making micronutrient such as zinc and iron available, which improves the nutrition quality of the diet (Meitzner and Price 1996; Oniango et al., 2002).

2.4 Ingredients used in Complementary Foods Formulation

2.4.1 Finger millet

Finger millet (*Eleusine coracana*) also known as African millet, wimbi (Swahili), bulo (Uganda) and telebun (the Sudan) is a tufted annual crop growing 40 to 130 cm tall, taking between 2.5 to 6 months to mature. It has narrow, grass like leaves and many tillers and branches (BOSTID, 1996). It probably originated in the Highlands of Uganda and Ethiopia where farmers have grown it for thousand of years (BOSTID, 1996). It is an important staple food in parts of Eastern and Central Africa and India. Finger millet can be stored for long periods without insect damage and thus it can be important during famine. Finger millet is a source of carbohydrate,

protein and mineral that is comparable to other cereal grains. It has a protein content of 7.4% (Sehmi, 1993; BOSTID 1996; Madibela and Modiakgotia 2004), which is comparable to that of rice (7.5%). Information on other nutrients in finger millet is shown in Table 1. Further the grain of finger millet is highly palatable and of excellent dietary sources of methionine, an amino acid limiting in diets of millions of poor who live on starchy foods such as cassava, polished rice and maize meal (BOSTID, 1996). However fingermillet has high levels of anti -nutrients like phytates and tannins, which reduce nutrient bioavailability but the effect of these can be reduced by suitable processing methods such as germination and fermentation (Sripriya *et al.*, 1997; Oniango *et al*; 2002). The grain is normally ground into flour and boiled to make porridge and can also be baked into bread. The flour can also be fermented and the grains be used as food as these two process are beneficial because they make the product easily digestible and nutritious (BOSTID, 1996).

Fermentation of finger millet for about 48 hours decreases pH due to formation of lactic acid and other acids and breaks down the starches thus increasing total sugars and free amino acids. It also decreases phytate contents. The phytates x Ca/Zn molar ratio decreases and this is indicative of increase in Zinc bioavailability (Sripriya et *al.*, 1997; Oniango *et al.*, 2002). The millet, however, is poor in protein compared with other common cereals. Differences in amino acid composition in different varieties of finger millet are large. Lysine is the limiting amino acid (*http://www.blackherbals.com/bd14538;* Sehmi 1993). The nutrition composition of finger millet is as shown in Table 1.1.

2.4.1.1 Anti-nutrients and toxic components in cereals

Cereals and other plant foods may contain significant amounts of toxic or antinutritional substances. Most cereals contain appreciable amounts of phytates, enzyme inhibitors, and some cereals like sorghum and millet contain large amounts of polyphenols and tannins. Some of these substances reduce the nutritional value of foods by interfering with nutrient bioavailability, and digestibility of proteins and carbohydrates. Relatively little is known about the fate of anti-nutrients and toxicants in traditional fermented foods (*http://www.blackherbals.com/bd14538*).

Phytates represents a complex class of naturally occurring phosphorus compounds that can significantly influence the functional and nutritional properties of foods. Phytic acid, myo-inositol 1, 2, 3, 4, 5, 6-hexakis (dihydrogen phosphate), is the main phosphorus store in mature seeds. Phytic acid has a strong binding capacity, readily forming complexes with multivalent cations and proteins. Most of the phytate-metal complexes are insoluble at physiological pH. Hence phytate binding renders several minerals biologically unavailable to animals and humans. Food preparation methods such fermentation should be used that reduce their content as (http://www.blackherbals.com/bd14538).

2.4.2 Amaranths

Amaranths can be grown in many drought prone semitropical countries to produce high protein flour which can be used to supplement cereal flour used for preparation of complementary foods (Mitra *et al.*, 1993; Eggum *et al.*, 2005). The variety Amaranths hypochondriacus is the most common used and has a protein content of approximately 15%. The protein is of high quality and rich in amino acid limiting in cereal, lysine and sulfur containing amino acid methionine which is usually low in legumes (Mitra *et al.*, 1993; Eggum *et al.*, 2005). Amaranths raw seed are high in anti-nutrients than can be reduced by cooking as evidenced by feeding trial of rats on cooked and uncooked amaranths seeds. These anti-nutrients can also be reduced by use of traditional processing methods such as fermentation (Mitra *et al.*, 1993; Eggum *et al.*, 2005).

Amaranths is an in expensive native crop that can be cultivated by indigenous people in rural areas as it is easily harvested and produces a lot of fruits (thus seeds) which are used as grains. It is highly tolerant to arid environment as grain hence a prospective grain. Amaranth grains grow rapidly and thus large seed head can weigh up to 1kg and contains many seeds. Amaranth species are reported to have 30% higher protein value than other cereals like millet, wheat flour, oats and rye.

Amaranth grain can be toasted much like popcorns, and ground into flour to be used in making porridge or enrich other cereals, it can be fermented and used to make porridge or can be used in form of flour to enrich cereals like maize, wheat, millet in order to improve on other nutritional value(Singhal and Kulkami, 1988).

Amaranth grain is very palatable, easy to cook and easy to digest. It is called a pseudo grain because of the flavor and cooking similarity to cereal grains. It is a dicot plant seed containing complete protein from plant source. It is also a good source of dietary fibre and dietary mineral like iron, magnesium, phosphorus, copper and manganese.

The crude protein of selected light seeded grain amaranth has been reported to range between 12.5 to 17.6 grams, (Kauffmna and Weber, 1990). Amaranth grain is reported to have a high level of lysine a nutritionally critical amino acid ranging from 0.73 to 0.84 % of the total protein content (Kauffman and Weber, 1990). The limiting amino acids include leucine and threonine, threonine being more limiting than leucine (Kauffman and Weber, 1990).

To compliment amaranth protein it has been combined with wheat, sorghum, and maize. Ordinary maize supplemented with as little as 12.7 % by weight of toasted amaranth flour provide a nutritionally superior source of protein that can satisfy a good nutrition of a protein requirement of young children and approximately 70% of diet energy. A combination of rice and amaranth in the ration of 1:1 has been reported to approach WHO protein specification for children under five years (Kauffman and Weber, 1990; Singhal and Kurkami, 1985). Combining amaranth with finger millet might also produce similar effects. The starch component of amaranth is distinctive. The starch granules are polygonal, measures 1-3mm in diameter and have

a swelling power. There is a distinctive gel characteristic of starch (Kauffman and Weber, 1990). Amaranth grain contains 6-10% oil which is found in the germ. It is predominantly unsaturated oil (16%) and is high in linoleic acid which is a vital nutrient in human bodies, (Kauffman and Weber, 1990).

Due to the fact that grain amaranth has high protein, as well as high fat content there is a potential to use it as energy food. Milling and toasting of amaranth product improves digestion as well as absorption once consumed by human being (Morales et al, 1988). The balance of carbohydrates, fats and protein allow amaranth the opportunity to achieve a balanced nutrient uptake with lower amount of consumption than with other cereals. The digestibility and protein efficacy ratio are improved if the grain is heat processed (Kauffman and Weber, 1990). Heat processing removes pectin's thus improving the protein efficiency ration of amaranth flour (Singhal and Kurkami, 1985). Other methods used in processing amaranth grains to improves on it quality includes popping, flour milled from toasted grains, fermenting the grains and milling to flour, heat rolling to flakes, extrusion and wet cooking gruel (Kauffman and Weber, 1990). It can also be processed in combination with other grains to produce cold and hot breakfast cereals, pancakes, bread, cakes and pastes, thus grain amaranth has a potential of improving the nutritional status of children as well as adults (Kauffman and Weber, 1990).

2.4.3 Pumpkin

Pumpkin is a gourd – like squash of the genus Cucurbita and family Cucurbitaceace. It is typically orange or yellow with a thick shell on the outside and pulp on the inside. They are like squash fruit ranging from less than 0.45kg to over 453.59kg. The main nutrients are lutein, alpha and beta- carotene. It is a warm weather crop that requires soil temperature of 15.5 ^oC and soil that holds water well but it is however a hardy crop. After maturity pumpkin can be boiled, baked, steamed or roasted to be used as food. It is also used to make soup and purees. It can also be mashed with potatoes or incorporated into soups.

Pumpkin is a good source of vitamin A –carotenoids (alpha and beta carotenes), vitamin C , K, E magnesium, potassium and iron. Beta carotene found in pumpkin is a powerful anti-oxidant as well as inflammatory agent. It helps build up cholesterol on the arterial walls thus reducing chances of stroke. Alpha carotene present in pumpkin assist in slowing process of aging and also prevent cataract formation. Pumpkin also reduces macular degeneration, a serious eye problem that result in blindness. Pumpkin has 0.2g fibre which is believed to be good for bowel health of an individual. It has 197.2 mg potassium which is associated with lowering risk of hypertension. Pumpkin also has zinc believed to boost immunity and improving bone density

(BOSTID, 1996; http:/lifestyle. Iloveindia.com/lounge- benefit of pumpkin and Sehmi 1993). The nutrient composition is as shown in Table 2.1.

Name of food stuff	Energy (KJ)	(kcal)	Moisture (ml)	Protein (g)	Fat (g)	CHO (g)	Fibre (g)	Ash (g)	Ca (g)	Phosphoru s (mg)	Phytin Phosph orus(mg)	Fe (mg)	Mg (mg)	Na (mg)	K (mg)
Finger Millet	1,237	129	11	6.37	1.8	66.9	2.9	3.2	190.5	209.9	39.6	20.5	228.2	5.80	60.9
Amaranth Seed	1340	319	10.0	14.7	1.9	600.7	9.6	3.1	510	397	11.0	0.07	0.21	0.5	1
Pumpkin (raw without peel)	130	31	88.7	1.8	0.4	6.6	0.46	1.02	13.8	46.4	1.02	33.5	1200	0.05	0.02
CH	O- carboh	ydrates													

Table 2.1 Proximate and mineral content of finger millet, Amaranth and Pumpkin

Source (Sehmi 1993, BOSTID 1996)
2.5 Fermentation in Increasing Nutrient Availability

Cereals are the most common staple foods for most community in rural and urban Kenya. Maize, millet and sorghum are the main cereals consumed as whole grain, roasted, boiled or milled as flour (Oniango *et al.*, 2002). The cereals are cooked as porridge to feed children. Traditional house hold methods of food preparation such as fermentation and malting are also used. Other thriving traditional household technology is the processing of cereals into fermented flour (Oniango *et al.*, 2002; Food chain, 1995; Sharma and Kapoor 1994). The naturally occurring bacteria make food acidic; this improves the taste and also has the advantage that diarrhea causing organisms cannot grow easily. Fermentation also imparts the desired flavor to flour while also increasing its shelf life due to the increased lactic acid content. The simple sugars produced in the process are easily absorbed into the body providing the nutrient requirement of the first developing child (Food chain, 1995). The fermentation process also breaks down some of the fibres in the food and increase iron absorption (Oniango, *et al.*, 2002).

Fermented foods contribute to about one-third of the diet worldwide. Cereals are particularly important substrates for fermentation and fermented cereal product arc consumed in all parts of the world including the Indian subcontinent, in Asia, and in Africa. Fermentation causes changes in food quality indices including texture, flavor, appearance, nutrition and safety. The benefits of fermentation may include improvement in palatability and acceptability by developing improved flavours and textures; preservation through formation of acidulants, alcohol, and antibacterial compounds; enrichment of nutritive content by microbial synthesis of essential nutrients and improving digestibility of protein and carbohydrates; removal of antinutrients, natural toxicants and mycotoxins; and decreased cooking time. The content and quality of cereal proteins may be improved by fermentation. Natural fermentation of cereals increases their relative nutritive value and available lysine. Bacterial fermentations involving proteolytic activity are expected to increase the biological availability of essential amino acids. Starch and fiber tends to decrease during fermentation of cereals. Although it would not be expected that fermentation would alter the mineral content of the product, the hydrolysis of chelating agents such as phytic acid during fermentation, improves the bioavailability of minerals (Hautrast *et al.*, 1989; Food chain, 1995; Oniango *et al.*, 2002; Hemalatha *et al.*, 2007). Changes in the vitamin content of cereals with fermentation vary according to the fermentation process, and the raw material used in the fermentation. B-group vitamins generally show an increase during fermentation. Another benefit of fermentation is that normally the product may not require cooking or the heating time required for preparation is greatly reduced.

2.6 Standards for Complementary Foods

Cereals suitable for human consumption are locally available and are suitable ingredients for production of formulated supplementary foods for older infants and young children. They should be processed in such a way as to reduce fibre content (when necessary) and to eliminate tannins or other phenolic materials which may lower protein digestibility and mineral availability.

Beside carbohydrates cereals contain a significant quantity of protein (8-12 %) but they are limiting in lysine (KEBS/CAC, 1991). The cereals should be treated to obtain wholesomeness and clean raw material of good quality. They should be cleaned to eliminate dirt, damaged grains, foreign grains noxious seeds, insects and insect excreta and any other adhering material. During milling care must be taken to minimize loss of nutritional value and avoid undesirable changes in technological properties of the ingredients. This applies to both commercially processed as well as home made prepared complementary foods.

During processing of cereal based products, the aim should be to obtain an energy density not less than 3.3 KJ/g (0.8Kcal/g), protein with a chemical index of at least 80% of that of the reference protein casein or the Protein Efficiency Ratio (PER) of the protein in the mixture shall be equal to at least 70% of that of the reference

protein casein. Lipid content not exceeding 1.1g/100 KJ (4.5 g/100 kcal) and vitamin A of 14-43 μ g/100KJ or 60-180 μ g/Kcal. These guideline are as per Kenya Bureau of Standard and serve as guideline during formulation of the feeds.

All ingredients, including optional ingredients, should be clean, safe, and of good quality. All processing and drying should be carried out in a manner that minimizes loss of nutritive value, particularly protein quality. The moisture content of the products should be governed by good manufacturing practice for the individual product categories and should be at such a level that there is a minimum loss of nutritive value and at which microorganisms cannot multiply.

The processed cereal-based foods should have a texture appropriate for the spoon feeding of infants or young children of the age for which the product is intended. The product should comply with any microbiological criteria established in accordance with the Principles for the Establishment and application of microbiological Criteria for Food (KEBS/CAC, 1997).

The declaration of nutrition information should contain the energy value, expressed in kilocalories (Kcal) and kilojoules (KJ), and the amount of protein, carbohydrate and fat expressed in grams (g) per 100 g or 100 ml of the food as sold, and where appropriate, as per specified quantity of the food as suggested for consumption; and the average amount of each vitamin and mineral per 100g. Also directions for preparation and use of the food, and its storage and keeping quality before and after the package has been opened, should be indicated (KEBS/CAC, 1997).

2.7 Dietary intake

Dietary intake estimates are obtained primarily as a proxy for more fundamental biological variables such as tissue concentration or blood concentration of the nutrients. Among methods used to estimate dietary intake are food records, dietary recall method, food frequency, food account among others.

2.8 Biochemical Assessment of Nutrient Intake

Biochemical methods are used to assess nutrient level in the body. Blood, urine, saliva, breast milk are among the body tissues used for biochemical assessment. Level of nutrient in these body fluids or in the cell can be used to measure nutrient levels in the body. The cell mass is an indicator of nutrition status of body mass protein. Proxy indicator of nutrient are protein, or nitrogen content in cells or serum level proteins, ascorbic acid , iron, zinc, retinol (vitamin A) among others.

CHAPTER THREE

STUDY SETTING AND STUDY METHODOLOGY

3.0 Study Setting; Thika District Profile

The study was carried out in Thika district hospital located in Juja constuency, Thika municipality division, Thika district. Thika district is one of the eleven districts that form Central Province. It was curved out of the larger Kiambu and Muranga Districts in 1995. It lies between latitude 3° 35" and 1° 45" south of equator and longitudes 36° 35" and 37° 25" East. It borders Nairobi city to the south, Muranga south district to the North, Gatundu District to the west and Machakos District to the east. The District has 1479.1 Km sq. It is divided into five administrative divisions, 11 location, 38 sub location. Most of the study children's came from Municipality and Ruiru divisions with a few from Gatanga and Kakuzi division.

The district is divided into two climatic regions, with the area which borders Kiambu, some parts of Muranga and Nairobi having adequate rainfall while the area bordering Machakos and some parts of Muranga receiving low rainfall, thus its mostly dry. There are two rainy seasons with long rains spanning from March to May and short rains from October to November. Most people in the district are mainly small scale farmers growing coffee and tea. Thika district harbor Thika town which is one of the industrialized towns in Kenya with several processing industries. There are few civil servants and business people residing in town, casual labourers in the urban areas constitute to a big percentage of the population because of the many industries/ factories in the town. Thika and Ruiru municipalities have various agricultural production industries where most of the population is employed either as permanent employee or casuals.

Thika district has a population of 544,166 with 3.24 % (14,715) being children under one year and 13.80 %(62,675) being children under 5 years and 25.9% (117,629) women of child bearing age. This is as shown in Table 3.1

Description	District proportions (%)	Eligible population 2009 - 2010
Total catchments population	100%	454,166
Children under 1 year (12 months)	3.24%	14,715
Children under 5 years (60 months)	13.80%	62,675
Women of child bearing age (15 -	25.90%	117,269
49 Years)		

Table 3.1: Demographic profile of Thika District

Source: MOH Thika district annual operating plan (AOP 5) 2009

Distribution of the top five diseases in the district is as shown in Table 3.2. Respiratory tract infection is the leading cause of morbidity in the district (42.1%) followed by malaria (30.4%) and intestinal worms (6.0%). Malaria and intestinal worms mostly affect young children and pregnant mothers thus during the study period children were dewormed and mother trained on malaria prevention strategy and issued with long lasting insectside treated nets (LLITNS). Malaria and intestinal worms infestation usually affect the nutritional status of children. Children suffering from malaria or those infested by intestinal worms are likely to suffer from iron deficiency hence the need to control mosquito and intestinal warm infestation

		0/ / 1 /
Disease	Number of cases	% contribution
Diseases of Respiratory	191,074	45.8
Malaria	127,072	30.4
Diseases of skin	43,974	10.5
Diarrhoea	30,316	7.3
Intestinal warms	25,115	6.0
TOTALS	417,551	100

Table 3.2 Top five diseases in Thika district

Top five causes of out patient diseases year 2009

Source: MOH Thika district annual operating plan (AOP 5) 2009

3.1 METHODOLOGY

3.1.1 Study Design

The study consisted of a longitudinal intervention with analytical component. The children with moderate weight for age z scores at the maternal and child health clinic (MCH) in Thika district hospital were sampled randomly and assigned to two equal groups. One group was supplemented with fermented porridge flour (FPF) while the control group was supplemented with non fermented porridge flour (NFPF). The children were evaluated for the effect of fermented porridge on their nutritional status, vitamin A, iron and zinc status in comparative with the unfermented product.

3.1.2 Study population and sampling frame

The study population consisted of moderately malnourished children aged 7- 24 months screened at Thika district hospital MCH The sampling frame consisted of all moderately malnourished children with a weight for age Z-score of between -2 to -3 with no disease condition and had not been enrolled to any food support programme.



Figure 3.1 Study Design

3.1.3 Sample size determination

An increase of 1g/dl was considered desirable to allow for detection of difference of mean haemoglobin concentration of the two groups. The standard deviation of the haemoglobin concentration in each group was set to be 1.1g/dl according to INACG

(1984). The power of test was set at 95% with a significant level of 0.5. The following formula was used (Kirkwood, 1988);

 $n = (u + v)^{2} (sd_{1}^{2} + sd_{2}^{2})$

 u_1 - u_2

Where

n = sample size,

 u_1 - u_2 = difference between the group haemoglobin concentration means,

 $sd_1 = standard deviation of group one$

 $sd_2 = standard deviation of group two,$

u =one sided percentage of the normal distribution corresponding to 100% - power of test (95%) which is equal to 1.64

v = percentage of normal distribution corresponding to two sided significance level (0.5) the v = 1.96)

Applying the formula

$$n = (1.64+1.96)^2 (1.1^2+1.1^2)$$
$$(1.0)^2$$

n = 31.36

Approximately 31 children were included in the study with each product. An attrition of 10% was allowed giving a sample size of 34 children.

3.1.4 Sampling procedure

Thika district hospital was purposively selected because it is as is one of the districts with high prevalence of malnutrition. Children with no complication and with a Z score of between -3 to -2 SD (weight for age) were screened from Children attending growth monitoring at the maternal child health (MCH) clinic. A total sample of 62

children was exhaustively selected and an attrition of 10 % was allowed which made a total sample size of 68 children. The children selected were randomly assigned to two groups fermented porridge flour group (FPF) and non- fermented porridge flour group,(NFPF).

3.1.5 Inclusion and exclusion criteria

The children who were eligible for the study were those aged between 7 to 24 months with weight for age between -3 to -2 Z score. Only health children with these characteristic were included. Sick children were not included in the study. The cut off point of seven month was chosen as all children at seven month are supposed to have been started on complementary foods. The age 7 - 24 month is the most vulnerable group during complementary feeding.

3.1.6 Screening and screening procedure

The Maternal Child and Health (MCH) clinic at Thika district hospital was used for screening as this is where children attend growth monitoring and promotion (GMP). The District Medical Officer of Health (DMOH), Director of Medical Services Officer (DMSO), MCH in-charge and District Nutrition Officer (DNO) were contacted one week before conducting the anthropometric measurement. Children attending GMP daily were screened until the required sample size was reached. Three hospital nutritionist, a nurse and a clinical officer were present during the screening period. The clinical officer and the nurse chosen were those trained on management of childhood illness to identify disease condition during screening and offer treatment to the sick children. The principal investigator with the assistant of the nutritionist took weight, height and determined age of the children attending GMP. Data on anthropometric measurement was analyzed using ENA Nutrisurvey soft ware after anthropometric measurement during screening period and those children met the inclusion criteria were recruited. A total of 68 children were selected for the study.





3.1.7 Sensitization of the parent for consent and recruitment of study children The mothers/guardians were informed about the study and its benefit to the nutrition of their children. The mothers/guardians of the children whose weight for age corresponded with the reference weight for age were advised and encouraged to continue taking good care of their children in all aspects related to health and nutrition. The mothers/guardians whose children weight for age indicated underweight (-3 to -2 Z score) and therefore qualified for the study were counseled and explained about the study procedure including blood sample collection at the start and end of the study, informed about the confidentiality and their consent sought for their children to be included in the study. One to one talk was preferred in order to give a clear explanation to the parent about the study and also to enhance a sense of commitment for the child to be enrolled in the study. Those parents/ guardian who agreed to have their children included in the study signed an informed consent form represented in Appendix 3. Time was given to those who needed to consult their spouse for their children to be included in the study and ask consent for blood sample collection before the children were recruited for the study. The parents whose children weight-for-age was below -3Z score were counseled and referred for further investigation by the pediatrician and therapeutic care.

3.1.8 Allocation of study group

A sample of sixty eight children was allocated randomly to two groups; one group was given fermented porridge (FPF group) and the other non fermented porridge (NFPF group) as shown in Figure 3.1. Then 100 gm of the porridge flour was given to each child as a daily ration with follow -up done for a period of six weeks.

3.2 Formulation of the Flours

Amaranth seed, finger millet seed and pumpkin (butternut) were purchased from local market in Thika and transported to the pilot plant Department of Food Science, Nutrition and Technology for processing. Recommended daily allowance (RDA) (FAO, 2005) as shown in Table 3.3 was used in the formulation.

Table 3.3: Recommended dietary Allowance (RDA) of the children aged 1-3yrs

Nutrient	Energy	Protein	Iron	Vitamin A	Zinc	
Quantity	1022kcal	14gm	6mg	400ug	8.4mg	

Source: Disvestro, (2004); FAO (2005)

The formulation aimed to achieve minimum 60 % RDA of protein, iron and zinc with a maximization of vitamin A level as per recommended daily allowance shown on Table 3.3, using nutrient compositions of the amaranth, finger millet and pumpkin (Sehmi, 1993).

3.2.1 Non fermented Amaranths-finger millet- pumpkin porridge flour processing

Amaranths and finger millet were washed separately, drained, dried and milled using Norris and hammer mill. The pumpkin was also washed and cut into halves. The seeds were removed, and the cut pieces peeled and sliced to 5mm cubes. The sliced cubes were steam blanched at 95° C for 15 minutes and drip drained. They were comminuted in a blender, and then mixed with the amaranth and finger millet flour. The mixture was dried at 60° C in the oven to an approximate moisture content of 10-12 %. Samples for proximate and microbial analysis were drawn from the flour mixtures. Samples were also drawn for analysis of vitamin A, iron and zinc. The formulation is summarized in Figure 3.3.

3.2.2 Fermented amaranths-finger millet- pumpkin porridge flour processing Finger millet and amaranth were washed separately, drained, dried and milled using Norris and hammer mill. They were then mixed after milling. The pumpkin was washed, peeled, seed removed, sliced to 5mm cubes and the cubes steam blanched at 95° C for 15 minutes. They were macerated in a blender to homogeneity and the

macerate mixed with finger millet and amaranth flours. The mash was then mixed with 30% (w/w) water to make slurry that was allowed to fermented spontaneously for 48 hours to a pH of 4.0.

The fermented slurry was dried in an oven at 60 ⁰ C to an approximate moisture content of 10-12%. Samples for microbial and proximate composition, iron, vitamin A and zinc determination were drawn randomly after drying. The formulation is summarized in Figure 3.4.

3.3 Chemical and Microbial analysis

A sample of 50 gm and 100 gm were drawn randomly from the fermented and nonfermented amaranth-finger millet-pumpkin flour and analyzed for proximate composition, beta-carotene, zinc and iron and microbial quality.

3.3.1 Determination of proximate composition of the porridge flours

The fermented and non-fermented amaranth-finger millet-pumpkin composite porridge flour was analyzed for total moisture, crude fibre, crude protein, total ash and soluble carbohydrates according to AOAC methods (AOAC, 1999).

3.3.1.1 Determination of moisture

Moisture contents of the samples were determined by standard analytical AOAC methods (AOAC, 1999). About 5g of each sample were weighed accurately in triplicates into aluminum dishes. Each dish and contents were put in an air oven maintained at 105°C and dried for four hours. The dishes were cooled in desiccators to room temperature then weighed and results recorded. Loss of weight due to drying was converted to percent moisture content.



NFPF flour

Figure 3.3 Processing of Non Fermented amaranth-finger millet-pumpkin porridge flour





Figure 3.4 Processing of Fermented amaranth-finger millet-pumpkin porridge flour

3.3.1.2 Determination of crude protein

Crude protein was determined as total nitrogen by the Kjeldahl method (AOAC 1999). Triplicate samples of about 0.5g were weighed accurately in nitrogen free filter paper. Each sample was folded carefully and placed in a 100ml Kjeldahl flasks followed by 5.5g Kjeldahl catalyst tablet (CuSO₄: k_2SO_4 =1:10). Then 20ml concentrated Sulphuric acid were added. A blank analysis was carried out without the sample at the same time. The flasks were heated on the kjeldahl heating assembly until all frothing stopped and a clear blue obtained. After cooling, distilled water was added to have the liquid fill ³/₄ of the flask. Some drops of phenolphthalein indicator were added. Four hundred (400) ml conical flasks containing 50ml of 0.1N HCl solution and some drops of methyl orange indicator were placed under the outlet of Kjeltic system 1002 distilling unit. The diluted digest was mixed with 40% NaOH to make sufficient alkaline then steam distilled up to a volume of 200ml. The ammonia in the distillate was determined by back titrating with 0.1N HCl. Percent crude proteins were calculated as

Nitrogen $\% = (v_1 - v_2) \times N \times f \times 0.014 \times 100/s$, where,

 V_1 = tire for the sample (ml) V_2 = tire for the blank (ml) s= weight of the sample taken in grams

N = normality of the standard HCl solution= 0.1

Protein % = nitrogen % x protein factor (6.25).

3.3.1.3 Determination of Crude Lipids

Crude lipid was determined by standard AOAC methods (AOAC 1999). About 5g samples were weighed accurately in triplicate into cellulose extraction thimbles and extracted with analytical grade petroleum ether of boiling point 55°C in soxhlet extraction unit for 16 hours. The ether extract was transferred to 300ml flat bottomed

flasks that had been previously washed dried in an oven at 105°c cooled in a desiccator and weighed. The excess petroleum ether was evaporated and residual extract dried in an oven at 80°C to constant weight. The residue was calculated as percentage crude lipids.

3.3.1.4 Determination of crude fiber

Crude fiber determination was carried out according to standard AOAC method (AOAC 1999). Triplicate samples of about 2 g were weighed accurately into a graduated 500ml beaker. Small amount of boiling distilled water together with 25ml of 2.04 N sulphuric acid solutions was added. The content was adjusted to 200ml with boiling distilled water and maintained at this volume while boiling gently on a hot plate for 30 minutes. The content was then filtered using a buchner funnel slightly packed with glass wool and then the residual was washed three times with boiling distilled water.

The residues together with glass wool were transferred quantitatively to the beaker. Small amount of water was added as well as 1.73N KOH solution. The volume was made to 200ml with distilled water and this volume was maintained as the contents boiled on a hot plate for 30 min. Then the contents were filtered using glass wool and the residue washed as above. The residue was again washed three times with small amount of ethanol. The residue was transferred quantitatively to a porcelain dish dried in a air oven for 2 hours, weighed and then the dish and the content ignited at 550°C to constant weight.

Then after cooling in a dessicator the weight of each was taken. The weight of the fiber content was calculated as follows;

Fiber $\% = (w_1 - w_2)/\text{sample weight } x 100$

3.3.1.5 Determination of total ash

Total ash was determined by standard AOAC method (AOAC 1999). About 2g of samples were weighed accurately in triplicate into porcelain ashing dishes previously cleaned and dried in an air at 105°C cooled in a dessicator and weighed. The dishes and the content were then held in a muffle furnace at 550°C overnight. They were

then removed cooled to room temperature and weighed. Percent ash was then calculated as follows

Ash %= weight of ash remained / weight of sample x100

3.3.1.6 Determination of soluble carbohydrates

Total carbohydrates were determined by difference; 100 – (crude fibre + crude protein + crude fat + total ash + moisture content)

3.3.2 Determination of total Energy

Energy was calculated using Atwater factors [energy content= (g carbohydrate x 4) + (g fat x 9) + (g protein x 4)]

3.3.3 Determination of pH

Measurement of pH of the samples was done in a homogenate prepared with 10% (w/v) flour in distilled water using a glass electrodes pH meter (Hanna pH 210) pH metre specification.

3.3.4 Determination of beta-carotene

Beta-carotene contents of the composite porridge flours were determined using AOAC methods (AOAC, 1999).

3.3.5 Determination of iron and zinc

The content of zinc and iron were analyzed in the samples of the formulated flours using Atomic absorption Spectrometre (AAS) (Unicam 939/959 PYE-Unicam, Cambridge, UK) equipped with an air acetylene flame and hollow cathode lamps, and using lamp specific for the elements. The device was operated under standard condition using wavelength and slit widths specified for each element (AOAC, 1999). For analysis 1 g of the finely ground sample was accurately weighed and incinerated to constant weight. The ash was extracted with 10ml of HCL:water (1:1) and the extract was quantitatively transferred to 50 ml volumetric flask and made up to the mark. Appropriate dilutions were made and the element analyzed against a standard.

3.3.6 Microbial analysis

Samples from fermented and non fermented porridge flour were drawn randomly and analyzed for total plate count, coli form, molds and yeast. Microbial analysis was done according to compendium of food microbiological methods. This analysis was done in the two products. About 25 g of each sample were accurately weighed and was placed in 225ml sterile saline solutions and shaken to prepare 10⁻¹ dilution. Then decimal serial dilutions were prepared in the saline solution. Aliquots 0.1ml was used to inoculate on to the surface agar media a spread plate technique. Additionally aliquots (1.0ml) were used in an agar pour plate procedure for total viable counts of bacteria, coliforms, yeast and molds. Colonies on the agar plate were counted (cfu/g) and a proportional sub sampling procedure was used to select colonies of bacteria for identification.

Medias were prepared according to consumer specifications, autoclaved at 121°c for 15min to sterilize and then held at 50 °C in water bath to prevent solidification before pour plating. This was done for all medias except for violet red bile agar (VRBA), which doesn't require autoclaving. Relatively saline water was prepared using 0.85% NaCl. Then 225ml of saline solution was put in a triplicate in conical flask and 9ml saline solution were put in test tube all totaling to 18 diluents. After autoclaving the saline solution was cooled to room temperature and aseptically stored before use.

A sample of each porridge flours was aseptically measured and made into a solution in 225ml saline solution. This was labeled as 10⁻¹ diluents. One- milliliter was taken from 10-¹ diluents using a sterile pipette and put in 9ml and labelled 10⁻². This was repeated 5 times for each sample in every experiment up to 10⁻⁶. Then 1ml of appropriate diluents was directly inoculated in triplicate by pour plating on the media. Plate count agar (PCA) agar was incubated anaerobically at 37 ⁰C for 48 hours to enumerate total viable count. Violet red bile agar was incubated anaerobically at 37[°]c for 48 hours to enumerate enterobacteria (coliforms). Malt extract agar for enumeration of yeast and mold was incubated for 30 [°]C for five days but close monitoring was done daily on each media for detection of microorganisms. The procedure was repeated for every experiment.

3.3.7 Sensory evaluation of the porridges

The composite flour was prepared into porridge. One kilogram of each flour was weighed using a kitchen scale and slurred in to one litre of water separately then poured into two litres of boiling water. Continuous stirring was done to prevent lump formation and sticking of the flour to the bottom of the pot as the porridge cooked. The porridge was then cooled into 40^oc and poured into coded cups. In each cup 100 ml of the porridge was poured and together with evaluation sheets were presented to the panelist.

Sensory evaluation was carried out using acceptance/consumer/ preference test. The quality attributes that were investigated were colour, taste, flavour, and texture. A seven point hedonic rating scale was used with 7= like very much and 1 = dislike very much.

The mother were explained on how to rate the attribute as per the seven point hedonic scale. Each mother was given two samples on coded cup A and B to taste and rate each on the provided sheet as shown in the sensory evaluation sheet provided in sensory evaluation sheet in appendix 1 number 3.

3.4 Baseline Data Collection

3.4.1 Social demographic characteristic of household members

Data on social demographic characteristic of the household members was collected using the field questionnaire section one attached in appendix one section 4. Among the data's collected included education level, marital status, occupation, age and gender.

3.4.2 Anthropometric measurement (Weight and height measurements)

Anthropometric measurement of children attending MCH clinic during screening days was taken. It included weight, height, and recording of age and sex. Principal investigator took the measurements with assistance of the nutritionist. Child name was also recorded.

The weight of the children was taken using Salter scale. Two readings were taken for every child, to the nearest 0.1 kg and the average taken. The length of the children was taken using a length board. The board was laid horizontally on a flat surface, and the child laid on it with the head positioned against the fixed head board and eyes looking vertically up. The knees were extended and feet flexed at right angle to the lower leg. The up-right sliding foot piece was moved to obtain the firm contact of the heels. Two readings were taken for every child to the nearest 0.1cm and the average computed.

3.4.3 Age determination

The mothers of the study children were asked the date of birth of their children. This was verified using the child health clinic card. Date of doing the anthropometric measurement was also recorded. These age were entered into MS excel and the age calculated by difference between the two dates. Also date on end of feeding was recorded in order to determine age at end of feeding.

3.4.4 Blood sample collection

Blood samples were drawn from the children at the start of the study and after feeding the children for six weeks. Blood drawing, blood sample preparation and blood analysis were carried out with the help of a qualified medical laboratory technologist from the hospital. Blood samples were collected on every child, on first morning after arrival at the nutrition centre. Ten-mililitres (10ml) of blood was

obtained from each child using an antecubital puncture. Then I ml of the blood sample was used to determine the hemoglobin concentration and 9 ml used for determination of serum protein, serum zinc and serum retinol

Sample collection and preparation was done in as much controlled environment as possible because blood serum result may vary due to changes in intravascular pressure at the time of blood withdrawal (Brown *et al.*, 2005). The subject remained sealed to the blood drawing process. Blood samples which could not be analyzed immediately were stored under refrigeration to prevent release of nutrient from platelet and blood cell leading to slightly increased concentration of the nutrients.

3.4.5 Determination of total serum protein, serum zinc and serum retinol

Serum was separated from whole blood. This was by centrifugation to pack up all the blood cells leaving a clear serum. The serum was separated and used for serum protein, serum zinc and serum retinol analysis.

3.4.5.1 Serum protein determination

Serum protein was analyzed by use of Biuret reaction. The principle of the method is that serum protein forms a coloured complex in the presence of copper salt in alkaline solution. The reagent composition was potassium iodide 6mmol/l, potassium sodium tartrate 21mmol/l, copper sulphate 6mmol/l and sodium hydroxide 58mmol/l. A standard was analyzed with albumin solution in distilled water, together with a blank.

During analysis 3 μ l of distilled water, 3 μ l standard and 3 μ l of sample were each mixed with thoroughly 200 μ l as shown in Table 3.4 and incubated for 225 seconds after which their absorbance (A) value were read at a wavelength of 546nm at an optical path of 1cm at 25^oC.

Table 3.4: Serum protein analysis

	Blank	Standard	sample
Reagent	200 µl	200 µl	200 µl
Distilled water	3 μl	-	-
Standard	-	3 μΙ	-
Sample(serum)	-	-	3 μl

3.4.5.2 Serum zinc determination

The protocol derived from Dawson, (1968) were adopted for zinc analysis. A ratio of 1 unit of serum to 9 unit of distilled water was vortex mixed. This approach is believed to be free from interference effects of protein. All samples were analyzed within three hours of reconstitution. The determination of serum zinc was done using an atomic absorption spectrophotometer (AAS). The AAS was calibrated using serum zinc standard before carrying out the analysis. The measurement was carried out at 231.09nm, using a gas flow rate of 2.4l/ minute of analytical grade acetylene and 8 l/minute of filtered air background correction mode.

3.4.5.3 Serum retinol determination

Serum retinol was measured using a high performance liquid chromatography (HPLC). One hundred (100) microlitres of serum, 15 microlitres of internal standard solution (4 microgram/ml of) retinyl acetate and 100 microlitres of methanol were transferred to a conical flask tube. The content in the tube were mixed using a vortex mixture. Then 200 microlitre of extraction solvent (diethyl ether: dichloromethane: isopropanol; 80: 19: 1) by volume were added to the tube. The tubes were then capped with a polythene cap. The tubes were volexed intermittently for a total period of 60 seconds and centrifuged. Then 100 microlitres of the supernatent was injected into the machine. Elution was done with the same solvent used in extraction. Then the HPLC condition displayed was recorded.

3.5 Food consumption patterns and Dietary Intake

This was obtained using food frequency table. Information on types of food consumed and their frequencies were obtained. Twenty four (24) hour dietary recall was also done to determine the dietary intake of the children.

3.6 Deworming and Malaria Prevention

After collection of the baseline data the children were dewormed with levamasole syrup (5ml per child). The prescription was done by a clinical officer trained in integrated management of childhood illnesses (IMCI). At the age of 7- 21 children creep and eat a lot of dirt hence need for deworming.

Thika district is a malaria zone area especially at the municipality division, with malaria being the second cause of morbidity as shown in Table 3.4. To prevent malaria mothers/ guardian were trained on malaria prevention methods which included use of insect side treated net (ITN), drainage of stagnant water. This was done, by a qualified public health officer in charge of malaria prevention in the district. The mother/ guardian were also provided with each a long lasting insectside treated net for their children as a malaria prevention strategy during the study period. Follow up was done to ensure that the mothers used the net for the children by public health technicians.

3.7 Feeding trials

After collection of baseline information the children were randomly assigned in two groups. Each mother was given a daily ration for the child on weekly basis of the fermented and non porridge flour for a period of six week. The children were fed on the porridge for a period of six weeks. The mothers/guardian were trained on how to prepare the porridge and record daily consumption. A quantity of 100gm was recommended for daily preparation of the porridge hence of 700gm was given on weekly basis. Daily record on porridge consumption was done by the mothers and weight of the child taken on weekly basis by the principal investigator with assistant of the research assistant as explained in section 3.5.2. Mother/ guardian came to the nutrition centre weekly for supply of the flour and weight determination of the children. After a period of six week the nutrition status (weight for age), protein, vitamin A, zinc and iron were determined using procedure explained in section 3.5.1 to 3.5.5. Since height was also taken height for age and weight for height Z score were also calculated using ENA Nutrisurvey software (NHCS,WHO 2007). Follow up was done at household level to ensure the porridge flours were consumed by the children with no sharing.

3.8 Data collection and management

Weight change during feeding was collected for a period of six weeks on weekly basis. Blood analysis for serum protein, serum retinol, serum zinc and hemoglobin levels was done at the start of the study and after six weeks of feeding. It was collected and recorded on laboratory request forms.

Dietary intake was determined using food frequency and 24 hour dietary recalls. Anthropometric measurement data were recorded on the table provided in the questionnaire.

Nutritional status was determined before and after feeding using ENA Nutrisurvey software and recorded.

3.9 Data Analysis

Data on social demographic characteristic of the household were subjected to independent sample t-test, and chi-square test to examine the difference between study group and association between groups' characteristics.

Morbidity indices between and within groups were compared using independent sample t-test and paired sample t-test respectively. Anthropometric data at baseline and after six weeks of feeding were analyzed for within and between groups using paired sample

t-test and independent t-test respectively. Effect of feeding on anthropometric and biochemical data were measured as difference between group changes from baseline to 6 weeks in FPF group compared with NFPF group.

Complete biochemical data were available for 55 children (28-FPF, 27-NFPF). The reason were mainly refusal of blood sample withdraw (3-FPF, 3-NFPF) from children by their parents at the end of the study and drop out before six weeks of feeding were over (3-FPF and 4-NFPF). Biochemical data at baseline and after six weeks of feeding were analyzed for within- group and between group responses to feeding using pared samples t test and independent sample t-test.

3.10 Ethical clearance

Permission to conduct the study was approved by Kenyatta National Hospital Ethical and Research Committee. Research clearance was obtained from Research Coordination section, Ministry of Higher Science and Technology. The ethical clearance are as per attached appendix five.

CHAPTER FOUR RESULTS AND DISCUSSION

This chapter presents the result of proximate composition, microbial quality and sensory evaluation of fermented and non fermented porridge flour. In addition the results of the dietary intake, morbidity status of the study children and the effect of the flours on nutritional status, serum protein, serum retinol, serum zinc and haemoglobin levels are also presented and discussed.

4.1 Proximate composition of the porridge flours

Result of proximate composition, iron, zinc and beta carotene are shown in Table 4.1. The moisture contents of the FPF and NFPF were 10.1% and 8.2%. The moisture content of the FPF was significantly higher (p<0.05) than that of NFPF. The moisture content of the FPF and NFPF were within the range recommended for a long shelf life infant food by Codex Alimentarius (KEBS/CAC, 1991). The protein content of the FPF and NFPF were 9.2% and 8.8% respectively, while crude lipids content was 5.6% and 5.10%. The protein contents of the FPF and NFPF were below the recommended Codex Alimentarius 15g/100g and contributed 61.3% and 58.7% of the recommended protein content for formulation of infant food (KEBS/CAC, 1991; WHO, 1985; FAO, 2004; FAO, 2005). However these values were within the target formulation, which was to achieve 60% recommended daily intake for both flour. Fermented porridge flour (FPF) had significantly higher (p<0.05) levels of protein than the non-fermented porridge flour (NFPF). The protein content was higher in FPF than in the NFPF because of the proliferation of lactic acid bacteria which made contribution to the protein biomass. The content for ash, carbohydrates, and energy are as shown in the Table 4.1. The energy content of the FPF and NFPF were 382KCal per 100gm and 365kcal per 100gm respectively. The energy content of NFPF (365kcal per 100gm) was significantly lower (p<0.05) than that of FPF (382kcal per 100gm). Fermentation leads to increased energy density and this therefore increased the energy in the fermented flour. The energy content for

both porridge FPF and NFPF 382 kcal/100gm and 365 kcal/100gm respectively were lower than that of 400 KCal/100gm recommended by Codex Alimentarius for the formulation of supplementary food (KEBS/CAC, 1991; WHO, 1985; FAO, 2004; FAO, 2005). Addition of about 5g sugar or 5 g fat, would therefore increase the energy content to between 402 - 417.1KCal/100g and 385 - 400 KCal/100g for fermented porridge flour and that of non fermented porridge flour respectively, and this would bring the levels to within the recommended amount of 400 KCal/100grams (KEBS/CAC 1991; FAO, 2001; FAO, 2004). From the food frequency data most children consumed fat (64.5% and 66.7%)| for FPF and NFPF group respectively or sugar (58% and 56.7%) for FPF and NFPF group respectively. The sugar was mainly added to the porridge during preparation and this could easily increase the energy content of the porridge to the level recommended by Codex Alimentarius (KEBS/CAC 1991; FAO, 2001; FAO, 2004).

Carbohydrate content for FPF of 78 g per 100g was lower than 84g per 100g of NFPF. Fermentation leads to decrease in carbohydrates which are used as substrate for fermentation. During fermentation sugars are converted to organic acids; lactic and acetic acids causing a reduction in pH and reduction of carbohydrates as nutrients sources. This could explain the low levels of carbohydrates in the fermented porridge flour (Oniang'o, *et al.*, 2002).

The beta- carotene content of FPF (180 μ g) was significantly higher (P<0.05) than 146 μ g of NFPF. Pumpkin, one of the ingredients in the flour formulation had beta carotene content of 740 μ g. The level of crude lipid in both flours (5.59g/100g and 5.10g /100g) of FPF and that NFPF respectively were lower than the recommended 10 - 20g by Codex Alimentarius for formulation of supplementary foods (/KEBS/CAC, 1991), however FPF had higher levels than NFPF.

The level of crude fibre were lower than the recommended 5g/100g by Codex Alimentarius of supplementary foods, but the levels were acceptable (3g/100g) and (3.3 g/100g) of FPF and that of NFPF respectively (KEBS/CAC, 1991). Because of their low levels, therefore the fibre would therefore be expected to have little effect on utilization of protein and minerals.

	%Moisture	%Crude protein	%Crude fat	%Crude fibre	%Ash	%soluble carbohydrates	Energy in kcal/100 g	Iron (mg/100g m)	Zinc (mg/100g)	Beta carotene µg/100g	Retinol equivalen (RE/100g
NFPF	10.1	8.8	5.1	3.0	2.71	78	365	4.9	2.6	146	24.3
FPF	8.2	9.2	5.6	3.3	2.72	84	382	5.2	2.4	180	30
Key	NFPF- Non fern	nented amaran	th-finger mill	et porridge fl	our						

Table 4.1 Proximate composition, iron, zinc and beta carotene content of the fermented and non fermented finger millet amaranth

NFPF- Non fermented amaranth-finger millet porridge flour FPF – Fermented amaranth-finger millet porridge flour

flour per 100 gm

pumpkin porridge

4.2 Microbial Quality of the Flours

The results of microbial analysis of the flours are shown in Table 4.2. The pH before fermentation was 6.1 and on fermentation the pH dropped to 4.0. The drop in pH was due to conversion of sugars to organic acids, lactic and acetic acids (Oniang'o, *et al.*, 2002).

The total viable counts ranged from $6.0\log_{10} \operatorname{cfug}^{-1}$ to $9 \log_{10}\operatorname{cfug}^{+1}$ and $9.0 \log_{10} \operatorname{cfug}^{-1}$ to $12 \log_{10}\operatorname{cfug}^{-1}$ for fermented and non fermented flour respectively. The mean total viable count was 8 log ${}_{10}\operatorname{cfug}^{+1}$ and 11 log ${}_{10}\operatorname{cfug}^{-1}$ for the fermented and non fermented porridge respectively. This suggests presence of other acidophilic bacteria and which contribute to spoilage of several type of food. This can be so especially considering the methods of millet and amaranth threshing after harvest. The coliforms in the samples were < log ${}_{10}\operatorname{cfug}^{-1}$ suggesting a hygienic handling practice during processing of the flours. The total viable count in the fermented porridge flour were significantly lower (p<0.05) than those in the non fermented porridge flour and this could be due to the inhibitory role of acid and substances produced by lactic acid bacteria as fermentation take place. Such products as peroxide and diacetyl secondary reaction products and bacteriocins produced during fermentation have potential to inhibit a variety of other microorganisms (Oniang'o, et *al.*, 2002).

Type of microorganism	Fermented p flour (n=5)	orridge	Non Fermente (n=5)	P value	
	Mean ± sd	Range	Mean ± sd	Range	
Total plate count	7.7±1.2*	6-9*	10.5±1.0*	9-12*	0.001 a
Coliform	<log<sub>10*</log<sub>	<log<sub>10*</log<sub>	<log<sub>10*</log<sub>	<log<sub>10*</log<sub>	
Yeast and molds	2.6±1.2*	1-4	2.6±1.3*	1-5*	1.0
pН	4±0.2	3.8-4.2	5.8±0.2	5.7-6.1	0.00 ^a

Table 4.2 Microbial quality of the flours and pH

* $(\log_{10} cfu/g)$

cfu- colony forming unit significant different

4.3 Acceptability Score of the Porridge Samples

The results of sensory evaluation of fermented and non fermented porridge are presented in the Table 4.3. The mean rating of colour of fermented porridge was significantly higher (p<0.05) than that of non fermented porridge. The mean rating for taste, flavour and overall acceptability for fermented porridge were significantly lower (P<0.05) than for non fermented porridge. This indicated that the mothers liked the flavour and taste of the non fermented porridge more than that of fermented porridge. During fermentation sugars are converted to organic acids; lactic and acetic acids causing a reduction in pH and making the food acidic. Fermentation also impart desired flavor to fermented cereals while also increasing its shelf life due to the increased lactic acid content (Oniango *et al.*, 2002). The sour taste and the flavour produced during fermentation were not familiar to the mothers probably because they do not naturally consume fermented porridge in their diet. This indicated why mothers liked the non fermented porridge better compared to fermented porridge. The two products scored above the value of 4.0 which indicates indifference meaning that the two products were acceptable in all parameters attribute tested. This is as shown in Table 4.3.

Attributes	Porridge samples	P- value			
	FPF	NFPF			
	(mean±s.d)	(mean±s.d)			
Colour	6.08±1.12	5.68±1.31	0.001		
Consistency	5.56±1.58	6.0 ± 1.29	0.005		
Flavour	5.32±1.86	5.72±1.28	0.012		
Taste	5.36±1.75	6.12±1.30	0.000		
Overall	5.32±1.84	6.04±1.14	0.000		
acceptability					

	Table	4.3	Mean	rating	for sensory	characteristics of	the	porridge s	ample
--	-------	-----	------	--------	-------------	--------------------	-----	------------	-------

FPF fermented porridge NFPF None fermented porridge

4.4 Baseline characteristic of Household of the study children

4.4.1 Social demographic characteristics of the households of the study children The study children came from household whose majority members were females comprising of 53.4% and 46.6% male. Out of these 31.1% had no formal schooling, 2% were in pre school, 34.2% had attended primary education, 28.3% secondary education and 3.7% post secondary education as show in Figure 4.1 Only 2.7% of the house hold members were permanently employed, 5.5% temporally employed 13.2% in business, 9.6% in casual labour, 2.3% in farming and 62.1% unemployed. From the household members only 31.5% earned income as shown in Figure 4.2. There was no significant difference between social demographic characteristics of household for the children in FPF and NFPF.

The distribution of family size of the house hold from where the children came from is shown in Table 4.4. Most of the households (36.1%) had a family size of 3 followed by a family size of 4 (31.1%). However there was no significant difference between the household size of the families in the two groups of children. The mean

house hold size was 3.8 for the household of the two groups of children (FPF and NFPF). The mean household size for the family of the two groups of children was lower than that of Kenyan national population mean household size of 4.4 (CBS, 2004).



Education level




GROUP B FPF

Figure 4.2 Occupation status of household members of the study children.

Number of	G	Froup	
Household Members			
	FPF (%)	NFPF (%)	Total (%)
2	6.4	13.3	9.8
3	35.5	36.7	36.1
4	32.3	30	31.1
5	19.4	6.7	13.1
6	3.2	6.7	4.9
7	3.2	3.3	3.2
8	0	3.3	1.6
Mean ± sd	3.8±1.2	3.8±1.4	3.8±1.3
P value		0.399 ^{ns}	

4.4.2 Mother's profile

The occupation status of the mothers is shown in Table 4.5. Most mother from the two groups (64.5% and 63.3%)for FPF and NFPF respectively were housewife's. However, they was no significant difference in occupation status of mothers of the children from the two groups (p>0.05).

Table 4.6 shows the education status of the mothers of the children. Forty eight percent (48.4%) and 41.9% of mothers for children in FPF group had attained upper primary and secondary education respectively, while 53.3% and 46.7% of mothers for the children in NFPF group had attained upper primary and secondary education respectively; however there was no significant difference in level of education of mother of the two groups of children

Total (%)
10tat (70
3.3
3.3
11.5
3.3
1.6
63.9
13.1

^{ns}-not significant by t-test

The numbers of mothers who had attained primary education in the families of the two groups of children was lower as compared to the national figure of 58% (CBS, 2004). The percentage of mothers who had attained secondary and tertiary education were however higher than the national figure of 23.8% and 59% (CBS, 2004).

Table 4	4.6	Education	status	of	the	mothers
---------	-----	-----------	--------	----	-----	---------

Education	Gro	oup	
	FPF (%)	NFPF (%)	Total (%)
Primary	48.4	53.3	50.8
Secondary	41.9	46.7	44.3
Tertiary college	9.7	0	4.9
P value		0.228 ^{ns}	

^{ns} -not significant by t-test

Eighty four percent (84%) and 83% of mother for the children in FPF group and NFPF group respectively were married while 16% and 17% of mother for the children from FPF group and NFPF group respectively were singles. However there was no significant difference in marital status of mother of the two groups of children as shown in Table 4.8. These figures were higher as compared to the national figure of 54.5% of married women (CBS, 2004)

The occupation status of the mother was positively associated with stunting as well as wasting of the children. In addition the household size was also associated with stunting of the children as shown in Table 4.7. This is supported by a study that indicated education of the child caretaker, household size, positively associated with stunting of the children (CKDAP, 2006).

Statu	Status of Canadian								
	Stunting((HAZ)	Underweigh)	Underweight(WAZ)		VHZ)			
Variables	Pearson Correlation	P – value	Pearson Correlation	P - value	Pearson Correlation	P- value			
Occupation of the	0.281	0.028*	-0.320*	0.012	-0.099	0.447			
mother									
House hold size	-0.455*	0.000	-0.204	0.115	-0.036	0.783			
Education of the mother	0.082	0.529	-0.118	0.367	0.179	0.167			
Marital status of mother	0.072	0.582	-0.080	0.539	0.103	0.428			

 Table 4.7 Correlation between social demographic characteristic and nutrition

 status of children

* Correlation is significant at the 0.05 level

Marital status	Gro	oup	
	FPF (%)	NFPF (%)	Total (%)
Single	16	17	16.4
Married	84	83	83.6
P value		0.956 ^{ns}	

Table 4.8: Marital status of the mothers

^{ns} -not significant by t-test

The mean age for mothers of FPF group of children was 26 and that of mother of children of NFPF group was 25.8. There was no significant difference in the mean age of mothers between the mothers of the children of the two groups.

4.4.3 Morbidity status of study children

The most common illness by the study children were malaria, diarrhoea, pneumonia, common cold and cough with malaria contributing the highest percentage of 16.1% and 16.7% for FPF and NFPF followed by diarrhoea with 12.9% and 13.3% for FPF and NFPF and pneumonia was third contributing to 12.9% and 10% for children in FPF and NFPF respectively. Up to 48.4% and 50% of the study children in FPF and NFPF respectively did not have any illness by the start of the study as indicated in the Table 4.9. There were no significant differences in the proportion of children who were ill seven days prior the study in the two groups.

Other characteristic of study children at baseline are presented in the Table 4.10.The results shows that there was no significant difference in the child characteristic between the FPF and NFPF group. The children not fully immunized (n=3) were those who had not attained the immunization age of nine months for measles vaccine. In terms of immunization there was no significant difference in the two groups of children as shown in the Table 4.10. All the children in the two groups had received BCG, OPV, and DPT vaccines.

Type of disease	Gi	roups
	FPF (%)	NFPF (%)
No disease	48.4	50
Malaria	16.1	16.7
Diarrhoea	12.9	13.3
Pneumonia	3.2	3.3
Common cold	12.9	10
Cough	9.6	6.7

Table 4.9 Type of illness suffered by study children

Table 4.10 Other characteristic of study children

Characteristic	FPF (n=31)	NFPF(n=30)	P value	Statistical
				significant
Initial weight	7.16±1*	7.17±+0.79*	0.515	ns
Age (month)	13.5±4.9*	12.8±3.9*	0.699	ns
Age group				
6-11	48.4%	53.3%	0.964	ns
12-24	51.6%	46.7%	0.920	ns
Sex				
Male	45.2%	40.0	0.989	ns
Female	54.8%	60.0%	0.772	ns
Not fully immunized	3.7%	6.25	0.657	ns
Illness previous seven	48.4%	50%	0.922	ns
days				
Not dewormed	87.1%	100	0.504	ns
Child breastfeeding	90.3%	80%	0.263	ns

 $mean\pm s.d$, ns – Not significant by t-test or chi-square test

4.4.4 Vitamin A Supplementation

The numbers of children supplemented with vitamin A less than six months prior to the study are shown in the Table 4.11. Seventy two percent (72%) of the children had received vitamin A supplement prior to the study with 47.7% from FPF group and 52.3% from NFPF group. However there was no significant difference in the level of supplementation.

		Grou	Total	
		FPF	NFPF	
Vitamin A supplemented	Yes	21(47.7%)	23(52.3%)	44(72%)
	No	10(58.8%)	7(41.2%)	17(28%)
P value			0.312 ^{ns}	

Table	.11 Proportion of children who had received Vitamin A supplement	ation
less	han 6 months prior to the study	

4.5 Dietary intake

4.5.1 Food frequency analysis

The most commonly consumed food was maize meal in form of ugali, followed by irish potatoes, green bananas, rice, beans, milk, green grams, spinach, pumpkin, pawpaw, passion fruits, oranges, fat and sugar in the proportions of 71%, 74.2%, 87.1%, 51.6%, 74.2%, 83.9%, 67.7%, 64.5%, 54.8%, 51.6%, 54.9%, 54.8%, 64.5% and 58% of children from FPF group and 63.4%, 70%, 83.3%, 60%, 66.6%, 90%, 56.6%, 70%, 76.7%, 37.3%, , 66.6%, 66.7%, 66.7% and 56.7% of children from NFPF group respectively. Milk had the highest proportion of children consuming, followed by green bananas.. Most of the protein came from milk, green grams and beans, and carbohydrates from maize, irish potatoes and green bananas. The Irish potatoes were consumed in combination with either green bananas in mash or pumpkin only or combined. Fat was used for stewing food and sugar was added to porridge or milk. Sugar and fat had the highest contribution to energy intake of the study children. The frequency of consumption in the two groups of children is shown in the Table 4.12. There was no significant difference in frequency of consumption of

food commodities of the study children in the two groups (p>0.05). Sweet potatoes, yams, finger millet, sorghum, cabbage, night shade, pineapple, mangoes and watermelon despite their abundance in the local markets where most of the study children came from were rarely consumed. Also, yoghurt meat and fish were rarely consumed.

4.6 Dietary intake of study children

Food intake was measured by means of the 24- Hour dietary recall using a subsample of 30 children. The food intakes were used to calculate the intakes of total energy, protein, iron, zinc and vitamin A as retinol equivalents. The results are presented in Table 4.13 These results shows that the daily dietary energy intake before supplementation with porridge flour ranged between 650.4-1518.9 Kcal with a mean of 980.2kcal for FPF group and 484.4-1317.0 Kcal with a mean of 838.4 for NFPF group. After supplementation with porridge the daily energy range changed to 1032-1900 kcal with a mean of 1362Kcal, and 849-1682 Kcal with a mean of 1203Kcal for FPF and NFPF group respectively. The mean energy intakes before supplementation were below the recommended daily intake of 1022Kcal for children aged 6-36months (FAO, 2005). Supplementing children diet with the porridge flour improved the energy content for both group of children to above FAO recommended value, and children from FPF group consumed significantly higher (<0.05) energy after supplementation compared to children from NFPF group. This was contributed by higher energy levels of fermented porridge of 385Kcal compared to 365Kcal from the non fermented porridge.

The daily intake of protein ranged from 8.7- 41g with a mean of 25g FPF group and 1.4 - 35.7g with a mean of 17.7g for NFPF group before supplementation. After supplementing the children diet with the porridge flour the daily protein intake ranged from 17.9 - 50.2g with a mean 34.2g for FPF group and 10.2 - 44.5 g with a mean of 26.5g for NFPF group. The daily iron intake before supplementing the children diet with the porridge flour ranged between 2.0 - 13.1mg with a mean of

7.8mg and 1.6 - 9.2 mg with a mean of 5.7mg for FPF and NFPF group respectively. However on supplementation the children with the porridge flour the daily iron intake ranged from 6.9 - 18.3 mg with a mean of 13mg and 6.7 - 14.1 mg with a mean10.6mg of for the FPF and NFPF group respectively.

Food sources	FPF group (n=31)				NFPF group (n=30)			
	% Never consumed	1 -2 times a week	3times a week	4-7 times a week	% Never consumed	1 -2 times a week	3times a week	4-7 times a week
Spinach	35.5	9.7	22.6	32.2	20	23.4	10	46.6
Amaranth	77.4	3.2	6.5	12.9	73.3	3.3	10	13.4
leaves								
Kales	67.7	12.9	12.9	6.5	76.6	10	6.7	6.7
Pumpkin fruit	45.2	25.8	12.9	16.1	23.3	30	6.7	40
Tomatoes	93.6	3.2	0	3.2	90	0	3.3	6.7
Carrots	83.9	0	0	16.1	73.3	0	6.7	20
Pawpaw	48.4	38.7	3.2	9.7	63.3	23.4	3.3	6.7
Ripe bananas	61.3	6.4	6.4	25.9	56.7	23.3	6.7	13.3
Passion fruit	45.1	19.4	6.5	29	33.4	23.3	10	30
Black current	58.1	16.1	3.2	22.6	56.7	16.1	3.3	23.3
fruit								
Oranges	45.2	19.4	6.4	29	33.3	23.4	10	33.3
Green	12.9	32.3	16.1	38.7	16.7	23.3	6.7	43.3
bananas								
Finger millet	77.4	0	3.2	16.2	86.7	0	3.3	30
Irish potatoes	25.8	12.9	12.9	38.7	30	13.3	16.7	30
Sorghum	74.2	6.5	0	19.3	76.7	3.3	6.7	13.3
Maize meal	29	32.3	19.4	19.4	36.6	43.3	6.7	13.4
Rice	48.4	35.5	9.7	6.4	40	43.3	10	6.7
Beans	25.8	45.2	12.9	16.1	33.4	40	13.3	13.3
Green grams	32.3	48.4	16.1	3.2	43.4	33.3	13.3	10
Red meat	74.2	22.6	3.2	0	76.7	20	0	3.3
Eggs	54.8	45.2	0	0	53.3	46.7	0	0
Milk	16.1	6.5	0	77.4	10	3.3	10	76.7
Fat	35.5	3.2	0	61.3	33.3	6.7	3.3	56.7
Sugar	42	3.2	3.2	51.6	43.3	3.3	0	53.4

The daily zinc intake for the children ranged from 2.2 - 7.3 mg with a mean of 4.5mg and 1.3 - 7.9 mg with a mean of 3.4mg for FPF and NFPF respectively before supplementation and on supplementation it ranged from 4.8 - 9.9 mg with a mean of 7.1mg and 3.5 - 10.3 mg with a mean of 5.7mg for the FPF and NFPF group respectively.

The daily retinol equivalents intake for the children ranged from $32.8 - 1102.8 \ \mu g$ with a mean of 414.6µg and 4.9 - 717.9 µg with a mean of 397.3µg for FPF and NFPF group respectively. On supplementation the daily retinol equivalents ranged from 212.8 - 1248.1 µg with a mean of 592.2µg and 150.9 - 863.7 µg with a mean of 545.4µg for FPF and NFPF group respectively.

Supplementing children diet with porridge improved the daily protein, iron zinc and vitamin A as retinol equivalent intake of the study children. The mean protein and iron intake for study children in FPF group were significantly higher (p<0.05) after supplementing their diet with porridge flours than that of NFPF group. Fermentation improve protein quality and increase bioavailability of nutrient as reported earlier (Oniango *et al.*, 2002), and this could suggest the higher mean protein and iron intake of the children in FPF group after supplementing their diet with the porridge flour. Fermentation also improves nutrient availability (Hautrast *et al.*, 1989; Food chain, 1995; Oniango *et al.*, 2002; Hemalatha *et al.*, 2007) hence it was possible that the available protein, zinc and iron was better in the FPF than in the NFPF group.

Nutrient	FPF group (Mean ± sd)	NFPF Group(Mean ± sd)		
-	Before supplementation	After supplementation	Before supplementation	After supplementation	
Energy (Kcal)	980.2 ± 251.6	1362 ± 251.6*	838.4 ± 247.4	1203 ± 247.4	
Protein(g)	25.0 ± 10.4	34.2 ± 10.4*	17.7 ± 10.2	26.5 ± 10.2	
lron(mg)	7.8 ± 3.9	13.0 ± 3.8*	5.7 ± 2.3	10.6 ± 2.3	
Zinc(mg)	4.5 ± 1.6	7.1 ± 1.6	3.4 ± 2.2	5.7 ± 2.2	
Retinol(µg)	414.6 ± 349.1	592.2 ± 344.1	397.3 ± 225.3	545.4 ± 224	

Table 4.13 Dietary intake of study children before and after supplementation with porridge

* Significantly higher

Dietary adequacy of energy, protein, iron, zinc and vitamin A as RDA 4.6.1 Diet adequacy was determined by comparing the actual foods consumed with the RDA of energy, protein, iron, zinc and vitamin A as retinol equivalent for children aged 6-36 months. These values are shown in Tables 4.14 and 4.15. The RDA for energy before supplementation of the two groups was 95.9% for FPF group and 82% for NFPF group and after supplementation it improved to 133.3% and 117.7 % respectively. The RDA for protein before supplementation of the two groups was 187.2% for FPF group and 137.3% for NFPF group and after supplementation it improved to 255.5% and 206.4 % respectively. The RDA for iron before supplementation of the two groups was 121.2% for FPF group and 78.8% for NFPF group and after supplementation it improved to 201.1% and 147.7% respectively. The RDA for zinc before supplementation of the two groups was 54.1% for FPF group and 42.1% for NFPF group and after supplementation it improved to 85.0% and 70.5 % respectively. The RDA for vitamin A as retinol equivalent before supplementation of the two groups were 101.9% for FPF group and 99.3% for NFPF group and after supplementation it improved to 146.3% and 136.3% respectively. The protein, iron and vitamin A content of children in the FPF group were within the RDA(FAO 2005), while only protein in the NFPF group were within the RDA before supplementation (FAO 2005), however these difference in RDA for all nutrient were not significant within and between groups (p>0.05) before

supplementation. On supplementation the RDA were within the recommended range except for zinc in the two groups (FAO, 2005). The percent RDA for protein and iron (255.5% and 201.1%) for children in FPF group were significantly higher (p<0.05) than that of children in NFPF group (206.4% and 147.7%) on supplementation. The percent RDA range for energy before supplementation of the two group was 64-149% for FPF group and 47-129% for NFPF group and after supplementation the ranges changed to 101 - 186% and 83-165% respectively. The percent RDA range for protein before supplementation of the two group was 62 -293 % for FPF group and 10-255% for NFPF group and after supplementation the ranges changed to 128-139% and 73 - 318 % respectively. The percent RDA range for iron before supplementation of the two group was 22-146% for FPF group and 18-102% for NFPF group and after supplementation the ranges changed to 77-203% and 74-157% respectively. The percent RDA range for zinc before supplementation of the two group was 26-87% for FPF group and 15-94% for NFPF group and after supplementation the ranges changed to 57-118% and 42-123% respectively. The percent RDA range for vitamin A as retinol equivalent before supplementation of the two group was 8.2-276% for FPF group and 1.2-179% for NFPF group and after supplementation the ranges changed to 53-312% and 38-216% respectively. Before supplementing the children diets with the porridge 57.1% of children from FPF group and 75% from NFPF group did not meet RDA for energy but on supplementation with the porridge all the children in FPF were able to meet there RDA for energy and the proportion of children not able to meet there RDA for energy in the NFPF reduced to 18.8%. The proportion of children that did not meet there RDA for protein from FPF and NFPF were 14.3% and 37.5% before supplementing the children diet with the porridge but on supplementation all the children in FPF met there RDA and only 6.3% from NFPF did not meet RDA protein. In addition 37.5% from FPF and 75% from NFPF did not meet RDA for iron before supplementing the children diets with the porridges and on supplementation all the children in FPF were able to meet there RDA for iron and only 6.3% from NFPF did not meet there RDA. Also before supplementing the children diets with

the porridge 50 % from FPF and 57.1% from NFPF did not meet RDA for vitamin A as retinol equivalents and this proportion on supplementation reduced to 37.5% and 50% for FPF and NFPF respectively. All the children in the two groups did not meet RDA for zinc before supplementation but on supplementation only 78.6% from FPF and 87.5% from NFPF were unable to meet there RDA for zinc. Supplementing the children diet with the porridges improved the RDA for energy, protein, iron, zinc, and vitamin A as retinol equivalents for the two groups of children with children in FPF having a better outcome compared to those in NFPF. Fermentation leads to improved energy density, protein and micronutrient (Oniang'o *et al.*, 2002). This suggests the improved better outcome in FPF children on supplementation.

	·		FPF gi	roup			NFPF g	roup			
		Nutrient	Nutrient intake from home meal without				Nutrient intake from home meal without				
Nutrient		porridge				porridge					
	RDA	Mean intake mg/day	Mean % RDA intake	Mean RDA range	Proportion not meeting RDA (%)	Mean intake mg/day	Mean % RDA intake	Mean RDA range	Proportion not meeting RDA (%)	P value	
Energy (Kcal)	1022kca	980.2kcal	95.9±24.6	64-149	57.1	838.4±247.4	82±24.2	47-129	75	0.131 ^{ns}	
	1										
Protein(g)	14g	25.0±10.4	244.6±744	62-293	14.3	17.7±10.2	189.1±73. 2	10-255	37.5	0.061 ^{ns}	
Iron(mg)	9mg	7.9±3.6	87.5±40.5	22-146	37.5	5.7±2.2	63.4±24.9	18-102	75	0.057 ^{ns}	
Zinc (mg)	8.4mg	4.5±1.6	54.1±19.4	26-87	100	3.4±2.2	42.1±22.8	15-94	100	0.136 ^{ns}	
Retinol (µg)	400µg	414.6±349.1	101.9±89.6	8.2-276	57.1	397.3±225.3	99.3±56.3	1.2-179	50	0.925 ^{ns}	

Table 4.14 Dietary intake of study children without porridge supplementation

Table 4.15 Dietary intake of study children with porridge supplementation

			FPF gro	oup			NFPF gi	roup		
		Nutrient intake from home meal with porridge				Nutrient intake from home meal with porridge				
Nutrient		supplementation				supplementation				
	RDA	Mean intake mg/day	Mean % RDA intake	%Mean range RDA	Proportion not meeting RDA (%)	Mean intake mg/day	Mean RDA intake	%Mean range RDA	Proportion not meeting RDA (%)	P value
Energy(Kcal)	1022kcal	1362.2±254.6	133.3±24.6	101-186	0	1203.4±247.4	117.7±24.2	83-165	18.8	0.093 ^{ns}
Protein	14.1g	34.2±10.4	244.6±74.4	128-359	0	26.5±10.2	189.1±73.2	73-318	6.3	0.049*
Iron	9mg	13.0±3.7	144.8±41.1	77-203	0	10.6±2.3	118.7±24.4	74-157	6.3	0.041*
Zinc	8.4mg	7.1±16	85.0±19.5	57-118	78.6	5.7±2.2	70.5±23.0	42-123	87.5	0.074*
Retinol	400µg	592.2±344.1	146.3±88.4	53-312	50	545.4±224.0	136.3±56.0	38-216	37.5	0.713 ^{ns}

^{ns}- not significant by t test* -significantly higher

4.7 Knowledge of mothers on fermentation

Eighty nine percent (89 %) of mother whose children were fed on fermented flour had heard of fermentation but none had given the child fermented porridge and 75% of mothers whose children had been fed on non fermented flour had heard of fermentation and only 3% had given their children fermented porridge. Mothers had perception that fermented porridge is not good for young children. This is because they thought it would make them sick. Most of the mothers indicated that fermented porridge is good for pregnant and lactating mother and not for children, as they thought it may cause illness in children ranging from diarrhoea to stomach upset. There was no significant difference on fermentation knowledge of mothers/guardian of the two groups of children as shown in Table 4.16.

Characteristic	FPF	NFPF	p value	Statistical
				significant
Heard of fermentation	24(88.9%)	24(75)	0.172	ns
Given the child	0	1(3%)	0.354	ns
fermented porridge				

Table 4.16 Knowledge of mothers on fermentation

ns-not significant by chi-square test

4.8 Morbidity experience of study children during feeding

The most common illness during feeding final trials was malaria, cough, pneumonia and diarrhoea. Children in NFPF group had a higher percentage of children who were ill (46.7%) as compared to children in FPF group (12.9%). These differences were significant (p<0.05). This is shown in Table 4.17. Fermentation leads to increased acidity, and production of products such as peroxide, diacetyl secondary reaction products and bacteriocins which have an inhibitory role to microorganism (Oniang'o, *et al.*, 2002). The reduced microbial load in fermented product safe product with reduced microbial load and this suggest the low rate of diarrhea in children fed on fermented porridge. In addition fermentation leads to increase nutrition density of a food product as well as increase bioavailability of micronutrient (Oniang'o, *et al.*, 2002) hence children fed with fermented porridge had a better nutrition outcome as well as improved micronutrient as indicated in the nutritional and micronutrient status of the children that were fed with fermented porridge. Good nutrition status help an individual fight infection due to improved immunity (Disilvestro *et al.*, 2004) and this could suggest the low significant levels of illnesses for the FPF group of children during feeding.

		Illness						
		No disease	Malaria	Diarrhoea	Pneumonia	Cough		
Group	FPF	27(87.1%)	1(3.2%)	2(6.5%)	1(3.2%)	0	0.007*	
	NFPF	16(53.3%)	6(20%)	1(3.3%)	1(3.3%)	6(20%)		

able 4.17 Morbidity	experience	during	feeding for	the two	groups of	children
---------------------	------------	--------	-------------	---------	-----------	----------

* Significant difference by paired t test

4.9 Consumption and sharing of the porridge

Table 4.18 indicates consumption of porridge during follow up period throughout feeding period. The children who shared flour with other members of the family were 6.5% and 16.7% from FPF and NFPF group respectively. There was no significant difference between the children who were sharing flour in the two groups of children (p > 0.05).

Table 4.18 Sharing of porridge flour by children

Group	Children shar	P value	
	Yes	No	
FPF	6.5%	93.5%	0.217 ^{ns}
NFPF	16.7%	83.3%	

^{ns} not significant by paired t test

4.10 Nutritional status of the study children

Table 4.19, 4.20 and 4.21 show the prevalence of underweight, stunting and wasting before and after feeding of the study children. The prevalence of malnutrition was high in all the 61 study children. Using the NHCS/WHO 2007 cut off of -2, 80.3% of the children were moderately underweight (weight for age z-score of -2), 36.4% of the children were moderately stunted (height for age z score below -2) and 47.5% of the children were moderately wasted (weight for height z score below -2). Of the underweight, stunted and wasted children 51.0%, 19.7% and 22.9% came from the FPF group while 39.3%, 14.7% and 24.6% from NFPF respectively. There was no significant difference in proportion of underweight, stunted and wasted children between the two study group (p>0.05 before feeding.

On feeding after a period of six weeks the nutrition status of the children improved. The level of malnutrition with regards to stunting, underweight and wasting for both groups of children were higher before feeding, when compared to the national levels reported, where 22% were underweight, 30.2% stunted, and 5.5% wasted (CBS, 2004) before feeding. On feeding the children for six weeks the prevalence of underweight, stunting and wasting were lower when compared to the national figure of 22% underweight, 30.2% stunted, and 5.5% wasted (CBS, 2004). However, the prevalence was significantly lower (P<0.05) in the FPF group as compared to NFPF group post intervention.

Children in the FPF group had a 3.7kg gain in weight compared to 1.2kg for the NFPF group and 4.1cm gain in height for FPF compared to 1.9cm gain in NFPF group as shown Table 4.22. These change in weight was significantly different (p<0.05), however the change in height was not significant. The nutrition status of the children before and after feeding is shown in the Table 4.23. There was no significance difference (P>0.05) in nutrition status children between the two groups (FPF and NFPF) before feeding. However there was a significance difference

(p<0.05) in nutrition status after feeding with children fed on fermented porridge (FPF group) having a mean of WAZ= 0.71, HAZ=-0.52 and WHZ = 1.30 compared to mean WAZ= -1.31 HAZ=-1.44 and WHZ= -0.79 for NFPF group.

	Initial WAZ					WAZ post intervention				
Group	Normal	Moderately under	Mild under weight	Severe under weight	P- value	Normal	Moderately under	Mild under	Severe under	P - value
		weight					weight	weight	weight	
FPF (N=31)	0	25(41 %	6(9.8%)	0	0.92 ^{ns}	28(45.9%)	0	3(4.9%)	0	0.000*
NFPF (N=30)	0	24(39 %)	6(9.8%)	0		11(18.0%)	10(16.4%)	9(14.7%)	0	
TOTAL	0	49(80%)	12(19.7%)	0		39(63.9%)	10(16.4)	12(19.6%)	0	

Table 4.19 Prevalence of underweight before feeding and after feeding of study group

*-Significantly higher ^{ns}-not significant by t test

Table 4.20 Prevalence of stunting before feeding and after feeding of study group

	Initial HAZ					HAZ post intervention				
Group	Normal	Moderately stunted	Mild Stunted	Severe stunted	P value	Normal	Moderately stunted	Mild Stunted	Severe stunted	P- value
FPF (N=31)	0	12(19.7%)	11(18.0%)	8(13.1%)	0.811 ^{ns}	21(34.4%)	4(6.6%)	5(8.2%)	1(1.6%)	0.013*
NFPF (N=30)	4(6.5%)	9(14.7%)	8(13.1%)	9(14.8%)		11(18.0%)	10(16.4%)	6(9.8%)	3(4.9%)	
TOTAL	4(6.5%)	21(36.4%)	19(31.1%)	17(27.9%)		32(52.4%)	14(23%)	11(18.0%)	4(6.5%)	

*-Significantly higher ^{ns}-not significant by t test

	Initial WHZ							
Group	Normal	Moderately wasted	Mild wasted					
FPF (N=31)	3(4.9%)	14(22.9%)	11(18.0%)					
NFPF (N=30)	3(4.9%)	15(24.6%)	8(13.1%)					
TOTAL	6(9.8%)	29(47.5%)	19(31.1%)					

Table 4.21 Prevalence of wasting before feeding and

*-Significantly higher ^{ns}-not significant by t test

ner leeding	i of study g	roup				
		WHZ post i	ntervention			
Severe wasted	P value	Normal	Moderately wasted	Mild wasted	Severe Wasted	P- value
3(4.9%)	0.752 ^{ns}	30(49.2%)	0	1(1.6%)	0	0.000*
4(6.5%)		16(26.2%)	4(4.9%)	11(18.0%)	0	
7(11.5%)		46(75.4%)	4(4.9%)	12(19.7%)	0	

.

	FPF gr	oup(n=31)	NFPF group(n =30)		
variable	Before intervention	After intervention	Before intervention	After intervention	
Weight kg (Mean±sd)	7.2±01.0	10.9±2.4*	7.2±0.8	8.4±1.9	
Height cmMean±sd	71.8±5.8	75.9±7.1	71.3±4.7	73.2±6.0	

Table 4.22 Change in anthropometry during the intervention

Table 4.23 Mean Z score of study children at baseline and post intervention

	Baseline			End of int	tervention	
variable	FPF group n=31	NFPF group n=30	P value	FPF group n=31	NFPF group	P value
					n=30	
WAZ (mean ± sd)	-2.05±0.57	-2.19±0.63	0.344 ^{ns}	0.70±1.15	-1.31±1.24	0.000*
HAZ (mean ± sd)	-1.35±1.27	-1.33±1.29	0.961 ^{ns}	-0.52±1.10	-1.44±1.49	0.013*
WHZ (mean ± sd)	-1.52±0.91	-1.70±1.16	0.391 ^{ns}	1.30±0.67	-0.79±1.13	0.000*

WAZ- Weight for age Z score HAZ- Height for age Z score WHZ- Weight for height Z score FPF - Fermented porridge flour NFPF - None fermented porridge flour *significant higher FPF ns-no significant difference

The children were undernourished at baseline but on feeding for six week the proportion of those undernourished reduced. Those from FPF group had 9.7% underweight, 29.0% wasted and 3.2% stunted post intervention as compared to 63.3 % underweight, 63.3% wasted and 43.3% stunted in the NFPF group. The reduction

in level of malnutrition in the two groups suggested that supplementing children diet increased the energy intake and other nutrient of the diet consumed leading to improved nutrition status. However the difference in nutrition status post intervention of children in FPF group in terms of underweight, wasting and stunting as compared to that of children in the NFPF group were significantly higher (p<0.05) suggesting that fermentation of the flour could have been responsible of this. Supplementing the children diet with porridge flour increased the weight and height of the two groups; however the increase in weight was significantly higher (p<0.05) in FPF as they had a 2.4 kg increase in weight compared to NFPF. The significantly higher levels in children fed with fermented porridge (FPF group) could have been contributed by increased energy density of the porridge on fermentation (Oniango *et al.*, 2002, Hemalatha *et al.*, 2007.

There was no significant difference (p>0.05 in nutrition status of children aged 6-11 months and 12-24 months of the two groups at baseline and after feeding as shown in the Table 4.24

	Baseline		End of intervention				
	7-12 Group	13-24 Group	P value	7-12 Group	13-24	P valu	
	n=31	n=30		n=31	Group		
					n=30		
WAZ (mean \pm sd)	-2.38±0.38	-2.34±1.14	0.094 ^{ns}	-0.73±0.37	-0.04±1.72	0.116 "	
HAZ (mean ± sd)	-1.38±1.06	-1.76±1.24	0.676 ^{ns}	-1.08±1.46	-0.91±1.47	0.689 ¹¹⁸	
WHZ (mean \pm sd)	-2.20±0.69	-2.02±1.01	0.094 ^{ns}	-0.17±1.11	0.52±1.72	0.121 "	

Table 4.24 Nutrition status distribution by age at baseline and post intervention

ns-no significant difference

4.11 Micronutrient Status

4.11.1 Haemoglobin levels

Change in haemoglobin level of study children is shown in Table 4.25. There was no significant difference (P>0.05) in mean haemoglobin level of the children in the FPF and NFPF groups before start of feeding. However there was a significant difference in mean haemoglobin level between and with groups within children from FPF group having significantly higher levels (p<0.05) compared to children in NFPF group.

Table 4.25 Haemoglobin levels of study children

	FPF gro	oup (n=28)	NFPF group(n=27)	
Variable	Baseline	End	Baseline	End
Haemoglobin level(g/dl)	9.8±0.8	11.4±0.6*	9.5±0.7	10.9±0.5
Iron status(anaemia prevalence)	92.1%	14.3%*	92%	66.7%

significantly higher for FPF group

NB: A child is anaemic if haemoglobin level is below 11.0g/dl

The proportion of children with a mean haemoglobin concentration below 11.0g/dl before feeding among the FPF group (92.1%) was not significantly difference from that of NFPF group (92%). Therefore there was no significant difference in the proportion of anaemic children between the groups before feeding as shown in Table 4.25. The decrease in prevalence of anaemia post intervention among FPF children was significantly higher (p<0.05) compared to the drop in anaemia prevalence of NFPF children group. Fermentation leads to increased micronutrient availability (Oniango *et al.*, 2002, Hemalatha *et al.*, 2007) and could suggest the improved haemoglobin level in FPF group

4.11.2 Change in total serum protein

Comparison of the total serum protein concentration before and after feeding is presented in the Table 4.26. There was no significant difference in the mean total protein serum concentration for FPF and NFPF group at baseline between the

81

groups. However, the mean total serum protein levels were significantly higher (p<0.05) in FPF group compared to the NFPF group post intervention.

Study group	Baseline	P value	Post intervention	P value
FPF(n =28)	7.15+-0.13	0.300 ^{ns}	7.46+-0.20*	0.004
NFPF (n= 27)	7.10+-0.23		7.28+-0.23	

Table 4.26 Mean change of total	protein (g/dl) of the study children
---------------------------------	--------------------------------------

ns-no significant difference

*significant higher

4.11.3 Changes in total serum zinc

The changes in serum zinc biochemical index are shown in the Table 4.27. There was no significant difference in mean zinc level for FPF and NFPF group at baseline between the study groups. However, the mean total serum zinc levels were significantly higher (p<0.05) in FPF group compared to the NFPF group post intervention.

Table 4.27 Change in serum zinc

		FPF group(n=28)		NFPF group(n=27)	
Variable		Baseline	End	Baseline	End
Serum zinc levels		66.7±3.5	71.6±3.6*	65.8±2.7	69.2±2.9
Zinc status	Deficient (<65µg/dl)	9(32.1%)	2(7.1%)	11(40.7%)	2(7.4%)
category	Adequate level	19(67.9%)	26(92.9%)	16(59.3%)	25(92.6%)
	(>65µg/dl)				

* Significant higher

The percentage of children who had different categories of zinc level is shown in the Table 4.27. The proportion of children with a serum level of <65 μ g/dl in FPF group (32.1%) was not significant different between that of children of NFPF group (40.7%) at baseline. The mean zinc of 71.6 μ g/dl for children in FPF group was significantly higher (p<0.05) compared to 69.2 μ g/dl for children in NFPF.

The design of the study allowed for examination of the effect of fermented amaranth finger millet pumpkin porridge flour on nutritional, zinc, iron and vitamin A status of 6-24 months old children. The effect of the intervention on serum protein, serum zinc and haemoglobin level were significant post intervention with children fed with fermented porridge (FPF group) having significantly higher level (p<0.05) than those fed with non fermented porridge (NFPF group). Children from FPF group had 1.32g/dl, 0.26 μ g/dl), 1.42 μ g/dl) level of haemoglobin, serum zinc, and serum protein respectively increase higher than the children on NFPF group and these increases were significant (p<0.05).

Fermentation of cereal flour increases mineral availability as phytates and other antinutrients reduce, thus making the mineral more available on consumption of fermented cereals, as reported earlier (Oniango *et al.*, 2002). During fermentation phytate: zinc molar ratio reduces and fraction zinc absorption increases. Reduction of phytates by either enzyme or fermentation increases zinc absorption. This might suggest the significantly higher level in children of FPF group who consumed fermented porridge. (Navert and Sandstorm, 1985; Anderson and Gibson 2009). Thus feeding children on fermented cereal porridge benefits them from increased micronutrient availability compared to those fed on non- fermented porridge. This suggest to the higher significant levels of serum zinc and haemoglobin levels post intervention for children fed on fermented porridge compared to those fed on non fermented porridge. Also fermentation increase energy density and protein quality of cereal and legume product. This suggest the better nutritional status and higher serum protein level in children fed on fermented porridge as compared to those fed on none fermented porridge (Oniango *et al.*, 2002, Hemalatha *et al.*, 2007)

4.11.4 Change in serum retinol

The Table 4.28 shows that there was no significant difference at baseline and post intervention in mean serum retinol between the two groups of children (FPF and NFPF)

83

The differences in increase post intervention in serum retinol for FPF and NFPF groups of children within and between groups were not significant.

The percentage of children from each group who were in different categories of vitamin A status based on serum retinol is shown in Table 4.28 After the intervention 34.5% of children in FPF had normal levels of vitamin A based on serum retinol as compared to 27.3% category of children in the NFPF group but these changes were not significant between and within groups.



		FPF group (n=28)		NFPF group (n=27)	
Variable		Baseline	End	Baseline	end
Serum retinol		1.00±0.11	1.075±0.10	0.98±0.12	1.047±0.11
(µmoll ⁻ 1)					
Vitamin A	Adequate	18(32.7%)	9(16.4)	21(38.2%)	12(21.8%)
status category	(0.07- 1.05 µmoll ⁻ 1)				
	Normal (>1.05 µmoll ⁻ 1)	10(18.2%)	19(34.5%)	6(10.9%)	15(27.3%)

Table 4.28 Change of serum retinol (µmoll⁻¹) of the study children

Seventy two percent (72%) of the children had been supplemented with vitamin A less than 6 month prior the study, this might have contributed to the no significant difference post intervention despite feeding the children on different types of flour. However the increased in level of serum retinol post intervention might have been contributed by the pumpkin added to the porridge flour with children supplemented with FPF (1.07 μ moll⁻¹) having a slightly higher level compared to NFPF retinol levels of 1.04 μ moll⁻¹.

CHAPTER FIVE CONCLUSIONS AND RECOMMENDATIONS

5.1 CONCLUSIONS

Fermentation had an effect on carbohydrates, protein, crude lipid and fat levels of the flour as there was an increase in energy densities, protein and fat for the fermented flour as compared to the unfermented flour. Also total carbohydrates reduced in the fermented porridge flour because of their use as substrate for fermentation.

Supplementation of the children diet with the fermented and non fermented porridge flours improved their nutrition status, but children fed on fermented flour improved their nutrition status significantly higher as compared to those fed on non fermented porridge flour.

Supplementation of the children diet with the fermented and non fermented porridge flours improved their serum protein, serum zinc, haemoglobin and serum retinol levels. However use of fermented flour on the children improved the serum protein, serum zinc and haemoglobin level significantly higher as compared to the use of non fermented porridge flour. Lactic fermented porridges increase haemoglobin levels, serum zinc as well as serum protein in moderately malnourished children significantly, but no significant differences were noted between test and control group for serum retinol

5.2 **RECOMENDATIONS**

There is need to advocate for fermentation of cereals gruels used for feeding children to increases energy density and bio-availability of micronutrient, in order to solve the problem of protein energy malnutrition as well as micronutrient deficiencies. Use of fermented product need also be advocated for adult in addition to children as they will assist in solving malnutrition problems as well as reduced morbidity due to improved nutrition status that lead to improvement of immunity so as to assist individual fight infections.

There is need of involving a multi-disciplinary team of clinician to take physical and clinical history of children as well as adult before advocating use of fermented cereal gruel J as other fermented product in order to determine those individual that are sensitive to consumption of acid product and only advocate fermented products to those individual who are acid tolerant

There is also need for nutrition education on suitability of fermented products for children to mothers. This will aid in changing the perception of mother that use of fermented porridge on the children may cause diarrhoeas and other illness like stomach upset in children. By so doing children will benefit with the increased nutrition value of use of fermented food.

There is need to conduct bioavailability tests of protein, iron, zinc and vitamin A on the meals consumed by the children to determine how much of the nutrients taken in are actually utilized by the body as indicator for status and to assess the effect of fermentation.

There is need to conduct more studies on fermentation of products which have added vegetable and fruits to determine whether the process increase bioavailability of micronutrients.

References

AOAC (1999). Association of Official Analytical Chemists, Official Method of Analysis, Washington DC.

Anderson.P., and R. S. Gibson (2009). A review of intervention based on dietary diversification or modification strategies with the potential to enhance intakes of total and absorbed zinc. International zinc nutrition consultative group technical document (2), Systematic review of zinc intervention strategies, *Food and Nutrition Bulletin* (30), 1, 108-143.

Benoist.B., W.Daniel, I. Egli, C. Mary, and M. Erin (2007). World wide prevalence of Anemia in preschool children, pregnant women and non pregnant women of reproductive age. The Guidebook for Nutrition Anemia. Sight for Life Press. Basel, Switzerland.

BOSTID. (1996). Board of Science and Technology for International Development Lost crop of Africa, grains (Finger millet), 1, 35 – 57.

Brown, K. H., S.Y. Hess, J. C. King, and J.M.Peerson (2005). Use of serum zinc concentration as an indicator of population zinc status. *Food and Nutrition Bulletin* 23 (3), 403-429.

Bruno. B., I.H. Danrton., L. Davidson., and F.Olivier (2005). Report of a WHO/UNCEF/IAFA/IZINCG Interagency meeting on Zinc status indicators. IAEA Headquarters, Vienna. *Food and Nutrition Bulletin* 28 (3) 260-279.

CBS (2004). Kenya Demographic and Healthy Survey 2003, Central Bureau of Statistics, Nairobi, Kenya.

Dawson (1968). The determination of serum copper and zinc, Pye Unicam Limited, Cambridge, United Kingdom.

Disilvestro. R.A., J.S.Hampl, and G.M. Wardlaw (2004). Perspectives in Nutrition, Sixth dition, Mc Graw Hill companies, New York.

Eggum B.O., K.E. Knudsen, and B.Pedersen (2005). The nutritive value of Amaranths grain, Plant food and human nutrition, Springer Verlag Netherlands. 1, 61 – 71.

UNICEF (1999). Nutrition essentials. A guide for health managers. United Nations Children's Fund. Eds, Philips, R and Shawthey, P. Washington DC: 13-44.

FAO (2005). Daily recommended intakes for energy and nutrients for groups of people. Food Agricultural Organization, Family Nutrition Guide.

FAO/WHO (2001).Energy requirements of infants from birth to 12 months, Food and Nutrition Technical report series, Human energy requirements, Report of joint Food Agricultural Organization /World Health Organization/United Nation Union Expert Consultation, Rome. pp 11-19.

FAO/WHO 2002. Human vitamin and mineral requirements. Report of a joint Food Agricultural Organization /World Health Organization Expert Consultation. Bangkok, Thailand, 21–30 September 1998. Food and Nutrition Division, Rome.

FAO/WHO/UNU (2004). Energy needs in Human Energy Requirements. Report of a Joint Food Agricultural Organization /World Health Organization/United Nation Union Expert Consultation. Rome, 17-24 October 2001, FAO, Food and Nutrition Technical Report Series(1) Rome. Food Chain (1995). Yeast, mold and bacteria, and Indigenous fermented cereals in Kenya, ITDG 15, 9-16.

FSAU (2003). Nutriton a guide to data collection, analysis, interpretataion and use, Food Security Assessment Unit of Somalia, Nairobi, Kenya.

Grases .F., J.G.March, R. M.Prieto, and B. N.Simonet (2001). Phytates level in diverse rat tissue: Influence of dietary phytates. *British Journal of Nutrition* 86 (2)(225).

Guthrie. H. A, and F. M.Picciano (1995). Human nutrition. Times Mirror Mosby, St. Louis, Missouri.

Hautrast G. J., M. J. R. Nout, and F. Rombouts (1989). Accelerated natural lactic fermentation of infant food formulations. *Food and Nutrition Bulletin* 11 (1), 50-59.

Hemalatha S., K. Platel, and K. Srinivasan (2007). Influence of germination and fermentation on bio accessibility of zinc and iron from food grains. *European Journal of Clinical Nutrition* 61, 342–348.

Hofuande and B.A.Underwood (1987). Processed supplementary foods for older infants and young children, with special reference to developing countries. *Food and Nutrition Bulletin* 9 (1), 1-7.

Importance of finger millet. <u>http://www.blackherbals.com/bd14538</u>, assessed on 15/5/2008 at 3.0 0pm

INACG (1979). Iron deficiency in infancy and childhood, laboratory diagnosis of iron deficiency. A report of the International Nutritional Anaemia Consultative Group, The Nutrition Foundation, Washgton, D.C. pp 29.

KEBS/CAC (1997). Guidelines on formulated supplementary foods for older infants and young children. Kenya Bureau of Standards, / Codex Alimentarius Commission, Nairobi, Kenya.

Kirkwood, B. R (1988). Essential of medical statistics, calculation of sample size, Blackwell Scientific Publications, Oxford, London.

Lotfi, M., M. G. V.Mannar, R. J. H. M. Merx, and P. N. Heuvel (1996). Micronutrient fortification of foods, Current practices, research, and opportunities, Micronutrient Initiative and International Agricultural Centre.

Madibela O.R, and Modiakgotia (2004), Chemical composition and in Vitro dry matter digestibility of indigenous finger millet (Eleusine coracana), Livestock research for rural development, Botswana.

Manner. V (2007). The case for urgent action to address nutrition anemia in Badham. J., K.Klaus, Z. and B. Michael. The guidebook for Nutriton Anemia. Sight for Life Press. Basel, Switzerland. Pp 12-13.

Meitzner, L. S., and M. L.Price (1996). Amaranth to Zai Holes, Ideas for Growing Food under Difficult Conditions. Intermediate Technology, ECHO: North Fort Myers, Florida

Mitra S., Hea- Ran Lee Ashraf (1993), International Journal of Food Science and Technology 28(6).

MOH (1999). The Kenya National Mcronutrient Survey Report. Vitamin A, status of Iron and Zinc in Kenya, Minstry of Health, Nairobi, Kenya.
MOH (2009). Thika district Annual Operation Plan 5 (AOP 5) Report, Minstry of Health, Nairobi, Kenya.

MOMS/MOPHS (2009). National Guideline for Management of Acute Malnutrition. Ministry of Medical Services and Ministry of Public Health AND Sanitation, Nairobi, Kenya.

Morales. E., J. Lembek, and G,G.Grahams (1998). Nutritional value for young children of grain Amaranths and maize – amaranth mixture ; Effect of processing, *Journal of Nutriton*111:78-85.

Narvert B., and B. Sandstorm (1985). Reduction of the phytates content of bran by leavening in bread and its effect absorbtion in man. *British Journal of Nutrition* 53, 47-63

O'Dell. B. L., and R. A. Sunde, (1997). Handbook of nutrition essential mineral elements, Library of Congress, New York.

Oniango R. K., A. O.Makoha, S.M. Njoroge, and O. K.Kamar (2002). Effect of traditional fermentation and malting on phytic acid and mineral availability from sorghum *(Sorghum bicolour)* and finger millet (*Eleusine coracana*) grain varieties grown in Kenya. *Food and Nutrition Bulletin* (24) 241-245.

Railey. K (1999). Amaranthus, A Health grain for Vegetarian Recipes

Rathod, P., and S.A. Udupi (1991). The nutritional quality and acceptability of weaning foods incorporating amaranth, *Food and Nutrition Bulletin*, 13, (1),70-82

Sehmi. J (1993). National food composition table, Government of Kenya GOK, Nairobi.

Sharma A., and A.C. Kapoor (1994). Effect of various types of fermentation on invitro protein and starch digestibility of different processed pearl millet. Nahrung,40:142-145.

Singh, R. S., P. R.Kulkami (1988). Review; Amaranths an underutilized resource. International Journal of Food Science and Technology (23) 125-139.

Sripriya. G., U.Antony, T. S. Chandra (1997). Changes in carbohydrate, free amino acids, organic acids, phytate and HCl extractability of minerals during germination and fermentation of finger millet (Eleusine coracana), *Journal of Food Chemistry* 58 (4), 345-350.

Tina S., V.A. Mark, B. Jean, and F. John F (2007), A Report for Ten Year Strategy in Reduction of Vitamin and Mineral Deficiency; Vitamin and Mineral Deficiency Technical Situation Analysis, *Food and Nutrition Bulletin*, 28, (1), 160-161.

Tina.S., B.Reena, and H.Robin (2007). What is the extent of vitamin and mineral deficiencies? Magnitude and problem . *Food and Nutrition Bulletin*, 28 (1), 174-181

Tomkins, A., and F.Watson (1993). Malnutrition and infection, Nutrition Policy Discussion Paper, 5, 13-19.

WHO (1985). Energy and protein requirements, Technical Report Series 724, World Health Organization, Geneva.

www.http:/lifestyle.iloveindia.com/lounge-benefit of pumpkin assessed on 19/2/2010 at 9.37pm- Benefit of pumpkin. Appendix 1: Data collection tools

TITLE OF THE STUDY:

Effect of lactic acid fermented amaranths-finger millet-pumpkin composite porridge flour on nutritional, vitamin a, zinc, and iron status of malnourished children age 7- 24 months in Thika district hospital

1. Proximate, Vitamin A and mineral analysis form

TECHINICIAN NAME:	1)
	2)
DATE:	

Proximate composition, vitamin A, zinc, and iron composition of the composite

flours

	Fermented amaranth-finger millet-pumpkin flour	Non Fermented amaranth- finger millet-pumpkin flour
Crude fibre		
Crude protein		
Total ash		
Moisture		
content (for		
flours)		
Vitamin A(beta-		
carotene)		
Zinc		
Iron		
Carbohydrates		
Energy density		

2. Microbial analysis

	Fermented amaranth- finger millet- pumpkin composite porridge flour	None Fermented amaranth- finger millet- pumpkin composite porridge flour
Total plate count		
coliform		
Yeast and molds		
Ph		

3. Sensory evaluation

TITLE OF THE STUDY:

Effect of lactic acid fermented amaranths-finger millet-pumpkin composite porridge flour on nutritional, vitamin a,

zinc, and iron status of malnourished children aged 7-24 months in Thika district hospital

Date: -----

Code	Hedonic scale	Type 1(F	Type 1(Fermented porridge)				Type 2(no fermented porridge					
		Colour	Consistency	Flavour	Taste	Overall acceptability	Colour	Consistency	Flavour	Taste	Overall acceptability	
1	Like very much											
2	Like moderately											
3	Like slightly											
4	Neither like nor											
	dislike											
5	Dislike slightly											
6	Dislike											
	moderately								1.1			
7	Dislike very											
	much											

4. Field Questionnaire

TITLE OF THE STUDY:

This is a study to find out the effect and quality of lactic acid fermented amaranthsfinger millet-pumpkin composite porridge flour on nutritional status of malnourished children aged 7- 24 months attending Maternal Child Health clinic in Thika district hospital. You're requested to voluntary answer the questions and you are assured that the data will be used for the purpose of this study and will be treated with confidentiality and care.

Questionnaire no []

Name of the interviewer.....

Social economic and demographic information

Give information about member of the household starting with head of house hold.

No.	Name	Sex 1= male 2= female	Age*	Educati on	Occupation	Income I = yes 2= no	Marital status	Relationship to child
1.								
2.								
3.								
4.						_		
5.								
6.								
7.								

For adult record age in years and children < 5 years record age in months.

Sex	Age	Marital Status	Education level
	Record in years older than 5 years		
1=Male		1=Single	1=Uneducated
	Record in months for	2=Married	
2=Female	children below 5 years	monogamously	(Never been to school)
		(includes	2=Pre-pri and lower
Occupation	Relation to child	cohabiting)	pri(std 3)
1=Permanent		3=Married	
employment	1=Mother	polygamous	3=Upper pri (std 4-8)
			4=Secondary
2=Temporary			
employment	2=Father	4=Divorced	
			5=preschool
3=Business	3=Grand parent	5=Widowed	
4=Casual laborers	4=Uncle/auntie	6=not applicable	6=Post secondary.
5=Unemployed	5= Sibling		
6=Farming	6 =Cousin		
7=student	7= Other(specify)		

```
Codes
```

2. Who takes care of the child? 1= Mother, 2= Father, 3=								
Grandmother/grandfather, 4= Aunt/uncle,								
5 = other specify.								
3. What is the main source of household in	(can have							
multiple response)								
1 = sales from farm produce	2 = casual labour	3 = formal						
employment								
4 = funds from relative	5 = business	6 = others						
(specify)								

4. Health and Nutrition status of the child

Immunization, vitamin A supplementation, deworming and growth

monitoring

Name	of child		Date	e of birth.		Sex		• • • • • • • • • • • • •		
Immu	inization								Vit.A last 6 months	Deworme d last 3months
BC G	OPV0	DPT1	OPV1	DPT2	OPV2	DPT3	OPV3	Measles		

5. **Disease morbidity**

Has the child been sick in the last 2 weeks [Indicate 1 or 2]

2 = no [skip to part]1= yes

If yes is the treatment card available? [Indicate 1 or 2] 1 = yes

$$2 = no$$

If the child has been sick, what type of illness was it, duration of illness, symptom and action taken.[enter information in table below using the codes

below the table for symptom and action taken]

Type of illness	No. of episode	Duration of illness(days)	Symptoms*	Source of treatment
4				* *
Malaria				
Diarrhoea				
Worms				
Wounds				
Bronchitis				
Pneumonia				
Common cold				
Otitis media				
Cough				
Amoebosis				
Typhoid				

Codes for symptom and source of treatment

Sym	ptom (multiple choice allowed)		Action taken
1	fever	1	went to health facility
2	vomiting	2	herbal treatment
3	running nose	3	self medication
4	loss of appetite	4	faith healing
5	weight loss	5	other specify
6	cough		
7	wheezing		
8	blood in stool		
9	difficult in breathing		
10	loose stool		
11	other(specify)		

6. Food consumption pattern

Food frequencies

I would like to know the type of food your child consumes in a week, fortnightly,

and monthly and rarely consumed and number of times per week they consumed

Foods	Frequency per fortnight	Frequenc y per month	Rarely consumed	7 day recall							
				0	1	2	3	4	5	6	7
Cereals											
Maize									-	-	
Sorghum											
Rice											
Irish						1					
potatoes				-							
Wheat				1							
Yams											
Arrow roots											
Sweet											
potatoes											
Finger											
millet											
Raw											
bananas											
Mixed flour											
(specify)											
Fruits and											
vegetables					ļ						
Spinach								_			
Kales											
Cabbage											
Pumpkin											

Managu						
Amaranths						
leaves						
Pumpkin						
leaves						
Pawpaw						
Water						
melon						
Pineapple						
Passion						
fruits						
Apple						
Oranges						
Ribena						
fruits						
Avocado						
Carrots						
Ripe banana						
Proteins						
Milk and						
milk						
product					 	
Milk						
Yorghut						
Mala						
Cheese						
Butter						
Animal						
products						
Eggs						
Meat (beaf)						
Pork						

Fish				
Omena				
Vegetable				
proteins				
Pigeon peas				
Peas				
Beans				
Lettuce				
Green				
grams				
Njahi				
Fat and oils				
Fat				
Oils				
Sugar				
Honey				

Have you ever heard of fermentation? [Indicate 1 or 2]-----

1. Yes 2. No

If yes do you ferment your child flour for porridge? [Indicate 1 or 2]-----

1. Yes 2. No

For the period you have given the child fermented porridge has (s) he

experienced diarrhea or any other disease. [indicate1 or2]-----

1. Yes 2. No

7. Anthropometric information

a. Baseline data

Now I would like to take the weight and height of your child.

Weight			Height		
Weight 1	Weight 2	Average	Height 1	Height 2	average

Date of birth -----

8 Morbidity recall

Has the child been ill during the feeding period?

Type of	Date of	Duration	Accompanying	Sources of
illness	illness		symptom	treatment.
Malaria				
Pneumonia				
Cough				
Diarrhoea				
Other				
specify				

Syn	nptom (multiple choice allowed)		Action taken
1	Fever	1	Went to health facility
2	Vomiting	2	Herbal treatment
3	Running nose	3	Self medication
4	Loss of appetite	4	Faith healing
5	Weight loss	5	Other specify
6	Cough		
7	Wheezing		
8	Blood in stool		
9	Difficult in breathing		
10	Loose stool		
11	other(specify)		

Has the child been breastfeeding?------

1 Yes 2 No

Did the child share the flour with other members of the family?.....

1 Yes 2 No

Anthropometric and biochemical data

a. Record of child weight for six weeks

Week	Weight					
	Weight 1	Weight 2	Average	Height 1	Height 2	Average
1						
2						
3						-
4						
5						
6						

b. Record of child biochemical indices

Period	Hb	Serum zinc	Serum retinol	Serum protein
Baseline				
After six weeks				

24-HOUR RECALL

Time	Dish	Name of Ingredients Used preparation	HH Measure used	Amount In H/H measure	Amount of ingredient In the dish	Volume of Complete Dish prepared	Volume of food served to the child	Amount Of left over	Food Consumed by the child

24-HOUR RECALL FOOD INTAKE RECORD SHEET

[Take household measures: tea spoon (tsps),table spoon(tbsp),cups, mugs, measuring jugs and sufurias and the food ingredients used in the household to show the exact amounts, sizes and units used in food preparation. Request to see the measures, sizes, foods and ingredients in order to practically measure and weigh the used levels].

1=tea spoon (tsp), 2=tablespoon (tbsp), 3=cup 1, 4=cup2, 5=cup3, 6=cup4, 7=mug, 8=measuring jugs, 9=sufurias, 10=bowl, 11=plate, 12=pot HH – house hold Appendix 2: Laboratory request form

Title of the study: effect and quality of lactic acid fermented amaranthsfinger millet-pumpkin composite porridge flour on nutritional status of malnourished children aged 7- 24 months attending Maternal Child Health clinic in Thika district hospital.

Name of Name of	analyzing ins laboratory an	stitution alyst				-		
2.								
Examina	ation requested	d for 2 pha	ises					
Child	Haemoglobin		Serum protein		Serum zinc		Serum retinol	
no.								
	Base line	After six weeks	Base line	After six weeks	Base line	After six weeks	Basel ine	After six weeks

Appendix 3 Training Curriculum

A training curriculum was developed for quality data Two training curriculum were used

Formulation of the flours

One of the research assistant was trained to assist the principal investigator on formulation of the flours. The research assistant who was used was a holder of BSC. Food Nutrition and Dietetic

Objectives

By the end of the training the research assistant was supposed to be able to describe the procedure for the formulation of the fermented and none fermented amaranthfinger millet- pumpkin composite porridge flour and be able to prepare them with the assistant of principal investigator. The preparation was as figure 3.3 and 3.4.

Training of Field research assistant

The field assistant included three nutritionist from Thika district hospital.

Objective

After the end of the training the research assistants were supposed to be able to;

- Explain recipe for porridge to the guardian of the children's,
- Administer the questionnaires to the guardian of the children,
- Take weight, height and determine the age,
- Take data on dietary intake and food consumption of the children's,
- Follow ethical issue and observe human right issues through out the study period,

• Follow up children at home and give a report on compliance to the feeds. In addition a nurse and a clinical officer trained on integrated management of childhood illness was involved throughout the study period to identify and treat any disease condition.

Curriculum

Rules and guidelines

The clinical officer, the nurse and the nutritionist were informed about the study, the children to be recruited in the study and ethical consideration to be followed through out the study period. Importance of observing confidentiality was emphasized. Other rules included were punctuality and remuneration.

Questionnaire briefing

The three research assistants were taken through the questionnaire so as to grasp what was expected of them. Emphasis was put on,

- Seeking consent from interviewer,
- How to ask questions,
- Probing where necessary,
- How to determine dietary intake using 24-hr recall and food consumption pattern using food frequency form.
- Exit strategy

The questionnaire was to be administered and anthropometric done after other question have been asked.

Procedure before weighing

I. Procedure before measuring

- Two trained enumerators are required to take a child's height. The measurer holds the child and takes the measurements. The assistant helps hold the child and records the measurements on the questionnaire.
- Measuring board and scale should be on a flat surface.
- Age should be determined to be within the range (7-24 months)
- Weigh and measure after verbal information has been recorded.
- Weigh one at a time, not all the children (complete questionnaire then weigh)
- Explain the weighing procedures to the guardian.

 Record measurements clearly in pencil. Keep pencils out of hands hair or breast pocket while weighing.

II. Measuring length

For children below 24 months the length is taken using a length board. Procedure for length taking was explained to the research assistant as described below and a practical off the same done.

• Procedure

- Place the length board on a hard flat surface on a flat bench.
- Ask the guardian of the child to remove shoes and unbraid hair that could interfere with the measurements.
- Ask the guardian of the child to place on the board with the head positioned against the fixed head board and eyes looking vertically.
- Extend the knees and flex the feet at right angle to the lower legs.
- Move the up sliding foot piece until a firm cont with heal is obtained.
- Take the reading and repeat the process again
- Take two reading to the nearest 0.1 cm and record.

III. Weight

Weight was taken using Salter scale and procedure was explained as described below.

Procedure

- i. Calibrate the Salter scale with known weight before taking the child weight.
- ii. Request the guardian of the child to undress the child and leave him/her with minimal clothing.
- iii. Request the guardian to place the child on the scale.
- iv. Record the reading and repeat the procedure again.
- v. Record the reading to the nearest 0.1kg.
- vi. Request the guardian remove the child from balance and dress him/her.

Dietary intake

Demonstration on how to do a 24 hour dietary recall and food frequency was also done by the principal investigator as the research assistant learned.

Training the guardians of the children's

The guardians were trained on how to prepare the porridge for the children's as per procedure below.

Procedure

- Weigh a 100 gram of the porridge (household measure with an equivalent of 100 gm was used).
- Mix the flour with water to make slurry.
- Weigh 900 ml of water and put in a sufuria, and heat to boil.
- Mix the slurry prepared earlier with the boiling water as you stir.
- After the porridge has thickened boil for 10 minutes.
- Cool and serve the porridge to the child 3 or 4 time a day
- Record amount consumed and amount in the form provided.

The guardian was also trained on how to record children porridge intake per serving and total intake per day was calculated by principal investigator.

Appendix 4: Informed Consent Form

This is a study to find out the effect and quality of lactic acid fermented amaranths-finger millet-pumpkin composite porridge flour on nutritional status of malnourished children aged 7- 24 months attending Maternal Child Health clinic in Thika district hospital. Participation to this study is voluntary and you're requested if you are willing to participate in the study to sign informed consent form.

Informed consent

I have understood all the procedure involved in this study and I hereby consent to participate and adhere to the protocol of the intervention feeding study.

Date
Signature of the parent
District Medical Officer of Health
District Nutritionist
Principal investigators