

**EFFECTS OF IRON FORTIFICATION AND
FERMENTATION ON PROCESSING, FUNCTIONAL,
MICROBIOLOGICAL AND STORAGE PROPERTIES OF
MAIZE-BLEND WEANING FOODS.**

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By

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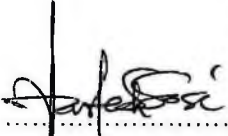
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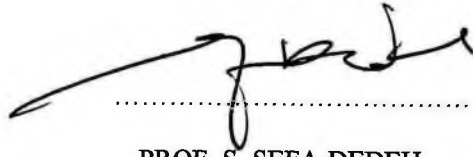
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DECLARATION

This research was conducted by me under the supervision of Prof. Sefa-Dedeh of the Nutrition and Food Science Department, University of Ghana, Legon.

A handwritten signature in black ink, appearing to read 'Sosa', written over a horizontal dotted line.

SAMUEL NARTEH SOSI

A handwritten signature in black ink, appearing to read 'Sefa-Dedeh', written over a horizontal dotted line.

PROF. S. SEFA-DEDEH

DEDICATION

To God the almighty and then to my wife, children and my father-in-law.

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Above all, to God be the glory.

ABSTRACT

Iron deficiency anaemia (IDA) has been a significant public health problem in the developing world. Fortification has been identified as one of the most cost-effective and sustainable approaches for controlling IDA. Addition of various bionutritional substances creates several technical challenges for the Food Technologists and therefore it is necessary to maintain the overall quality of the product in terms of the bioavailability of the fortifying agent.

The main objective of this work is therefore to investigate the effects of iron fortification on the physical, functional and microbiological properties of weaning foods. The iron compounds used were ferrous sulphate and ferrous fumarate.

Oven drying at 60°C was used to reduce the moisture content of the fermented maize dough to produce dehydrated product. A 4x3x2x2 factorial experiment with, fermentation time, iron compound, iron concentration and the mode of iron addition as variables were replicated. The indices measured were pH, titratable acidity, colour, fat acidity, water absorption, viscosity, total iron content, energy content and microbiological study.

A roasted weaning food - maize/cowpea/groundnut blend was prepared and response surface methodology studies using a central composite rotatable design for factor, $k=3$ (iron concentration, legume concentration and roasting temperature) conducted.

Samples were packaged in polyethylene bags and stored for 4 months and analysed for pH, titratable acidity, colour, water absorption and fat acidity.

Sensory evaluation/consumer acceptability test was performed on three iron-fortified cooked products namely, dehydrated fermented maize dough, maize/cowpea dough and roasted maize /cowpea/groundnut blend.

Even though iron fortification brings about relative increase in pH, the values (3.51-

4.11) were optimal for cereal phytase in fermented maize dough system. Breakdown of triglycerides was high in both iron-fortified and non-fortified products. Fermentation and iron addition were found to increase the total iron content of the products. Energy content of at least 460 kcal/100g was recorded for all samples. This is higher than a recommended energy for a quality weaning food. The concentration of iron added before fermentation did not interfere with the microbial growth of the fermented maize signifying that the levels added were not toxic to the microorganisms.

Even though storage had significant effect on the indexes measured, there was not much change in the characteristics when comparing the iron-fortified and the non-fortified products.

Regression models developed to predict product characteristics of roasted weaning food were influenced by the process variables. These were mainly legume concentration and roasting temperature. Iron addition however, affected the characteristics when legume concentration was high. Storage of the roasted blend was highly affected by iron addition.

Products were generally acceptable to mothers from the sensory evaluation and consumer acceptability test conducted in five communities in the Central region.

TABLE OF CONTENTS

DECLARATION	i
DEDICATION	ii
ACKNOWLEDGEMENT	iii
ABSTRACT	v
TABLE OF CONTENTS	vii
LIST OF TABLES	xii
LIST OF FIGURES	xiv
1.0 INTRODUCTION	1
1.1 Malnutrition	1
1.2 Protein Malnutrition (PEM)	1
1.3 Micronutrient Malnutrition (MM)	2
1.3.1 Vitamin A Deficiency (VAD)	3
1.3.2 Iodine Deficiency Disorders (IDD)	4
1.3.3 Iron Deficiency Anaemia (IDA)	4
1.4 Strategies to combat infant malnutrition	5
1.5 Proposed solution of micronutrient malnutrition	6
1.5.1 Technical challenges of mineral fortification	7
1.6 Objectives	9
2.0 LITERATURE REVIEW	10
2.1 Weaning practices	10
2.2 Weaning foods	10
2.2.1 Traditional weaning foods	11
2.3 Cereal and legume processing technologies	12
2.3.1 Soaking	12

2.3.2 Roasting	12
2.3.3 Fermentation	13
2.4. Functional and Chemical Properties of Weaning Food	14
2.5 Food Fortification	15
2.5.1 Protein Fortification	16
2.6.2 Mineral Fortification	17
2.6.2.1 Iron fortification	18
2.6.2.1.1 Level of Iron Fortification	20
2.6.2.2 Effect of iron fortification on food quality	22
2.6.2.2.1 Reduction potential and pH	22
2.6.2.2.2 Colour	23
2.6.2.2.3 Flavour	24
2.6.2.2.4 Energy	25
2.6.2.2.5 Effects of processing	26
2.6.2.2.6 Other quality changes	27
2.6.2.3 Toxicity	28
3.0 MATERIALS AND METHODS	30
3.1 MATERIALS	30
3.2 METHODS	30
3.2.1 Calculation of iron added to food samples	30
3.2.2 Effect of fermentation and drying on iron-fortified maize dough.	30
3.2.2.1 Sample Preparation	30
3.2.2.2 Analysis on fresh samples	31
3.2.2.3 Experimental Design	33
3.2.3 Effect on storage on dehydrated iron-fortified fermented maize dough flour.	33



3.2.4 Optimisation of concentration of legume and iron in roasting weaning blend.	33
3.2.4.1 Sample Preparation	33
3.2.4.2 Experimental Design and Statistical Analysis	34
3.2.5 Effect of storage on iron- fortified roasted weaning foods.	37
3.2.6 Laboratory Studies	37
3.2.6.1 Colour of food samples	37
3.2.6.2 pH and Titratable Acidity	38
3.2.6.3 Fat acidity	38
3.2.6.4 Total iron determination	39
3.2.6.5 Energy	40
3.2.6.6 Water absorption	40
3.2.6.7 Apparent Hot and Cooled Paste Viscosity	40
3.2.7 Microbiological analyses	41
3.2.8 Sensory Evaluation through field trial	41
3.2.8.1 Field Survey - questionnaire administration	41
3.2.8.2 Sample preparation of test samples used.	42
3.2.8.3 Sample Presentation	43
4.0 RESULTS AND DISCUSSION	44
4.1 Effect of fermentation on iron fortified maize dough	44
4.1.1 pH	44
4.1.2 Titratable acidity	47
4.2 Effect of fermentation and drying on iron-fortified maize dough	50
4.2.1 pH	50
4.2.2 Titratable acidity	52
4.2.3 Fat acidity	54

4.2.4 Colour	58
4.2.5 Water absorption index	61
4.2.6 Apparent Hot paste viscosity (90°C)	65
4.2.7 Apparent cooled paste viscosity (50°C)	68
4.2.8 Energy content	70
4.2.9 Total iron content	72
4.2.10 Microbiological Evaluation	74
4.3 Storage stability of iron-fortified and non-fortified dehydrated fermented maize dough	78
4.3.1 pH and Titratable acidity	78
4.3.2 Fat acidity	79
4.3.3 Colour	80
4.3.4 Water absorption index	80
4.4 Optimisation of concentration of legume and iron in roasting weaning blend	80
4.4.1 Colour	83
4.4.2 Fat acidity	83
4.4.3 Water absorption index	85
4.4.4 Apparent paste viscosity	87
4.4.5 Energy content	90
4.4.6 Total iron content	93
4.5 Storage stability of iron-fortified roasted blend	96
4.5.1 Fat acidity	96
4.5.2 Water absorption index	96
4.6 Sensory evaluation through field trials	100
4.6.1 Background data on respondents	100
4.6.2 Sensory evaluation of some quality attributes	101

4.6.3 Cost	106
5.0 CONCLUSIONS	108
5.1 Effect of fermentation and drying on iron fortification of fermented maize dough	108
5.2 Microbiological evaluation	108
5.3 Response surface methodology study on roasted maize/cowpea/groundnut blend	109
5.4 Shelf stability	109
5.5 Sensory evaluation/consumer acceptability	110
REFERENCES	112
APPENDICES	131

LIST OF TABLES

Table 1 Relative bioavailability of iron compounds used in food fortification (%)	19
Table 2 Recommended dietary intake (RDI) for iron (mg/day)	21
Table 3 Relationship between iron requirements and dietary iron supply	26
Table 4 Mineral Safety Indexes	29
Table 5 Process variables used in the Central Composite Rotatable Design (K = 3)	34
Table 6 Design matrix and variable combinations in experimental runs	36
Table 7 ANOVA summary table of pH on fresh and dried fermented maize dough samples	45
Table 8 ANOVA summary table of acidity on fresh and dried fermented maize dough samples	48
Table 9 ANOVA summary table of fat acidity on dehydrated and stored fermented maize dough	55
Table 10 Hunter colour values for dehydrated fermented maize samples fortified with ferrous sulphate	59
Table 11 Hunter colour values for dehydrated fermented maize samples fortified with ferrous fumarate	60
Table 12 ANOVA summary table of colour on dehydrated and stored fermented maize dough samples	61
Table 13 ANOVA summary table of water absorption on dehydrated and stored fermented maize dough samples	65
Table 14 ANOVA summary table of viscosity on dehydrated fermented maize dough samples	68
Table 15 Effect of fermentation and iron fortification on energy content of dehydrated maize dough (kcal/100g)	71
Table 16 ANOVA summary table of energy on dehydrated fermented maize	

dough samples	71
Table 17 Effect of fermentation and iron fortification on total iron content of dehydratedmaize dough (mg/100g)	73
Table 18 ANOVA summary table of total iron content on dehydrated fermented maize dough samples	74
Table 19 Effect of fermentation on total bacterial count for microorganisms of iron-fortified and non-fortified maize dough	75
Table 20 Effect of fermentation on lactic acid bacteria count of iron-fortified and non-fortified maize dough	76
Table 21 Effect of fermentation on yeast and mould count of iron-fortified and non-fortified maize dough	76
Table 22 ANOVA summary table of on pH on dehydrated and stored fermented maize dough samples	79
Table 23 ANOVA summary table for the full regression of the models and lack of fit (F-values)	82
Table 24 Age of last child of mothers	101
Table 25 Rank sums of sensory evaluation on fortified and non-fortified fermented maize dough porridges	104
Table 26 Rank sums of sensory evaluation on fortified and non-fortified fermented maize/cowpea dough porridges	104
Table 27 Rank sums of sensory evaluation on fortified and non-fortified roasted maize/cowpea/groundnut blend porridges	105
Table 28 Estimated cost of 250g per product by respondents	107

LIST OF FIGURES

Figure 1 Flow diagram of preparation of iron-fortified fermented maize dough	32
Figure 2 Flow diagram of preparation of iron-fortified roasted weaning blend	35
Figure 3 pH changes in maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation.	46
Figure 4 Changes in titratable acidity (g/100g) of maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation	49
Figure 5 pH changes in dehydrated maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation.	51
Figure 6 Changes in titratable acidity (g/100g) of dehydrated maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation	53
Figure 7 Changes in fat acidity (mg KOH/100g) of ferrous sulphate (FS) fortified dehydrated maize dough before (A) and after (B) fermentation	56
Figure 8 Changes in fat acidity (mg KOH/100g) of ferrous fumarate (FF) fortified dehydrated maize dough before (A) and after (B) fermentation	57
Figure 9 Changes in water absorption in ferrous sulphate (FS) fortified dehydrated maize dough before fermentation at 30°C (A) and 70°C (B) and after fermentation at 30°C (C) and 70°C (D)	63
Figure 10 Changes in water absorption in ferrous fumarate (FF) fortified dehydrated maize dough before fermentation at 30°C (A) and 70°C (B) and after fermentation at 30°C (C) and 70°C (D)	64
Figure 11 Hot paste viscosity changes in dehydrated maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation	67
Figure 12 Cooled paste viscosity changes in dehydrated maize dough fortified with ferrous	

sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation	69
Figure 13 Response surface plot for colour at 175°C roasting temperature for ferrous fumarate roasted blend	84
Figure 14 Response surface plot for fat acidity at 175°C roasting temperature for ferrous sulphate roasted blend	86
Figure 15 Response surface plot for water absorption at 30°C at 175°C roasting temperature for ferrous sulphate roasted blend..	88
Figure 16 Response surface plot for water absorption at 70°C at 175°C roasting temperature for ferrous sulphate roasted blend.	89
Figure 17 Response surface plot for apparent hot paste viscosity at 175°C roasting temperature for ferrous fumarate roasted blend.	91
Figure 18 Response surface plot for apparent cooled paste viscosity at 175°C roasting temperature.	92
Figure 19 Response surface plot for energy content at 175°C roasting temperature for ferrous fumarate roasted blend	94
Figure 20 Response surface plot for total iron content at 175°C roasting temperature	95
Figure 21 Response surface plot for fat acidity at 175°C roasting temperature for ferrous fumarate roasted blend at 4 months storage	97
Figure 22 Response surface plot for water absorption at 30°C at 175°C roasting temperature at 4 months storage	98
Figure 23 Response surface plot for water absorption at 70°C at 175°C roasting temperature at 4 months storage	99
Figure 24 The profile of respondents: Educational background (A), age group distribution (B) and primary occupation (C)	102

1.0 INTRODUCTION

1.1 Malnutrition

Malnutrition is usually the result of a combination of inadequate dietary intake and infection. In children, malnutrition is synonymous with growth failure - malnourished children are shorter and lighter than they should be for their age. Although many people still refer to growth failure as protein-energy malnutrition or PEM, it is now recognised that poor growth in children results not only from a deficiency of protein and energy but also from an inadequate intake of vital minerals (such as iron, zinc and iodine) and vitamins such as vitamin A and other essential fatty acids as well (UNICEF, 1998).

1.2 Protein-Energy Malnutrition (PEM)

Protein-energy malnutrition (PEM) affects about 500 million people globally (Lachance and Bauernfeind, 1991). More severe forms are characterised by retarded growth, apathy, loss of appetite and fluid accumulation (kwashiorkor) due to severe protein deficiency or by retarded growth and extreme thinness (marasmus) caused by deficiency of both calories and protein, or by a mixture of the two states (Marasmic-Kwashiorkor). PEM exists in many developing countries where parasitic infections and diarrhoea are complicating factors. In Ghana, 28% of children under five of age are underweight. It is estimated that about 45% of all child deaths beyond early infancy are due to protein-energy malnutrition, making this the single greatest cause of child mortality (GDHS, 1993).

Much attention has been focused on protein-energy malnutrition on account of its

widespread occurrence. However, other nutrient deficiencies such as vitamin A, iron and iodine are becoming prevalent owing to the heavy dependence on cereals and root crops among the poorer segments in developing countries.

1.3 **Micronutrient Malnutrition (MM)**

Minerals are needed in quantities, in the order of a few thousandths of a gram or less each day. They are consequently called micronutrients, which are needed for the production of enzymes, hormones and other substances that are required to regulate biological processes leading to growth, activity, development and the functioning of the immune and reproductive systems.

All the minerals that the body needs, for example, calcium, phosphorus, iron, zinc, iodine, sodium, potassium and magnesium have to come from the food we eat or from supplements. The body manufactures many of the complex organic molecules it needs from simpler building blocks but the vitamins - A, B complex, C etc. are not synthesised.

Micronutrients are needed at all ages but the effects of inadequate intake are particularly serious during periods of rapid growth, pregnancy, early childhood and lactation (UNICEF, 1998). An estimated 2,000 million people are at risk of deficiencies of vitamin A, iodine and iron; most of them are women and children living in the less developed countries (Combs *et al.*, 1996). Problems caused by micronutrient malnutrition are thus, now recognised as causes of human suffering on a global scale. They constitute major causes of morbidity and mortality and can impair cognitive development and learning. It is not unreasonable, therefore, to view micronutrient malnutrition both as an outcome and a cause of underdevelopment.

Common micronutrient deficiencies in developing countries include Vitamin A deficiency (VAD), Iodine deficiency disorders (IDD) and Iron deficiency anaemia (IDA). Certain conditions demand specific types of interventions such as supplementation, food fortification, promotion of breastfeeding, dietary modification through nutrition education, promotion of home gardens and disease reduction) (Bouis and Slack, 1996).

1.3.1 Vitamin A Deficiency (VAD)

Vitamin A deficiency is a leading cause of childhood blindness (xerophthalmia). It is estimated that more than 250 million children worldwide are at risk of vitamin A deficiency. This is believed to cause blindness in 250,000 - 500,000 children each year; two-thirds of these children die within months of going blind due to their increased susceptibility to infections (Combs *et al.*, 1996). Most nutritionists now agree that if an appropriate food carrier exists, to which vitamin A can be added, then fortification is the cheapest and most effective means of controlling xerophthalmia in most countries (Lachance and Bauernfeind, 1991).

Baseline data from two vitamin A supplementation trials conducted in 1989-91 in the savannah area of Ghana found the prevalence of clinical VAD to range from 0.7% to 1.5%, indicating that VAD was a problem of public health significance in some regions of the country. Moreover, low serum retinol (< 20µg/dl) was found in 65-70% of the children studied, reflecting high levels of subclinical VAD in the population (VAST, 1993). An average of 37.2% studied in the southern zone had severe VAD (below 10µg/dl for serum retinol) (MOH, 1998).

1.3.2 Iodine Deficiency Disorders (IDD)

Endemic goitre and cretinism (dwarfism), a result of iodine deficiency disorder (IDD), is influenced primarily by a lack of sufficient iodine in the inland soils in which food crops are grown. Iodine deficiency is prevalent in most regions of the world; an estimated 1,600 million people live in iodine-deficient areas. The most prevalent outcomes of iodine deficiency are reduced mental function, increased rates of still birth, abortions and infant deaths (in as much as 1% of pregnancies in deficient areas). Some 6 million infants born annually to iodine-deficient mothers develop severe mental and neurological impairment known as cretinism. An additional 20 million people are estimated to have mental retardation as a result of iodine deficiency (Combs *et al* 1996). Ghana has serious IDD problem, affecting, at least, 33% of the districts (Asibey-Berko and Orraca-Tetteh, 1995). In industrialised countries, control of IDD has generally been achieved through mandatory or voluntary iodisation of salt, a cheap and simple delivery system. This fortification has caught up with the developing countries.

1.3.3 Iron Deficiency Anaemia (IDA)

Iron deficiency is the most prevalent micronutrient deficiency, affecting over 2,100 million people, particularly women of reproductive age and pre-school children living in tropical areas. It also affects school-aged children and working men in many areas. Iron deficiency leads to several outcomes that compromise function, these include, anaemia, reduced work capacity, impaired learning ability, increased susceptibility to infections and increased risk of death associated with pregnancy and childbirth (Combs *et al.*, 1996). It is estimated that one-fifth of maternal mortality is due to the direct (heart failure) or indirect

(inability to tolerate hemorrhage) effects of anaemia. Severe anaemia is responsible for nearly one-third of fatalities among children who are not given immediate transfusions. While anaemia can have multiple causes (including malaria, intestinal parasitism, deficiencies of folate and vitamin B₁₂), it is thought that at least half of the anaemia worldwide is due to iron deficiency. Data from a national survey conducted in 1995 indicated that 83.5% of children under five years of age, 71% of school aged children, 65% of pregnant women and 58% lactating women were anaemic (MOH, 1998). This is similar to work done in other developing countries reported by Bothwell *et al.*, (1979) and Simmons and Gurney (1980).

1.4 Strategies to combat infant malnutrition

Approaches to control or prevent PEM include increased dietary intake of cereal-legume foods, more oil and fat in diet, protein or amino acid supplemental addition to the low protein foods, a greater effort on immunisation, parasitic disease control and oral dehydration treatment for diarrhoea (Lachance and Bauernfeind 1991).

Work in Ghana and elsewhere has shown that the protein quantity and quality of cereal gruels can be increased through fortification with legumes (Orraca Tetteh, 1972; Eyeson and Plahar, 1998; Mensah and Sefa-Dedeh, 1991; Osei and Sefa-Dedeh, 1993; Akpapunam and Sefa-Dedeh, 1995 and Nout, 1992).

Nout (1992) observed that the energy densities of traditional cereal foods are inadequate, particularly, when used for weaning. The protein quality and quantity are low due to limiting amino acids e.g. lysine; and the high bulk of the product causes gelatinisation of the large quantities of starch in the cereal products.

In Ghana, efforts have been made to use cowpea to develop high protein foods (Osei

and Sefa Dedeh, 1993; Akpapunam and Sefa-Dedeh, 1995). Blends of the cereals and legumes are encouraged in weaning food formulation to make use of locally available raw materials and to provide better nutritional value than that of cereal porridges (Sefa-Dedeh *et al.*, 1996).

Fermentation is another method used to improve quality of traditional weaning foods. It is one of the oldest methods of preparing and preserving food. It is intentionally carried out to enhance the taste, aroma, shelf life, texture, nutritional value and other properties of food (Motarjemi and Nout, 1996). Due to its anti-microbial effect in foods, fermentation is being used to afford microbial safety. For example lactic acid fermentation inhibits the survival and multiplication of a number of bacterial pathogens (Motarjemi and Nout, 1996). It reduces the contamination and proliferation of diarrhoeal pathogens in cereal porridge (Mensah *et al.*, 1988; Mensah 1990; Nout *et al.*, 1989).

Solid state fermentation of cereals is widely applied in the processing of cereals for the preparation of a wide variety of dishes (Obiri-Danso, 1994). In Ghana, the traditionally recommended weaning porridge is prepared from fermented maize dough.

1.5 Proposed solution of micronutrient malnutrition

For the last decades, more attention has been focused on the development of nutritious weaning foods to alleviate PEM in children (Roman *et al.*, 1987; Abbey and Nkanga, 1988; Valyasevi and Dhanamitta, 1987)). The joint FAO/WHO initiative to convene the International Conference on Nutrition (ICN) in 1992 recognised that micronutrient deficiencies constitute major public health problems in many countries and also have a serious impact on overall human well being and economic performance. The conference stressed the need for sustainable measures and called for the development and implementation of long-

term food-based strategies to prevent these deficiencies (Lupiem, 1996).

Most infant cereal diets are relatively low in iron content and often cannot fulfill the infant's iron requirement during the rapid growth phase of the first life (Pizarro *et al.*, 1991). In recent years, addition of bioavailable iron to infant cereal has been shown to protect against iron deficiency when significant amounts are consumed daily (Walter *et al.*, 1993). Iron fortification of infant formula has been shown to significantly reduce the prevalence of iron deficiency in countries such as the United States (Yip, 1989).

Sweden has a high level of iron fortification, which contributes up to 40% of dietary iron intake (Hallberg, 1992). Analysis of the Swedish experience of iron fortification of flour since 1976 indicates that the practice has contributed to the significant decline of iron deficiency anaemia among women of child-bearing age, between 1965 to 1975 from 30% to 7% without a noticeable impact on the incidence of haemochromatosis (Hurrell, 1992).

Evaluating the information on nutritional iron deficiency gathered over the last 40 years has led most investigators to conclude that iron fortification of the diet is the only cost-effective long-term strategy for reducing the prevalence of iron deficiency in most developing countries (Cook and Reusser, 1993; MacPhail and Bothwell, 1992).

1.5.1 Technical challenges of mineral fortification

The addition of various bionutritional substances creates several technical challenges for the Food Technologists (Sloan and Stiedemann, 1996). It is necessary to maintain the overall quality of the product in terms of the bioavailability of the fortifying agent. The stability of the fortified product can affect the biological outcome and the acceptability, and the effects of processing, storage, distribution and preparation practices on them also need to be considered. Iron, for example, may react with fatty acids in the fortified food, forming free

radicals that induce oxidation, which may produce unacceptable flavours in the product. Other characteristics that may be affected are colour, taste, odour and appearance, alterations that should be avoided altogether since they affect consumer acceptability of the product (Meija, 1994).

Since minerals are added to food for the purpose of improving nutritive value, it is obvious that the major factor, which must be considered in choosing a mineral additive for a particular food, is that it should be bioavailable and fulfil its intended purpose. It does not make sense to use a low cost mineral additive that does not degrade food quality if it is absorbed and utilised by the body. However, the converse is also true in that it does not make sense to use an exceedingly bioavailable form of a mineral if the food is rendered unpalatable, due to quality losses.

Bioavailability of a mineral in food is not strictly a function of the bioavailability of the mineral salt or additive chosen. However, minerals are often chemically reactive and their bioavailability will often be greatly affected by interactions with food components when added, or during processing and storage. Such reactivity will of course vary with the mineral additive used, the food, the process, and storage times and conditions. This means that the bioavailability of a mineral in food is ultimately dependent upon its electronic configuration since this governs its physical and chemical properties, which in turn govern its behaviour in food. Interestingly, the electronic configuration of the elements whose physical and chemical properties (electronic structure) are similar will act antagonistically to each other biologically. Therefore, in order to make judgements about both the bioavailability and functionality, so that an appropriate mineral additive may be chosen, it is essential to understand the major physiochemical properties that affect potential bioavailability, and how these may be influenced by their food environment and/or processing and storage (Clydesdale, 1989).

1.6 Objectives

The main objective of the study is to investigate effects of iron fortification and fermentation on product characteristics of weaning foods.

Specific Aims

1. To study the effects of fermentation and drying on product characteristics of ferrous sulphate and ferrous fumarate fortified maize dough.
2. To evaluate the microbiological properties of ferrous sulphate and ferrous fumarate fortified fermented maize dough.
3. To investigate the effect of legume and iron concentration on the physical, chemical and functional properties of roasted weaning food.
4. To evaluate the shelf stability of iron fortified weaning foods.
5. To determine the consumer's expectation of the quality characteristics of weaning foods.

2.0 LITERATURE REVIEW

2.1 Weaning practices

Weaning is the process by which the infant gradually becomes accustomed to the full adult diet. Weaning begins when the child is introduced to foods other than breastmilk and is completed when breastfeeding is stopped and the child is fully accustomed to the family diet.

Breast milk is nutritious and generally safe food for newborn infants. Exclusive breastfeeding, that is, giving the infant no fluid or food other than breast milk, protects against diarrhoea by minimising the infant's exposure to food-borne and waterborne pathogens. However, when the infant reaches 4-6 months of age, breast milk needs to be supplemented and later on (less than 2 years of age) substituted by appropriate foods until the child takes up the family food. The weaning period is the most vulnerable stage in the life of a child particularly in developing countries. Improper weaning practices are not due to ignorance and taboos but mainly to inadequate supply of right types of foods particularly protein-rich foods, which are essential for growth. The weaning period is the most vulnerable stage in the life of a child particularly in developing countries. Improper weaning practices are not due to ignorance and taboos. The introduction of weaning foods, prepared under unhygienic conditions to infants may expose them to infective doses of food-borne pathogens, leading to diarrhoeal diseases, malnutrition and death (Motarjemi *et al.*, 1993).

2.2 Weaning food

Weaning foods are described as semi-solid or solid foods designed to be fed to an

infant either partially or fully weaned from the breast (Jansen, 1992). They bridge the gap between breastfeeding and an “adult” family diet. The weaning foods in most cases are watery cereal porridge, very low in energy and protein and not adequate to provide all the nutrients required by the rapidly growing child. The development of weaning foods is based on the nutritional requirement of children aged 6 - 12 months. The total food intake must supply 3500kJ of energy and 14g protein daily for a child weighing 7 kg (Hiel, 1984), as well as a variety of micronutrients (Renner, 1989). In addition, weaning foods must have an easy-to-swallow semi-liquid consistency (1,000-3,000 cP), and must be microbiologically safe when consumed. The level of antinutritional factors occurring naturally in the ingredients must be minimised by adequate processing. Ingredients must be locally available and acceptable. These would usually include a staple cereal such as maize, sorghum, millet and rice or starchy tuber, such as yam and cassava. The protein quantity and quality of the starchy staple can be optimised by adding legumes (beans, pulses). Low-cost processing techniques must be employed using simple equipment and energy-conserving operations.

2.2.1 Traditional weaning foods

The diet of most people in developing countries is based on processed cereal grains such as maize, sorghum and rice or roots and tubers such as yam and cassava (Bressani, 1985). These foods, because they are eaten in large quantities, provide the bulk of protein, but the quality of this protein leaves much to be desired. Traditional weaning porridges are usually made from these local staples and the result is gruels that have low nutritional values, as they are not adequate source of micro and macronutrients (Brown, 1991).

2.3 Cereal and legume processing technologies

In Ghana, cereal processing technologies may be divided into two; traditional or indigenous and imported technology (Sefa-Dedeh, 1989): The utilisation of maize for example, follows a variety of processing procedures, including soaking, dehulling, grinding, roasting, boiling, fermentation and germination (Sefa-Dedeh, 1993). These processing techniques are employed to eliminate anti-physiological constraints and improve nutritional value of the food.

2.3.1 Soaking

Soaking of whole maize grains in water before milling affects milling characteristics, reduces starch damage; affects fermentation and cooked paste viscosity of the fermented maize dough (Ackom-Quayson and Sefa-Dedeh, 1993). The soaking also initiates the fermentation process. Soaking before cooking has however been reported to decrease the nutritive value of cowpea (Bressani, 1985) probably because of the leaching out of soluble nutrients.

2.3.2 Roasting

Roasting or toasting can be best described as dry heating. During roasting, several volatile substances including hexanal are removed by evaporation. The removal of such 'grassy' smelling substances and the simultaneous onset of Maillard reactions give rise to an attractive flavour. Roasting has a slight viscosity-reducing effect on some cereals, but this is

negligible in cereal-pulse mixtures. It does not reduce the content of tannins, phytic acid and oligosaccharides. However, roasting is carried out commonly in industrial practice to inactivate the Kunitz type of proteic trypsin inhibitors, haemagglutinins (Bressani and Elias, 1993) and lectins (van der Poel, 1990). Excessive roasting lowers the protein quality of the product since the Maillard reaction reduces the amount of available amino acids including lysine (Hans *et al.*, 1988). For those reactions, the time temperature combinations during roasting should be kept under control. From a microbiological point of view, roasting reduces the number of vegetative cells of bacteria and fungi. However, it does not protect the product from proliferation of post-processing contamination.

2.3.3 Fermentation

Fermentation is defined as the biochemical modification of primary food products brought about by the action of microorganisms and their enzymes. The soaking of the grains in excess water allows the selection of desirable microorganisms, such as lactic acid bacteria, yeasts and moulds (Akinrele and Bassir, 1967). The activity of these microorganisms reduces pH and increases the titratable acidity of the substrates. A number of fatty acids are also produced (Akinrele, 1970; Muller, 1970; Plahar and Leung, 1982). The sour taste developed as a result of the acids produced is a desirable sensory attribute. This tangy taste has been reported to appeal to infants especially those who are ailing and unable to feed properly (Philips and Abbey, 1989).

In 1990, Mensah, studying the properties of fermented maize dough observed significant inhibition of pathogenic bacteria. Cooking the dough into porridge reduced the antimicrobial effect but there was still significant inhibition of pathogen. The anti-microbial

property could be important for the reduction of the high levels of faecal bacteria in weaning foods in developing countries. The increase of niacin and riboflavin contents (Cameron and Hofvander, 1971) and the release of phosphorous from phytate (Lopez *et al.*, 1983) are among the several benefits derived from the fermentation of maize.

Another important characteristic of fermentation is the textural changes that occur. Findings on the effect of fermentation on viscosity are highly contradictory and appear to vary depending on the method of fermentation and particularly the microorganisms responsible (Ashworth and Draper, 1992; Wanink *et al*, 1994). Solid-state fermentation has been found by some researchers to increase cooked paste viscosity of maize dough (Ampadu, 1991; Anim, 1991; Osa-Mensah, 1991; Okoree, 1993). However, the presence of some admixtures in fermented maize dough decreases the cooked paste viscosity. The lactic bacteria (*Lactobacillus plantarum*, *Lactobacillus fermenti*) have been reported to eliminate flatulence-causing oligosaccharides (Duszkiewicz- Reinhard *et al.*, 1994). Additions of some legume have also been reported to improve protein digestibility (Barampama and Simard, 1994).

Phytic acid is major inhibitor of iron in cereals and legumes and can be removed by enzyme treatment or fermentation. Analysis of fermented whole grain sorghum and the germination reduces the concentration of phytic acid to the extremely low levels needed for significant improvement in iron absorption (Hurrell, 1990).

2.4. Functional and Chemical Properties of Weaning Food

Specific functional characteristics are required for every food system (Pour - E.L, 1981). The functional property of a food is important because it indicates the utility of products in specific applications and therefore reflects the properties encountered by their use

during the preparation of usable products (Sefa-Dedeh, 1993).

The functionality of food depends on its chemical composition and properties. This is because food is made up of aggregates of biological molecules including fats, carbohydrates and proteins, which react constantly depending on their environment. It is this reaction and interaction between the food constituents that determines the functionality of that particular food system. The ultimate application of cereals and legumes is the conversion into edible forms for human consumption. The application of processing technologies, for example, temperature, enzymes, size reduction and hydration in weaning food production affects the chemical properties and therefore the functionality of the food. Viscosity and water absorption characteristics are important functional properties of cereal- based porridges. Starch, a major carbohydrate in cereals and legumes has the ability to absorb water when suspension of the granules is heated above gelatinisation temperature (Rasper, 1982). The swelling of the granules and their consequent disintegration on prolonged heating leads to significant changes in viscosity and other rheological properties of the paste that are characteristic of the particular type of starch. Since the effect of mixing different components is not simply addition, evaluation of the individual components is at best, only a partial predictive tool (Scheon, 1977). Therefore, in product formulation, the determination of the functional properties of components of the mixture and the end product are clearly of prime importance and must be measured.

2.5 Food Fortification

Depending upon the causes of malnutrition and the resources available, different approaches to the improvement of nutrition may be adopted. Food fortification is only one of

the measures affecting the quality of food supply that are worthy of consideration in the design of programmes intended to improve upon the health of the population. It is used as a method of maintaining the quality of foods consumed by the population. For example, nutrients may be added to foods to replace those lost in processing, they may be to “imitate foods” to provide nutritive value equivalent to the foods that they imitate or they may be added to ‘ready to eat’ meals or similar preparations. Nutrients may have to be added to food mixtures compounded for special dietary needs, e.g., infant foods, weaning foods or energy ration (FAO/WHO, 1971).

The principles of food fortification are:

1. To conform to current food standards
2. To replace nutrients to a level representative of those in the prior to storage, handling and processing
3. Avoid nutritional inferiority in a food that replaces a traditional food in the diet
4. And balance the vitamin, mineral and protein content of a food in proportion to its total caloric content.

2.6.1 Protein Fortification

An important approach to increasing nutrient density has been to broaden the number and type of ingredients used for weaning food formulation (Philips, 1991). Cereals and legumes especially maize and cowpea are widely grown in developing and industrialised countries of tropical and warm temperate regions where they are accepted as part of traditional diets (Dovlo *et al.*, 1976; Ali and Wills, 1983). Maize lacks some nutrients and its content of lysine, tryptophan, and niacin is low. The whole grain contains about 10% protein

and has sufficient sulphur amino acids to form complementary mixtures with legume proteins (Ihekoronye and Ngoddy, 1985). Cowpeas, one of the most important grains in diets in Africa are used extensively in weaning foods where they provide abundant and high quality protein. Legume seeds in general are a rich source of proteins, vitamins and minerals (Borejszo and Khan, 1992).

Due to the high cost of commercial weaning foods and animal protein, most developing countries are encouraging the production of high protein-energy weaning foods from locally available materials (Sefa- Dedeh, 1991). Cereals and legumes are both cheap and available and whilst each has its nutritional deficiencies, together they are complementary (Akinyele *et al.*, 1988).

2.6.2 Mineral Fortification

The use of mineral additives in food, like other nutrient additives, was used historically as a public health measure to prevent the occurrence of deficiency diseases among population (Clydesdale, 1991). Over the years, many groups have been involved in attempting to establish principles and policies for the fortification of foods. This is because food-based programmes offer it as an important sustainable strategy for preventing mild to moderate deficiencies of micronutrients. They can prevent deficiencies of several micronutrients simultaneously without the risk of antagonistic nutrient interactions (Gibson, 1996).

A food vehicle should be chemically suitable, not adversely affect the bioavailability of the fortificant(s), and its organoleptic (taste, smell, colour) and functional properties (stability and shelf life) should not be affected by fortification under usual conditions of storage and use.

Fortificant is the term used to refer to the chemical compound, which is added to a

food during the fortification process. Fortificants differ in their bioavailability, organoleptic, and functional properties, and different compounds are suited to different food vehicles. The levels of fortificant to be added to the food vehicle are important; the concentration of the fortificant(s) in the food must be adequate to deliver an effective dose or the fortification programme will be ineffective.

2.6.3 Iron fortification

The addition of iron to food over and above that naturally found in food can prevent iron deficiency. The challenge is to identify a widely consumed, centrally processed food appropriate to the age groups of concern. Fortification of complementary foods has been achieved in many countries at low cost, which allows for effective targeting of infants and encourages the development of partnerships between government and the food industry.

Fortification with iron presents a greater challenge than other micronutrients. Most fortificants are inorganic iron compounds whose bioavailability is extremely variable and is greatly affected not only by the chemical composition of their food vehicle, but also by that of the whole meal in which they are consumed. There are many factors which reduce the bioavailability of iron, the main ones being phytates, oxalates in cereals and pulses, polyphenols including tannins in tea and coffee, soy protein, and phosphatin in eggs. Factors, which enhance bioavailability, are organic acids, notably ascorbic acid or vitamin C, animal protein, a low pH as in fermented foods and alcohol. Ascorbic acid can be added to improve absorption, but this will increase costs and would be prohibitively expensive on a national scale (Cook and Reusser, 1983). It has been recommended that the bioavailability of fortificant iron in typical foods eaten as part of the normal diet should be part of a feasibility

study of fortification (Baynes and Bothwell, 1990). Table 1 shows, as an example, the addition of several iron salts to different processed foods.

Table 1 Relative bioavailability of iron compounds used in food fortification (%)

Compounds	Rats	Humans	Fortified foods
Soluble in water			
Ferrous sulphate	100	100	infant formulas
Ferrous gluconate	97	89	infant formulas
Slightly soluble in water and soluble in diluted acids			
Ferrous fumarate	95	100	infant cereals
Ferrous succinate	113	92	infant cereals
Insoluble in water but soluble in diluted acids			
Ferric pyrophosphate	45	21-74	infant cereals
Ferric orthophosphate	6-46	25-31	infant cereals
Elemental iron	8-76	5-90	infant cereals

Source: Hurrell, 1992

Unfortunately, the most bioavailable soluble compounds, such as ferrous sulphate, also tend to be the most reactive and can catalyse oxidative reactions causing smells and colours (Cook and Reusser, 1983). This reduces the shelf life of the fortified products. There is an inverse association between shelf life and stability. Insoluble phosphate compounds cause fewer organoleptic and storage problems but have a low bioavailability.

Sodium iron ethylenediaminetetra-acetic acid (NaFeEDTA) and organic haemoglobin

are iron complexes whose bioavailability is good and relatively unaffected by other food components. Ethylenediaminetetra-acetic acid (EDTA) is an approved food additive and iron EDTA was given provisional approval by the Joint FAO/WHO expert Committee on Food Additives (JECFA) in 1993 for use in food fortification programmes.

Bovine haemoglobin has been used in Chile and Brazil (Nogueira *et al.*, 1992; Walter *et al.*, 1993), but requires a constant supply of uncontaminated blood, imparts a strong colour to foods, and would be culturally unacceptable in some countries. It also carries potential health risks and its use would require a rigorous quality control system. Encapsulated fortificants have been developed which are relatively immune to heat-, moisture-, and oxygen-induced degradation and so survive the effects of downstream processing and storage well, but these are much more costly than unencapsulated forms (Maxwell, 1990).

An alternative to chemical fortificants is a food-based approach using locally available and culturally acceptable micronutrients-rich foods to improve the micronutrient content of the foods. Trowbridge *et al.*, (1993) recommended food-based approaches as a feasible and cost-effective strategy to prevent malnutrition.

Fermentation and germination, which are traditional food technologies in many countries, have been suggested as a means to improve iron availability (Svanberg *et al.*, 1993)

2.6.3.1 **Level of Iron Fortification**

Numerous factors influence the recommended level of fortification hence there is no universal specification for level of fortification of the chosen vehicle. The main determinants of the fortification level are:

Recommended dietary intake (RDI) of the micronutrient

- Prevalence of micronutrient deficiency
- Per caput consumption of the food vehicle
- Extent of processing, transit, storage and food preparation losses
- Current dietary habits of the population in question with regard to food selection and preparation; and
- Other dietary ingredients affecting its absorption and bioavailability (Lofti *et al.*, 1996). The RDI of iron varies by age and physiological group and depends on the iron availability in the diet (Table 2)

Table 2 Recommended dietary intake (RDI) for iron (mg/day)

Iron availability in diet	Low (5%)	Medium (10%)	High (15%)
Adults			
Men (>16 years)	23	11	8
Women (menstruating)	48	24	16
Women (postmenopausal)	19	9	6
Women (lactating)	26	13	9
Children (age in years)			
0.25-1	21	11	7
1-2	12	6	4
2-6	14	7	5
6-12	23	12	8
12-16 (boys)	36	18	12
12-16 (girls)	40	20	13

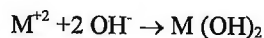
Source FAO/WHO (1988)

2.6.4 **Effect of iron fortification on food quality**

Successful fortification of a food is dependent upon the fortificant acting in a relatively benign manner in the food. Unfortunately this level of chemical inertness is often associated with insolubility and relatively poor bioavailability. The key to proper fortification is to deal with chemically reactive species, not inert compounds as the minerals are often thought. This reactivity becomes particularly apparent in a food in the presence of moisture and for adequate water activity when reactions may occur with free radicals, other food components, and oxygen or diffused packaging materials. Any or all of these reactions could result in undesirable quality changes and consideration must be given to such an occurrence. Ideally, mineral fortification should cause no change in colour, flavour, methods of preparation, appearance or catalyse any other undesirable changes in the food if it is to be used successfully in a general fortification programme (Clydesdale, 1991). This is a difficult ideal to achieve and some of the potential problems are discussed below:

2.6.4.1 **Reduction potential and pH**

The solubility product (K_{sp}) for a mineral (M) in the presence of hydroxyl ions (OH) may be represented by the following equation:



$$K_{sp} = [M^{+2}] [OH^-]^2$$

Since K_{sp} is constant, an increase in pH [OH^{-1}], causes a decrease in the free metal ion [M^{+2}] which follows first, second, and third powers of hydroxyl ion concentration with the mono, di, trivalent form of the metal respectively (Chaberek and Martell, 1959). It is therefore evident

that metal ion will decrease in solubility as the pH is increased. Wien and Schwartz (1985) gave a dramatic example of this. They reported that a pH increase of 0.9 units, from pH 6.2 to 7.1, caused a 40% drop in calcium solubility in three muffin formulations.

Another equally important reaction occurs with transition metal solubility as pH increases. The transition metals, including these nutritionally important ones such as Fe, Ca, Ni, Cu and Zn, exist in solution as hydrates rather than ions. As the pH is raised the hydrates lose protons and form the less soluble or insoluble hydroxides. Iron is the best known example of this phenomenon, existing in solution at low pH as $\text{Fe}(\text{OH})_2$ form the solubilities of 10^{-16}M and 10^{-1}M , respectively. Although neither of these is very soluble, it is clear that Fe^{+2} is much more soluble than Fe^{+3} and explains the greater bioavailability of Fe^{+2} . The difference in solubility between ions of different valence states is why the reduction potential of the medium is important. In the above case, an environment, which promotes reduction, will keep the Fe^{+2} form predominating and increase bioavailability. This is one of the reasons why vitamin C, which is a reducing agent, greatly enhances the bioavailability of iron. This means that acidic foods and those that contain vitamin C may be a logical point to consider fortification with iron or other minerals. Control of pH at appropriate points during processing may be simple yet it plays an effective role in increasing the final bioavailability of minerals in question.

2.6.4.2 **Colour**

Colour is one of the most obvious undesirable changes that minerals can cause to food. Some colour changes are due to the reactivity of the mineral used while others may be due to the amount of mineral used. Small amounts of minerals such as iron if added in a reactive form such as ferrous sulphate, can react with phenolics to form blue-black colours

and darken food products containing large amounts of tannins such as chocolate and barley flour (Waddel, 1973). Iron may cause colour problems in milk (Coccodrilli and Shah, 1985) and soy isolate infant formula (Purvis, 1985). Problems may also occur in powdered products and dry blends when liquefied. The Committee of Food Protection Food and Nutrition (C.F.P.) in 1975 have reported this, in a vanilla-flavoured product fortified with ferrous citrate. Colour changes can also be a problem with pale foods, such as rice, and in foods, which are eaten, in small amounts, such as sugar and salt, where fortificant levels are relatively concentrated. Coloured foods with strong flavours are more suited to fortification with reactive iron compounds (Draper, 1996). The reactivity and/or solubility of the mineral are critical.

Many fruit-based beverages are of low pH and as a result will tend to solubilise most mineral salts to increase their reactivity and potential for bioavailability, thus establishing the inverse relationship between functionality and bioavailability. Changing the fortificant source or adding it to the product at a different stage in the process may eliminate some of these colour problems. If iron is dry-blended into the finished powdered product, the colour may be better than if added during the liquid stage of processing due to minimisation of reactivity in the dry state (Clydesdale, 1991).

2.6.4.3 **Flavour**

The addition or presence of minerals in food can also cause undesirable flavour changes. The amount of mineral necessary to achieve nutritional significance has a direct bearing on flavour. The major flavour problem, however, may arise in foods, which contain lipids. Many minerals, particularly the transition metals possessing two or more valence states

with a suitable oxidation-reduction potential between them such as cobalt, copper, iron, manganese and nickel are major pro-oxidants. Even at levels as low as 0.1 ppm they can increase the rate of oxidation under certain circumstances, (Nawar, 1985) and cause unacceptable off-flavours.

Of all the minerals, iron is the one most involved with the formation of off-flavours in lipid-containing foods. It forms complexes and free radicals with oxygen that are energised sufficiently to remove hydrogen from the fatty acid chains and form free radical derivatives of fatty acids (Anderson, 1985).

It has been shown with physiological and non-physiological model systems that oxidation of lipids is greatly facilitated by the presence of iron whether the lipids peroxidation is dependent on the superoxide radical (Tien *et al.*, 1981) or the hydroxyl radical (Tien *et al.*, 1982). It is also apparent that different forms of iron may act as lipids peroxidation initiators. Ferrous, ferric and EDTA chelated forms in some systems may initiate the peroxidation reaction or the ADP-Fe₂-O₂ (perferryl) complex may be the initiator (Tien *et al.*, 1982). Interestingly, ferrous sulphate can be used in pasta without adversely affecting its colour, shelf life or flavour (Barrett and Ranum, 1985). Both ready-to-eat breakfast cereals and flours from wheat or corn are shelf-stable for the storage time required (Anderson, 1985).

2.6.4.4 **Energy**

On the basis of measurements taken in Western countries, dietary iron intake is closely related to energy intake (approximately 6mg Fe/1000kcal; Bothwell *et al.*, 1979). Relative differences between iron and caloric requirements in men, women, and children are reflected by significant variation in the percentage of dietary iron that must be absorbed to meet physiological needs (Table 3). Adult men, who have the highest caloric intakes and lowest

iron requirements, are least likely to suffer from iron deficiency, because absorption of as little as 5% of iron ingested may satisfy their needs. Infants in the post weaning period require fewer calories but almost as much iron. Women with high physiological losses require more iron but fewer calories.

Table 3 Relationship Between iron Requirement and Dietary Iron Supply

	Young children	Adults	
	6-18 Months	Male	Female
Mean weight (kg)	10	70.0	60.0
Food intake (kcal)	1120	3000	2300
Iron intake (mg)	6.7	19.2	13.8
Iron requirement (mg)	0.9	1.0	1.4 (2.2)
Absorption requirement (%)	13	5.0	10.0 (16.0)

Values in parentheses are for women with monthly menstrual blood loss exceeding 80 ml (10% of women)

Source: *Encyclopaedia of Food Science, Food Technology and Nutrition*

2.6.4.5 Effects of processing

Processing of food has been essential to convert the foods into relatively stable, convenient, packaged products subject to wide distribution and storage environments for year round availability in the feeding of humankind. To reach this objective, there occurs an inevitable loss of some nutrients, depending on the processing methodology.

The effects of processing on mineral bioavailability involve the form of mineral or mineral salt added to the food. Minerals are not heat sensitive, but their bioavailability may be altered as a result of chemical interactions within the food.

When fortifying with iron it is appropriate to utilize some form of elemental iron when the food is in dry form and undergoes storage. It has been shown that the bioavailability of elemental iron powders increases with decreasing particle size and surface volume, within limits (Patrick, 1985). It has also been shown that in a dry fruit-flavoured beverage mix fortified with elemental iron that 30% of the iron was converted to the ferrous form upon rehydration and 90% after standing overnight (Lee and Clydesdale, 1980). The solubility of both elemental iron and other sources can be affected by food process such as drying (Lee and Clydesdale, 1980), thermal processing (Lee and Clydesdale, 1981), and baking (Lee and Clydesdale, 1980). However, one study by Kivisto, *et al.*, (1986) found that extrusion had no effect on the apparent absorption of iron. This may be due to the decreased digestion of phytate found in the extruded bran products.

2.6.4.6 **Other quality changes**

Particle size is an important quality factor, since elemental iron if added to granular materials such as farina or semolina might be removed along with the dust when run through a purifier or any similar device that uses air separation. This problem can be prevented by addition of reduced iron at the end of the milling process. Elemental iron in ready-to-eat cereals can also cause physical problems, since it can gather on magnets that are in the cereal processing lines to remove tramp metal. Rearrangement of the magnets, however, can minimize this problem (Anderson, 1985).

In protein-containing liquid products, care must be taken in the amount and rate at which minerals are added, since overload might cause protein precipitation and a grainy defect. Some of these functional problems may be addressed by using a combination of

insoluble and soluble forms of the minerals (C.F.P. 1975) but care should be taken in the way in which such a choice affect bioavailability.

The oxidative properties of certain minerals such as iron and copper may catalyse the destruction of vitamins A and C as well as promoting flavour changes through oxidative rancidity.

Lee and Clydesdale (1980) evaluated various iron forms in chemically leavened bread and found out that the baking generated insoluble iron independent of the iron source. This may have been due in part to the alkalinity provided by the baking soda used to leaven the bread but regardless of the cause it would render the iron non-reactive and therefore not likely to cause quality problem on storage. Unfortunately it would also reduce bioavailability.

2.6.4.7 Toxicity

The use of supplements and fortification procedures has raised some concern about the possibility of toxic effects from minerals. Increased iron fortification, in particular, generated much debate when it was proposed in the seventies. For those minerals with large recommended daily allowance (RDA) such as calcium, phosphorous and magnesium there is little concern about toxicity from individual foods, which have been fortified since excessively high levels would cause quality problems such as sandiness or chalkiness. It was difficult to arrive at a measure of “safe intake” for any compound including the nutrients. However, Hathcock (1985) has proposed an interesting technique, which was used to arrive at a “mineral safety index” (MSI). This technique involved the use of a “recommended intake” (RI), which is the highest recommended daily allowance (RDA) value for an adult, except those for pregnancy and lactation, or the U.S. RDA, whichever is higher, and the “minimum toxic dose” (MTD) estimated from the literature. From these values, the mineral safety index

is calculated as:

$$MSI = MTD/RI$$

The MSI is thus defined in easily understood terms, and the result is a ratio that is independent of units as long as they are the same for the RI and the MTD.

The MSI for the three micronutrients of concern are shown in Table 4.

Table 4 Mineral Safety Indexes

Micronutrient	Recommended Adult Intake (RI)	Minimum toxic dose (MTD)	Safety index
Iodine	0.15mg	2mg	13
Iron	18mg	100mg	5.5
Vitamin A	5000IU	10,000-12,000IU	2-2.4

Source Hathcock (1985)

An individual's iron status regulates iron absorption to some extent (Gavin *et al.*, 1994). As the iron store increases, the bioavailability and, thus, the absorption of the dietary iron, decreases. Furthermore, the losses of iron from the body will increase because with increased iron stores, desquamated cells will have a higher iron content (Hallberg *et al.*, 1993). For this reason, there are no reports of iron toxicity in healthy subjects from iron-containing foods.

3.0 MATERIALS AND METHODS

3.1 MATERIALS

Maize (*Zea mays*) var "Obatanpa" and cowpea (*Vigna unguiculata*) var "Ayiyi" were obtained from Ejura Farms in Ghana. Groundnut was obtained from Mallam Atta market in Accra - Ghana. The iron compounds used to fortify the products were ferrous sulphate (dried FCC 111) and ferrous fumarate powder FCC 11. These were supplied by Dr. Paul Lohman GmbH KG in Germany.

3.2 METHODS

3.2.1 Calculation of iron added to food samples

The amount of iron added was based on the amount of a cereal- legume "weanimix" consumed by a Ghanaian child (Quarshie, 1988) and the recommended dietary intake (RDI) for iron for 5% iron bioavailability for cereal/legume foods in Table 2.

Sample calculation is shown in Appendix 1. The maximum amount of iron to be added was found to be 32mg/100g. Half of this amount was also added as the lower fortification to compare with the maximum level of fortification.

3.2.2 Effects of fermentation and drying on iron-fortified maize dough.

3.2.2.1 Sample Preparation

The maize was soaked in water for 24 hours, washed, drained and milled using the disc attrition mill (Agro Grinding mill, No.2A, India) to obtain a fine meal. The meal was

kneaded with water (50% moisture content) to form smooth dough. The maize dough was thoroughly mixed with ferrous sulphate and ferrous fumarate using Kenwood mixer (Model, PO9 2NH, Britain). The mixing of the maize dough with iron was done into two ways. The maize dough was divided into two parts:

Method I

Iron was added to the meal before mixing with water to form a 50% dough which was then fermented.

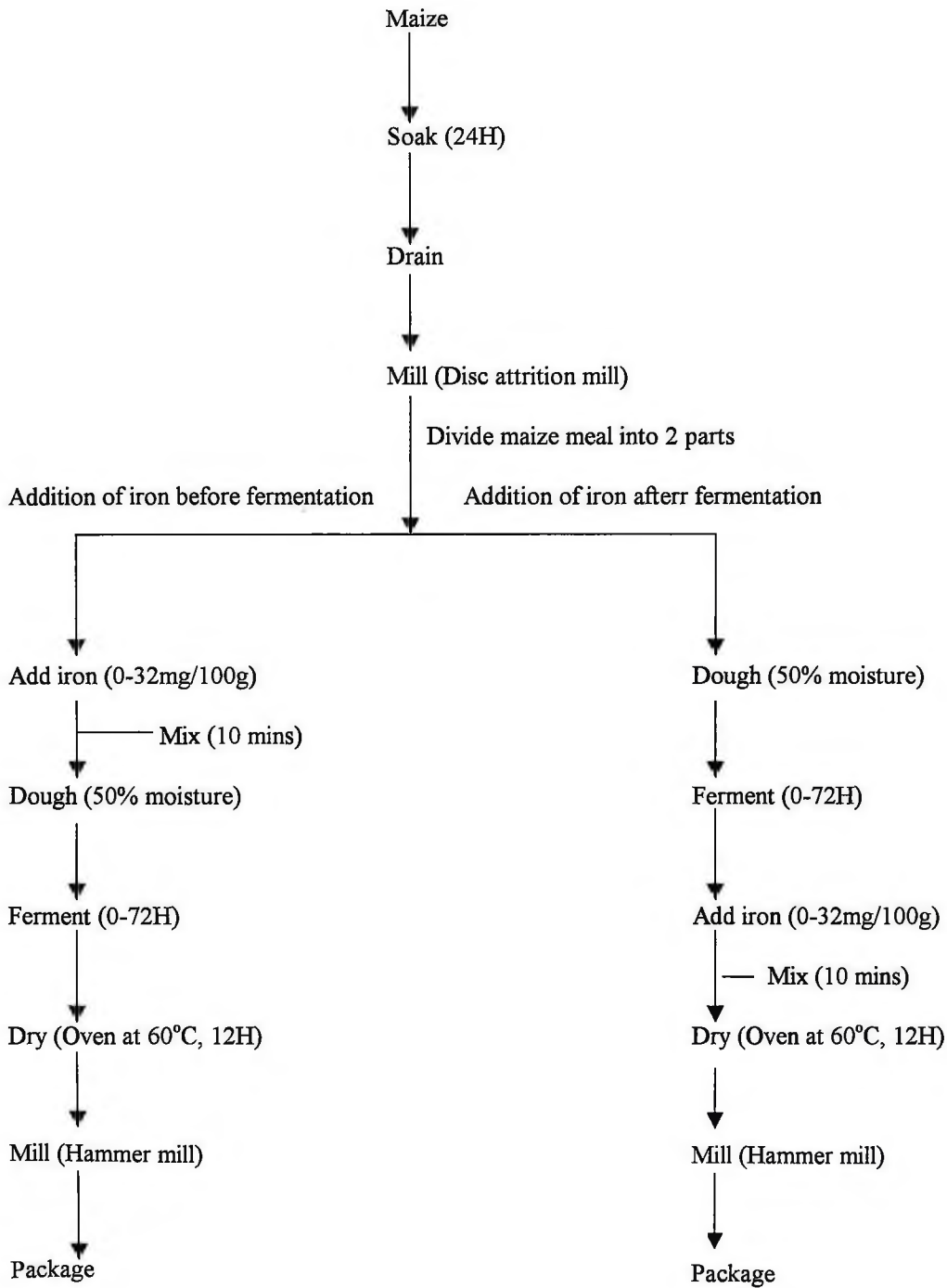
Method II

Dough was fermented then mixed with the iron supplement. Samples were dried in an oven at 60°C for 12 hours. These were milled into fine flour to pass through the No. 8 sieve using the hammer mill (Christy and Norris Ltd, Chelmsford, England). They were packaged in polythene and stored. The flow diagram of the sample preparation is shown in Figure 1. These were used in an experimental design as described in section 3.2.2.3

3.2.2.2 Analysis on fresh samples

Samples were fermented at room temperature for up to 72 hours and sub-samples collected at 12 hourly intervals. These were evaluated for pH and titratable acidity. Microbiological studies were also carried out on the samples in which iron has been added before fermentation.

Figure 1 Flow diagram of preparation of iron-fortified fermented maize dough



3.2.2.3 **Experimental Design**

Dehydrated fermented maize dough flour at fermentation time of 0, 36, 48 and 72 hours were used in 4x3x2x2 factorial design. The principal factors were:

<u>Variables</u>	<u>Levels</u>
Fermentation time (H)	0, 36, 48, 72
Iron fortification level (mg/100g)	0, 16, 32
Type of iron compounds	ferrous sulphate, ferrous fumarate
Mode of iron addition	before and after fermentation

These were evaluated for pH, titratable acidity, colour, fat acidity, water absorption, viscosity, energy and total iron determination. The data was subjected to analysis of variance and multiple range tests.

3.2.3 **Effect on storage on dehydrated iron-fortified fermented maize dough flour.**

Samples for the experimental design were stored at room temperatures (28°C) and were analysed after 2 months and 4 months. These were evaluated for pH, titratable acidity, colour, fat acidity and water absorption.

3.2.4 **Optimisation of concentration of legume and iron in roasting weaning blend.**

3.2.4.1 **Sample Preparation**

Cowpea and groundnut were roasted separately at 140°C for 15 minutes. The roasted groundnuts were dehulled by hand and winnowed. These were mixed in a proportion of 1:1 to make a legume concentration. Maize was roasted between 150°C - 200°C for 15 minutes. The

roasted maize was mixed with the legume blend. The legume blend was made up to 40% of the mixture. The mixture was milled into flour using the disc attrition mill (Agro Grinding Mill, No. 2A, India). The flour was allowed to cool and then mixed thoroughly with ferrous sulphate and ferrous fumarate (0, 16 and 32mg/100g) using Kenwood mixer. The samples were packaged in polyethylene bags. Flow diagram of the sample preparation is shown in Figure 2.

The samples were then evaluated for colour, fat acidity, water absorption, viscosity, energy and total iron.

3.2.4.2 Experimental Design and Statistical Analysis

The Central Composite Rotatable Design for $k = 3$ (Cochran and Cox, 1957) was used with the following independent variables; legume concentration, iron concentration and roasting temperature for maize. The levels of the variables used are summarised in Table 5.

Table 5 Process variables used in the Central Composite Rotatable Design (K = 3)

Independent variables	Variable levels				
	-1.682	-1.0	0	1.0	1.682
Legume concentration	0	8.1	20.0	31.9	40.0
Iron concentration	0	6.5	16.0	25.5	32.0
Roasting temperature	150	160	175	190	200

Figure 2 Flow diagram of preparation of iron-fortified roasted weaning blend

8 parts

One part

One part

Maize

Cowpea

Groundnut

Roast (150°C-200°C, 15 mins)

Roast (150°C-200°C, 15 mins)

Roast (150°C-200°
15 mins)

Mix

Mill (Disc attrition mill)

Flour mixture allow to cool

Add iron 0-32mg/100g

Mix (10 mins)

Package

Twenty sample combinations were generated in the design matrix (Table 6)

Table 6 Design matrix and variable combinations in experimental runs

No.	Level codes			levels		
	Legume	Iron	Temperature °C	Legume	Iron	Temperature °C
1.	-1	-1	-1	8	7	160
2.	-1	1	1	8	26	190
3.	1	-1	1	32	7	190
4.	1	1	-1	32	26	160
5.	0	0	0	20	16	175
6.	0	0	0	20	16	175
7.	-1	-1	1	8	7	190
8.	-1	1	-1	8	26	160
9.	1	-1	-1	32	7	160
10.	1	1	1	32	26	190
11.	0	0	0	20	16	175
12.	0	0	0	20	16	175
13.	1.682	0	0	40	16	175
14.	-1.682	0	0	0	16	175
15.	0	1.682	0	20	32	175
16.	0	-1.682	0	20	0	175
17.	0	0	1.682	20	16	200
18.	0	0	-1.682	20	16	150
19.	0	0	0	20	16	175
20.	0	0	0	20	16	175

The data were subjected to multiple regression analysis with the following dependent variables; fat acidity, colour, water absorption, total iron content, energy content and viscosity

(8% slurry concentration). Regression models were developed from Statgraphics software (Statgraphics, STSC Inc. Version 4.2 USA) using the stepwise regression technique. Models developed for each index were examined for lack of fit and three-dimensional response surface plots generated.

3.2.5 Effect of storage on iron- fortified roasted weaning foods.

The iron-fortified roasted weaning foods were stored at room temperature (28°C) for 4 months and evaluated for colour, fat acidity and water absorption.

3.2.6 Laboratory Studies

3.2.6.1 Colour of food samples

The colours of food samples were measured using Minolta Meter (CR - 310 Japan) with a 8mm wide diameter measuring area using diffuse illumination and a 0° degree-viewing angle. The instrument was standardised with standard white tile ($Y = 94.8$ $x = 0.3128$ $y = 0.3192$) under 'C' illuminant condition according to the CIE (Commission Internationale de l'Eclairage) were minimised by reporting an average of 3 readings per ground sample from each treatment and replication. This was in the L, a, b mode: "L" lightness, 0 = black to 100 = white, "a", red (positive reading) to green (negative reading), "b", yellow (positive reading) to blue (negative reading). L, a, b axes were defined as orthogonal axis and thus were physically independent.

Physico-chemical properties

3.2.6.2 pH and Titratable Acidity

(i) pH

AACC (1983) Method No. 02 - 31 was used.

Ten grams of sample (dry powder) was mixed with 10ml of distilled water and allowed to stand for 30 minutes with intermittent shaking. The supernatant was collected and its pH measured (TOA HMS - 305).

(ii) Titratable Acidity

For titratable acidity measurement, 17.6ml of the aliquot were pipetted into a conical flask and the pipette rinsed with 17.6ml distilled water was added to the aliquot. This was titrated against 0.1N NaOH to phenolphthalein end point. Titratable acidity was expressed as gram lactic acid per 100g of dry sample. All determinations were performed in duplicate.

Chemical Properties

3.2.6.3 Fat acidity

AACC (1983) Method No. 02 - 03A was used with some modification.

Ten grams well-mixed ground sample was weighed and transferred into an extraction flask. Fifty millilitres mixture of chloroform: methanol (2:1 v/v) was added and a stopper inserted. The mixture was shaken for 30 minutes using the Griffin flask shaker (Griffin and George Ltd, Great Britain). The meal was allowed to settle for about 3 minutes after the shaking and the liquid carefully decanted into a folded whatman No.1 filter paper inserted in a glass funnel. The funnel was covered with a petri dish to minimise evaporation. Ten millilitres of the filtrate was transferred into a 250ml Erlenmeyer flask and 10ml alcohol phenolphthalein solution

(prepared with 0.4g phenolphthalein in one litre 95% ethyl alcohol) was added. This was titrated with 0.0178N KOH. The determination was done in duplicate.

A blank was prepared by adding 10ml of alcohol phenolphthalein solution to 10ml chloroform:methanol mixture (2:1 v/v) and titrating with 0.0178N KOH.

Fat acidity was reported as mg KOH required neutralising free fatty acids from 100g samples on dry matter basis.

3.2.6.4 Total iron determination

Samples were wet digested with concentrated acid and 70% perchloric acid and the total iron was determined using atomic absorption spectrophotometer (AAS).

A Perkin - Elmer Model 3110 USA and Shimadzu, Japan with hollow cathode lamps and an air - acetylene flame were used for determining total iron of the samples. The specific standard operation conditions for the determination of iron is described in the AOAC 13th edition as follows: current 15mA; slit width 0.15nm; wavelength 248.3nm, Range 2.20ug/ml; Air pressure 81/min; Acetylene pressure 2.71 litres/min.

Six solutions with standard iron concentrations, which were read to obtain a calibration curve, were prepared from a standard stock solution of iron (Iron (111) nitrate 17.9mmol l⁻¹).

These were aspirated through the nebulizer of the atomic absorption spectrophotometer and the readings recorded. Each sample was then aspirated and the reading recorded. Two determinations were made for each sample and a standard sample was read between each test sample to ensure the proper calibration of the instrument. The burner was flushed with water between samples to re-establish a zero absorption point before each reading. The concentrations of iron in the samples were read from a calibration curve of absorption against

concentration.

3.2.6.5 **Energy**

The energy values of the samples were determined using the Ballistic Bomb Gallenkamp Calorimeter (Model CBB - 330, England)

Functional Properties

3.2.6.6 **Water absorption**

Five grams of sample were mixed thoroughly with 30ml of distilled water in a centrifuge tube and allowed to stand for 30 minutes at room temperature (30°C). The mixture was then centrifuged (Denley Model/BS 400) at 3000 rpm for 20 minutes. The supernatant was decanted and the sample weighed. The water retained in the sample was reported as water absorbed per 100g of sample on dry matter basis. Duplicate analysis was done. The measurement was repeated at 70°C.

3.2.6.7 **Apparent Hot and Cooled Paste Viscosity**

The Brookfield Digital Viscometer (Model Dv - L+ version 2.0, Brookfield Engineering Laboratories, Inc., Stoughton, MA, USA) equipped with a number 1 disc spindle and spindle speed of 5rpm at 6% solids was used for the fermented maize dough flour. A spindle number 3 and spindle speed of 5rpm at 8% was used for the roasted maize/groundnut/cowpea blend. The flours were mixed with water and the slurry was heated slowly on a hot plate until the temperature was 90°C. The viscosities were measured at this

temperature as hot paste viscosity. The sample was allowed to cool to 50°C and cooled paste viscosity determined.

3.2.7 **Microbiological analyses**

For all the samples, 10g were added to 90ml sterile diluent containing 0.1% peptone, 0.8% NaCl, with pH adjusted to 7.2 and homogenised in a stomacher (Lab blender, Model 80 Seward Medical, England) for 30 seconds at " normal" speed. From appropriate ten-fold dilutions; enumeration of aerobic mesophiles was carried out on Plate Count Agar (PCA, Merck, 5463, Darmstadt, Germany) incubated at 37°C for 48 hours. Lactic acid bacteria were enumerated on MRS (Merck, 10660, Darmstadt, Germany) incubated at 37°C for 72 hours. The pH of the sample solution was brought down to 5.5 before use. The mould and yeast counts were enumerated on PDA (Difco USA) incubated at 30°C for 72 hours. All determinations were duplicated.

3.2.8 **Sensory Evaluation through field trial**

3.2.8.1 **Field Survey - questionnaire administration**

Background data and information on the expectation of cost and quality attributes that are desirable in a weaning food was obtained by means of a questionnaire (Appendix 2) and consumer oriented sensory evaluation was also conducted on a total of 15 test samples and 2 manufactured weaning foods. This was done in 1997. Mothers were asked to rank the samples based on their preference for or degree of liking with respect to colour, flavour, taste and overall acceptability. The questionnaire was administered to mothers selected at random from

5 communities in 3 districts of the Central Region namely: Cape Coast in Cape Coast Municipal, Abandze, Taido Anomabu, Pomaase in Mfantseman district and Assin Jakai in Assin district.

3.2.8.2 Sample preparation of test samples used.

A) Dehydrated fermented maize dough.

The maize dough was fermented for 48 hours and oven dried at 60°C for 12 hours. This was hammer milled and 16 and 32mg/100g of ferrous sulphate and ferrous fumarate were added and mixed thoroughly using Kenwood mixer for 15 minutes. A sample with no iron was also presented.

B) Roasted maize /cowpea/groundnut blend

Cowpea and groundnut were roasted separately at 140°C for 15 minutes. These were mixed with roasted maize at 175°C for 15 minutes. The mixture maize, cowpea, groundnut was in the proportion 80%: 10%: 10%. The mixture was milled using disc attrition mill and allowed to cool. The resulted flour was mixed thoroughly with 16 and 32mg/100g of ferrous sulphate and ferrous fumarate using Kenwood mixer for 15 minutes. A sample with no iron was also presented.

C) Dehydrated fermented maize/ cowpea dough

Another product prepared from fermented maize/cowpea was used in this trial. Maize

was soaked in water for 24 hours, washed and drained. Cowpea was soaked in water for 4 minutes and dehulled using the disc attrition mill. Soaked maize and the dehulled cowpea were mixed in the proportion 4:1 and ground using the disc attrition mill to obtain a fine meal. The meal was kneaded with water (50% moisture content) to form smooth dough, which was left to ferment at room temperature for 48 hours. This was oven dried at 60°C for 12 hours and hammer milled. 16 and 32mg/100g of ferrous sulphate and ferrous fumarate were added to the dehydrated samples and mixed thoroughly using Kenwood mixer for 15 minutes. A sample with no iron was also presented.

3.2.8.3 **Sample Presentation**

Six test samples each of the three weaning foods including the proprietary weaning foods were prepared at 7% solids into porridge and coded. Mothers were requested to arrange in order of preference with respect to colour, taste, flavour and general acceptability. Each attribute was evaluated separately. The porridges were given scores one to six, one (1), indicating the most preferred and six (6), representing the least preferred and the rank sum of each sample calculated. Friedmann's table was used in the analysis.

4.0 RESULTS AND DISCUSSION

4.1 Effect of fermentation on iron fortified maize dough

4.1.1 pH

Fermentation of maize dough generally resulted in decrease in pH as reported by Akpapunam and Sefa-Dedeh (1995). Addition of iron compounds (ferrous sulphate and ferrous fumarate) before and after fermentation of the dough boosted the buffer activity of dough, thus slowing down the rate of decrease in pH.

Addition of iron compounds before fermentation reduced pH of the maize dough up to 36 hours of fermentation after which there was a rise. This was observed for samples fortified with 16 and 32mg of iron per 100g (Figure 3A).

Addition of 16mg of iron compounds after fermentation of maize dough did not change trend in pH of the system even though the values were slightly higher. However, addition of 32mg of iron compounds after fermentation of the dough increased pH values similar to that observed when maize dough was fortified before fermentation (Figure 3B). This implies that pH of the iron-fortified maize system may be influenced by fermentation and iron concentration. There was no effect of the type of iron compounds used on pH values.

In general, the samples prepared by fortifying the product with iron before fermentation had high pHs. In the case of fortification of maize dough with iron after fermentation, the samples with high iron fortification level (32mg/100g) had high pH products whilst those of lower iron fortification level (16mg/100g) produced low pH products similar to the non-fortified product. This implies that to produce an iron-fortified fermented maize dough with low pH similar to that

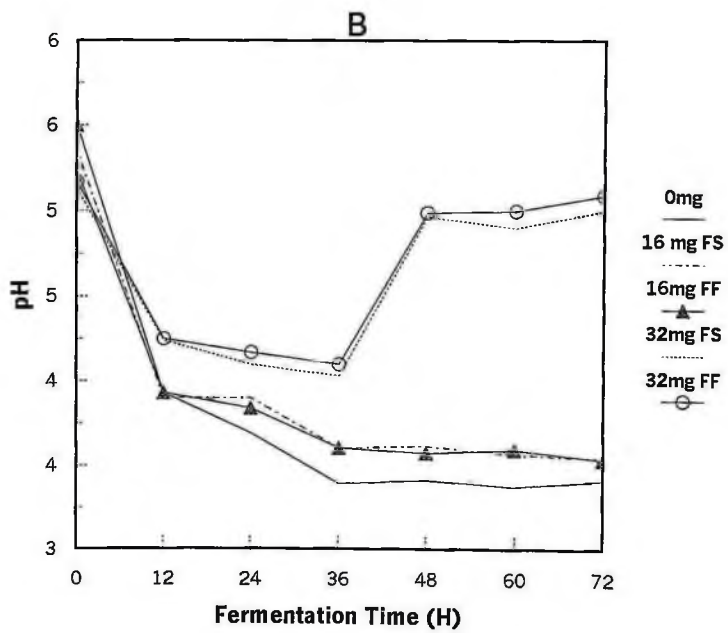
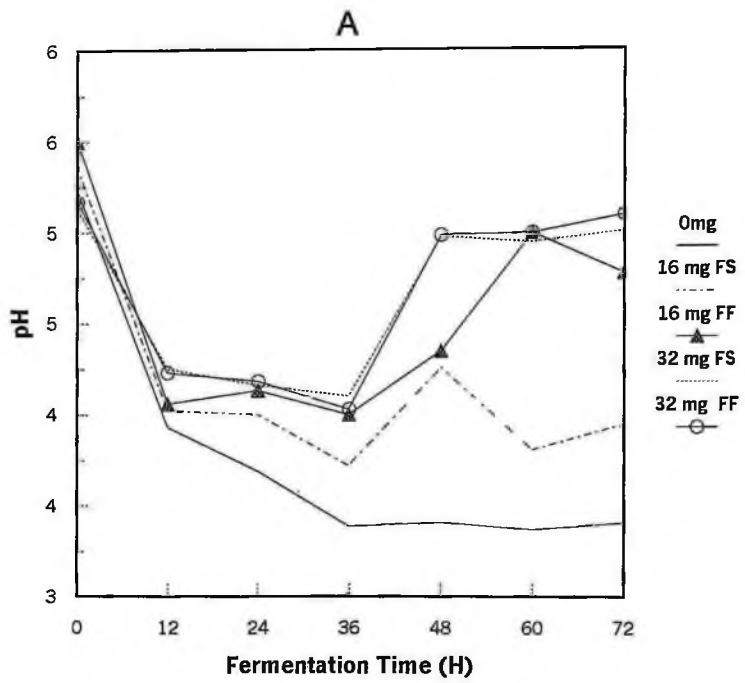
Analysis of variance (ANOVA) on the data (Table 7) showed that the critical parameters, which influence pH of the product, were fermentation time, the amount of iron added and the timing of fortification in relation of fermentation. The significant effect of fermentation time on pH is expected because of the production of carboxylic acids during fermentation. Akpapunam and Sefa-Dedeh in 1995 reported reduction in pH of the non-fortified fermented dough.

Table 7 ANOVA summary table of pH of fresh and dried fermented maize dough samples

Source of variation	Sum of squares	d.f	Mean squares	F-ratio
Iron compound	0.071539	1	0.0715389	1.867
Fermentation time	36.345897	6	6.0576495	154.494*
Iron level	13.65627	2	6.8212644	173.823*
Mode of addition	0.840588	1	0.8405882	21.420*
State of sample	0.093942	1	0.0939415	2.344
Fermentation time X iron level	6.825193	12	0.5687661	14.494*
Iron compound X mode of addition	0.302953	1	0.3029534	7.720*
Fermentation time X mode of addition	0.562787	6	0.0937979	2.390*
Iron level X mode of addition	1.635880	2	0.8179400	20.843*
Iron compound X state of sample	0.057910	1	0.0579096	1.476

* significant ($p \leq 0.05$)

Figure 3 pH changes in maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation.



A significant interaction ($p \leq 0.05$) between fermentation time and the amount of iron added to the product implied that pH of the fortified fermented dough at each fermentation was dependent on the amount of ferrous compounds added. An interaction between fermentation time and the mode of iron addition suggest that the pH developed for fermentation in the products at each fermentation time is dependent on when the ferrous salt was added. The type of iron compound and the mode of fortification are therefore important.

Duncan multiple range analysis showed that the effect on pH by fermentation time of 24, 36 and 48 hours was different from that of 12, 60 and 72 hours. The effect no unfermented products was entirely different from the fermented products. It also showed that the addition of iron compounds before fermentation had different effect on pH from the addition after fermentation. The effect of iron levels (0, 16 and 32mg/100g) on pH of the products was also different.

4.1.2 Titratable acidity

The titratable acidity in the maize dough showed consistent increase with fermentation time. When iron was added to maize dough there was increase in acidity up to 36 hours of fermentation after which there was a decline. This phenomenon was observed when iron compounds at the levels of 16 and 32mg/100g were added before fermentation. There was however, a sharp increase in acidity after 48 hours in the samples fortified with 32mg of ferrous sulphate was added before fermentation of the maize dough (Figure 4A). The decrease in titratable acidity may be due to reaction of acid with the iron to form $\text{Fe}(\text{OH})_2$.

When iron compounds were added after fermentation of maize dough, there was generally an increase in titratable acidity. However, addition of 32mg of ferrous fumarate recorded a

decline after 48 hours of fermentation. Addition of 32mg ferrous sulphate showed the same trend irrespective of when it was added (Figure 4A & B).

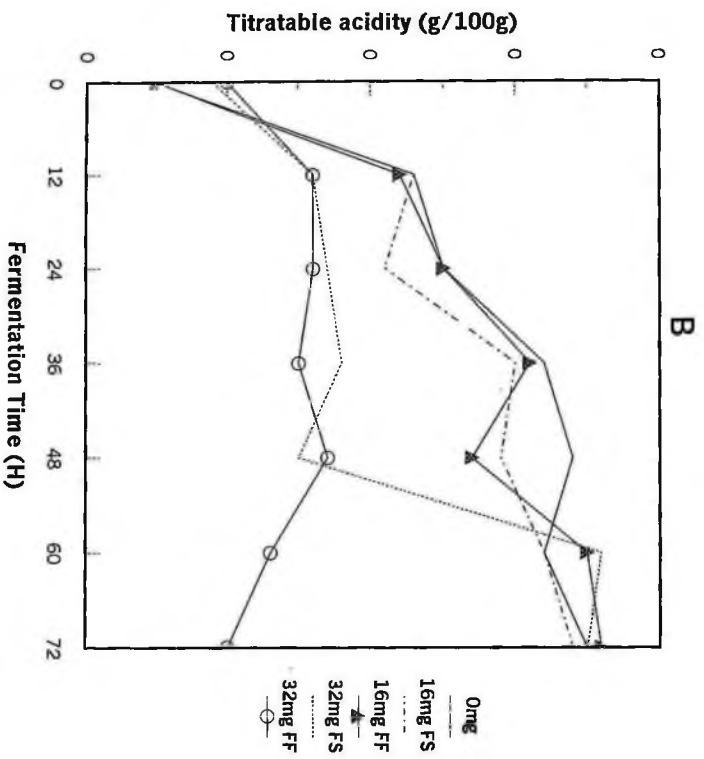
Analysis of variance (Table 8) revealed the influence of the same parameters as occurred in pH. Iron fortification has an effect on the acidity of the products. The interaction of iron with acid was strong during the initial stages of fermentation. This is because iron exist in solution at low pH as $Fe(OH)_2$. This suggests that the time of addition of iron fortificant may influence the quality or end product indices required.

Table 8 ANOVA summary table of acidity of fresh and dried fermented maize dough samples

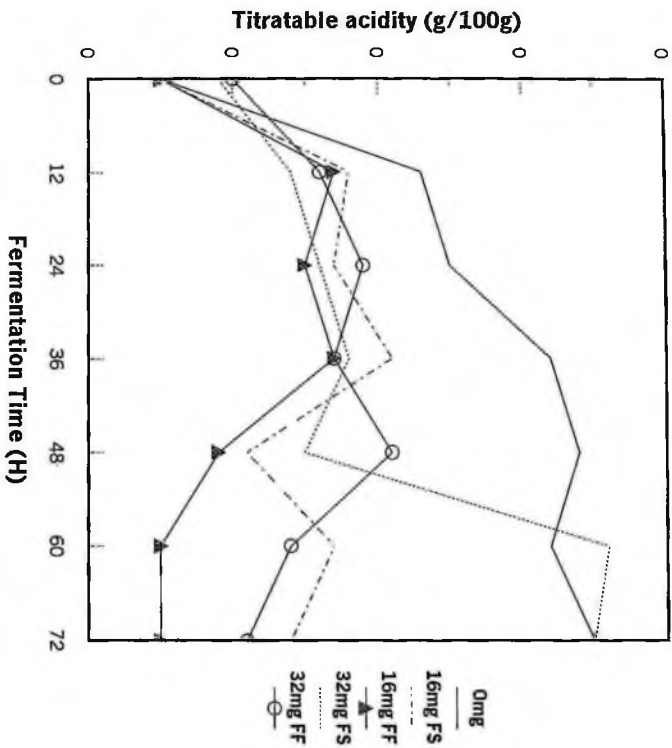
Source of variation	sum of squares	d.f	Mean square	F-ratio
Iron compound	0.0007807	1	0.0007807	0.55
Fermentation time	0.1489029	6	0.0248171	17.487*
Iron level	0.0319606	1	0.0319606	22.52*
Mode of addition	0.0230138	1	0.0230138	16.216*
State of sample	0.0041217	1	0.0041217	2.904
Fermentation time X iron level	0.0200348	6	0.0033391	2.353*
Iron compound X mode of addition	0.0018665	1	0.0018665	1.315
Fermentation time X mode of addition	0.0279648	6	0.00046608	3.284*

* significant ($p \leq 0.05$)

Figure 4 Changes in titratable acidity (g/100g) of maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation



A



4.2 Effect of drying on quality of fermented and iron-fortified maize dough

4.2.1 pH

Dehydrated fermented maize dough recorded relatively higher pH values than the wet product indicating the possible loss of organic acids during drying and consequent increase in pH. A sharp decrease in pH was recorded for all samples (both iron-fortified and non-fortified) up to 12 hours of fermentation after which there was a steady decrease for the non-fortified product.

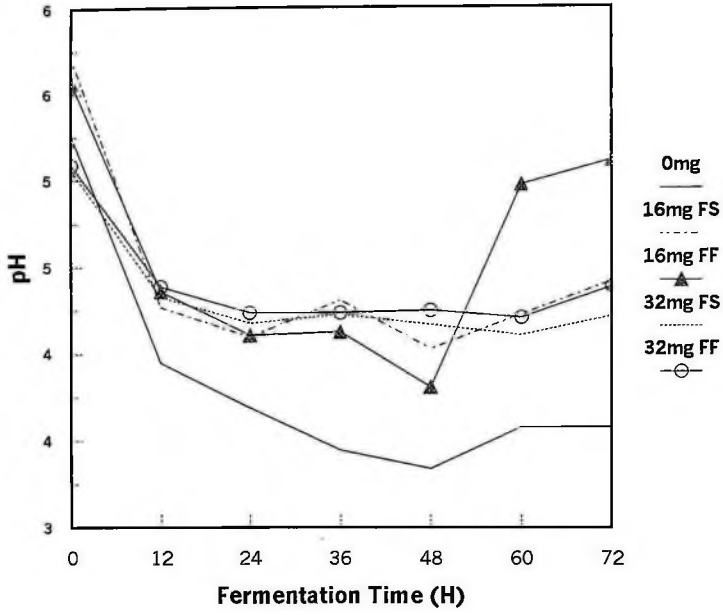
Maize dough fortified with iron before fermentation, when dried had similar pHs after a sharp decrease at fermentation time of 12 hours. There was however, an increase in pH when 16mg ferrous fumarate was added after 48 hours of fermentation. Generally the pHs of iron-fortified products were higher than that of the non-fortified product (Figure 5A).

In the case when maize fortified with iron after fermentation were dried, the pHs recorded for 16mg addition of both iron compounds were similar to that of the non-fortified product. The pHs of 32mg iron addition were however, relatively higher after a sharp decrease after the 12 hours of fermentation (Figure 5B).

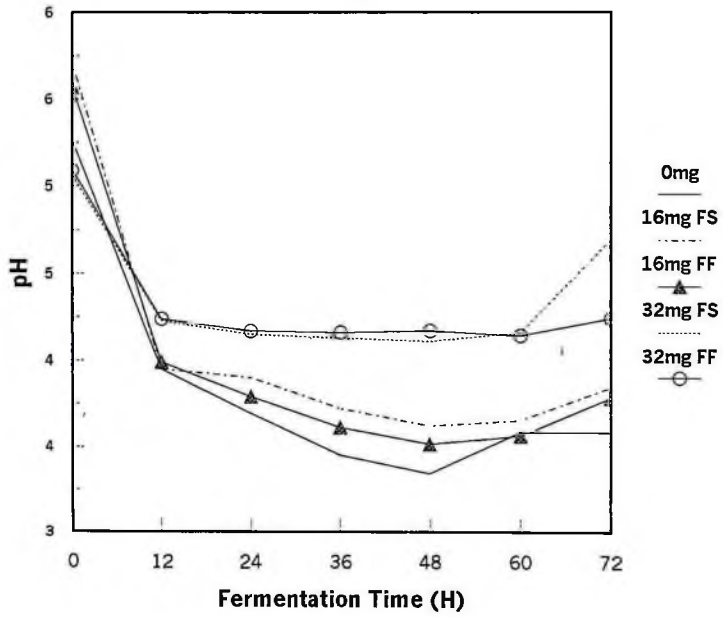
Analysis of variance showed that changes in pH with drying was not significant (Table 7). This means that drying of products in oven at 60°C had no effect on pH values of both iron-fortified and non-fortified fermented maize dough. Similar observation was made by Sefa-Dedeh and Akpapunam (1995). However, iron fortification affected pH of dehydrated products.

Figure 5 pH changes in dehydrated maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation.

A



B



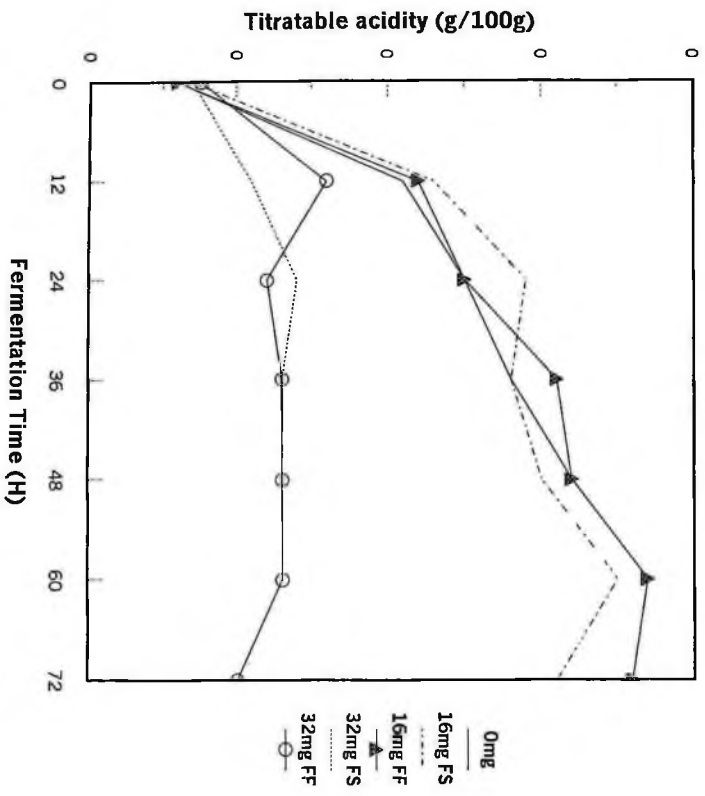
4.2.2 **Titrateable acidity**

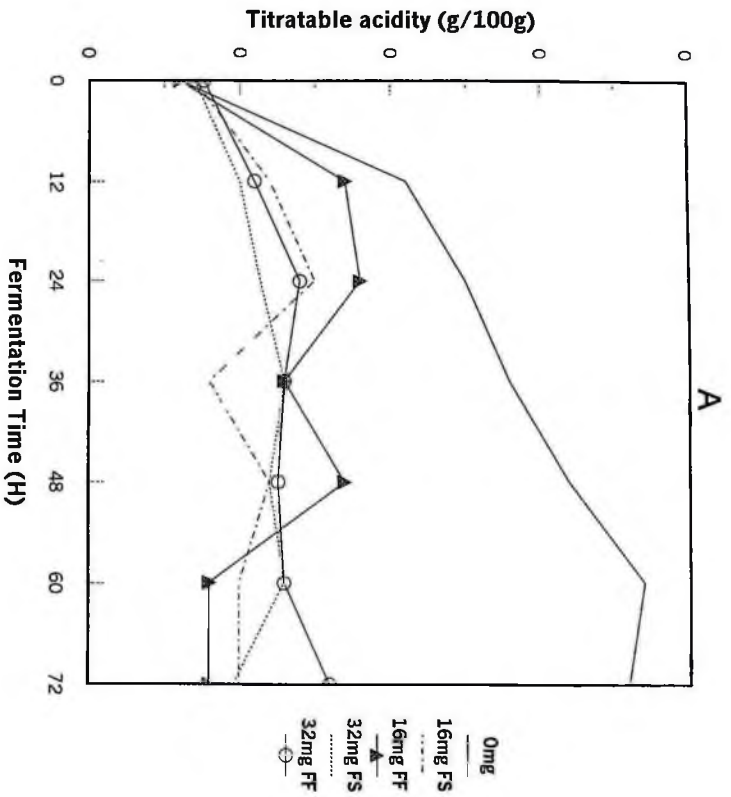
Lower acidity was recorded for dehydrated fermented maize dough as compared to the wet sample (Figure 6). When maize dough was fortified with the ferrous salts before fermentation and dried, titrateable acidity was lower than that of non-fortified sample (Figure 6A). When maize dough was fortified after fermentation, the titrateable acidity recorded for the low level of iron fortification (16mg) was similar to the non-fortified samples whereas that for the high level of iron fortification (32mg) was lower (Figure 6B). This suggests that the mode of iron addition influence the interaction between iron and acid. Analysis of variance however, indicated that changes in titrateable acidity were not significant with drying (Table 8).

It was reported by Smith (1983) that with acidic (pH at 4) protein source meal such as in beef, casein, egg albumin and soy protein isolate, the apparent retention of iron was greater than when pH was at neutrality. The lowest pH obtained for the iron-fortified products was around 4, which is suitable for iron retention and bioavailability.

Figure 6 Changes in titratable acidity (g/100g) of dehydrated maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation

B





4.2.3 **Fat acidity**

Determination of free fatty acids gives a good indication of quality changes occurring in the sample (Paredes-Lopez and Mara-Escobedo, 1983). Free fatty acid results from the hydrolysis of the glyceride molecule. Foods containing even minor amounts of lipids (e.g. <1%) can have a shelf life limited by lipid oxidation and subsequent rancidity (Nielsen, 1994). This is because hydrolysis of short-chained fatty acids are volatile. In addition to free fatty acids, acids phosphates and amino acids can contribute to acidity.

Hydrolytic changes in lipids were followed by determining fat acidity. There was an increase in fat acidity as fermentation time increased. This means that there has been breakdown of the triglycerides to yield free fatty acids as fermentation occurred. The fat acidity for the dehydrated fermented maize dough ranged between 33.96 and 143.68mg KOH/100g sample of which the highest value was recorded between 48 and 72 hours of fermentation. The high value could be due to the fatty acids produced during fermentation since it was observed that acidity was high at 48 hours of fermentation. This could also be due to the large surface area of the flour as reported by Nawar (1985) that the rate of lipid oxidation increases in proportion to the surface area of the sample to air.

Addition of 16 and 32mg/100g ferrous sulphate before and after fermentation raised the fat acidity (Figure 7A and B). A similar phenomenon was observed when ferrous fumarate was used in the fortification (Figure 8A and B). A high fatty acid value in a product can cause unacceptable off-flavours.

However, analysis of variance (Table 9) showed that fermentation time and mode of addition of iron were the parameters influencing fat acidity of the dehydrated products. Iron is most involved in the formation of complexes and free radicals with oxygen that are energised

sufficiently to remove hydrogen from the fatty chain and from free radicals derivatives of fatty acids (Anderson, 1985). This implies that fat acidity, which measures the rate of breakdown of triglycerides of fermented maize dough, did not depend on the type and concentration of iron added but rather the how it was added.

Duncan multiple range analysis indicated that 0 and 36 hours of fermentation had separate effect on fat acidity. However, 48 and 72 hours fermentation had the same effect on fat acidity.

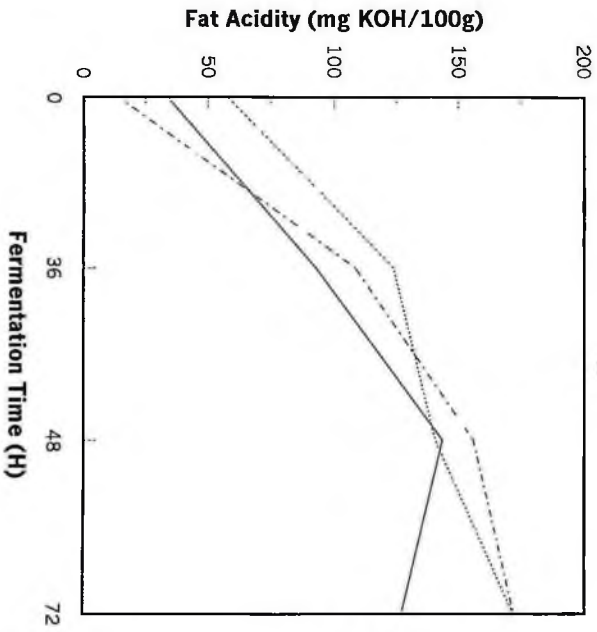
Table 9 ANOVA summary table of fat acidity of dehydrated and stored fermented maize dough

Source of variation	sum of squares	d.f	Mean square	F-ratio
Fermentation time	374768.37	3	124922.79	133.733*
Iron level	3038.51	2	1519.26	1.626
Mode of addition	5245.61	1	5245.61	5.616*
Storage	100949.59	2	50474.8	54.034*

* significant ($p \leq 0.05$)

Figure 7 Changes in fat acidity (mg KOH/100g) of ferrous sulphate (FS) fortified dehydrated maize dough before (A) and after (B) fermentation

B



0mg
16mg FS
32mg FS

A

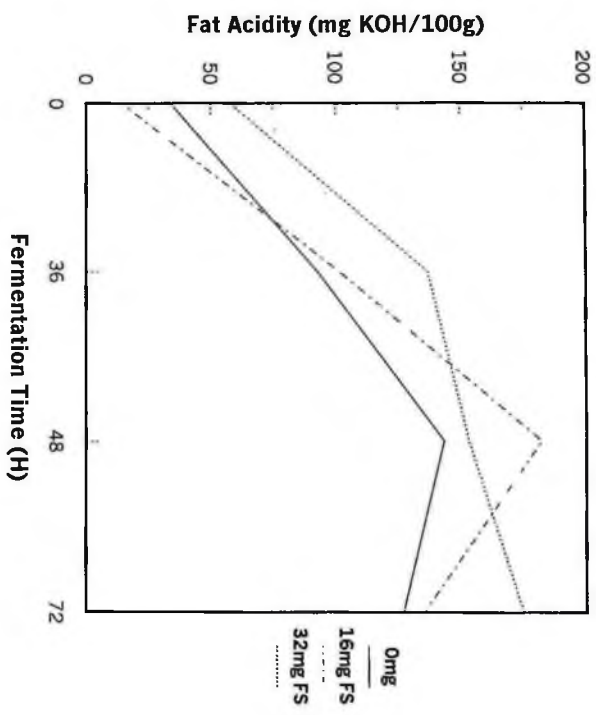
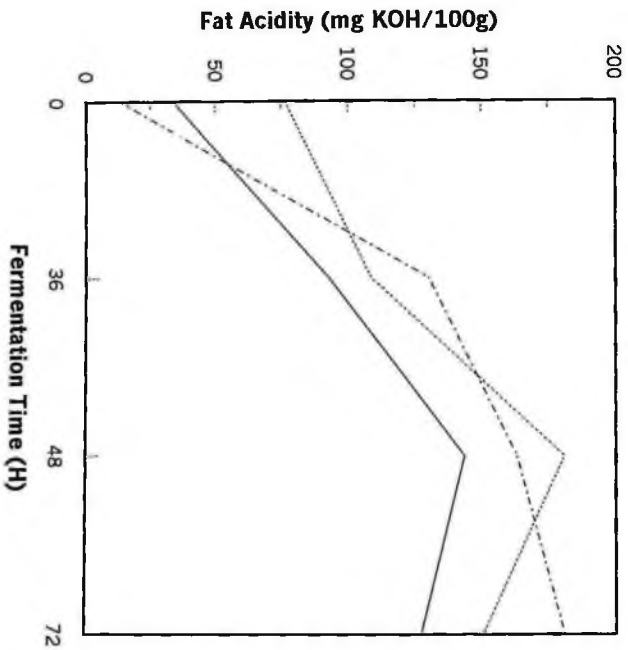
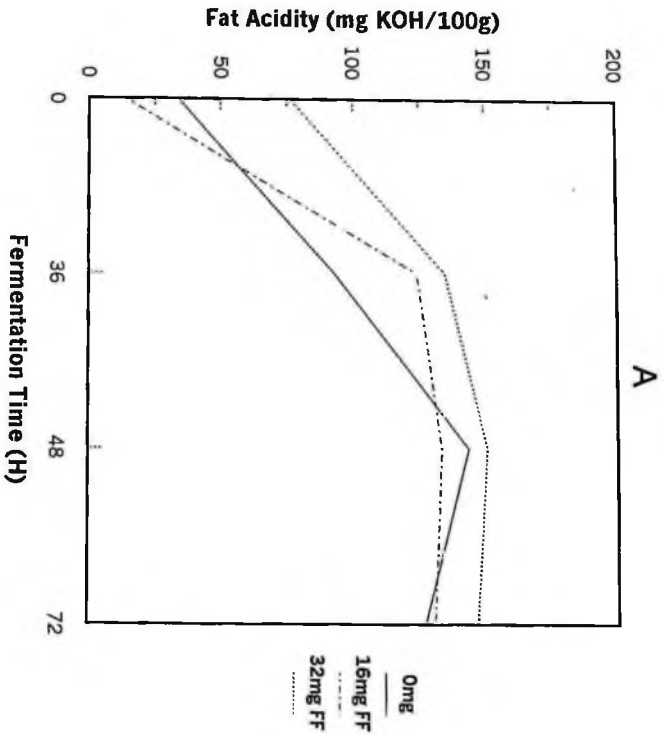


Figure 8 Changes in fat acidity (mg KOH/100g) of ferrous fumarate (FF) fortified dehydrated maize dough before (A) and after (B) fermentation

B



0mg
16mg FF
32mg FF



4.2.4 Colour

The flour samples of both iron-fortified and non-fortified fermented maize dough were visibly cream white in colour and free flowing. Colour measurement was done to determine whether there are changes as iron is added to the product. In general, fermentation resulted in gradual increase in lightness (L^*) of colour. The improvement in colour during fermentation might be due to acid production.

There was a decline in lightness of colour when iron was added and further decline with increase in iron fortification. From the results presented in Table 10 and 11, there was not much difference in lightness of colour due to the source of iron or mode of addition. Analysis of variance on the data of lightness of colour in Table 12 showed however that fermentation time and iron level had significant effect ($p \leq 0.05$) on lightness of colour of the products.

Colour is affected by the addition of iron since there might be a reaction of iron and tannin to form dark colour (Waddel, 1973). There was also a significant interaction between the type of iron and level added. The interaction means that the change in the level of iron added significantly affects the influence on the type of iron added on lightness of colour.

Multiple range tests indicated that 0 and 36 hours of fermentation had different effect on lightness of colour. There was however no difference in the effect of 48 and 72 hours of fermentation. Iron addition at each level had different effect on lightness of colour.

The values of a^* were positive signifying reddish colour co-ordinate for non-fortified and 16mg ferrous sulphate fortified products at all stages of fermentation. Negative values signifying greenish colour co-ordinate for 32mg ferrous sulphate addition were recorded. These were observed irrespective of the mode of addition. All the b^* values were positive signifying yellowish colour co-ordinate for all products.

Table 10 Hunter colour values for dehydrated fermented maize samples fortified with ferrous sulphate

Method of fortification	Iron level mg/100g	Fermentation Time (H)											
		0			36			48			72		
		L*	a*	b*	L*	a*	b*	L*	a*	b*	L*	a*	b*
	0	90.37	0.12	17.21	95.03	0.10	10.21	93.70	0.10	10.59	91.81	0.11	11.58
Before	16	89.40	0.07	18.62	94.48	0.16	10.49	93.22	0.18	10.63	91.61	0.26	12.36
Fermentation	32	83.02	-0.45	12.98	85.59	-0.52	10.93	85.38	-0.58	10.76	85.08	-0.56	11.10
After	16	89.40	0.07	18.62	94.30	0.04	10.66	93.30	0.05	10.71	92.13	0.06	11.79
Fermentation	32	83.02	-0.45	12.98	85.50	-0.56	10.98	85.28	-0.49	11.54	85.23	-0.41	11.03

*: 1. Average of three determinations

2. Absolute measurements

Table 11 Hunter colour values for dehydrated fermented maize samples fortified with ferrous fumarate

Method of fortification	Iron level mg/100g	Fermentation Time (H)											
		0			36			48			72		
		L*	a*	b*	L*	a*	b*	L*	a*	b*	L*	a*	b*
	0	90.37	0.12	17.21	95.03	0.10	10.21	93.70	0.10	10.59	91.81	0.11	11.58
Before	16	88.08	0.33	21.23	94.08	0.17	10.74	93.43	0.20	10.98	91.27	0.25	12.32
Fermentation	32	83.64	-0.31	12.38	85.53	-0.61	10.97	85.52	-0.61	10.65	83.97	-0.40	11.99
After	16	88.08	0.33	21.23	94.56	0.02	10.45	93.11	0.03	10.73	91.47	0.04	11.87
Fermentation	32	83.64	-0.31	12.38	85.26	-0.58	11.12	84.84	-0.36	11.42	84.01	-0.39	11.74

*: 1. Average of three determinations

2. Absolute measurements

The other colour co-ordinates, that is, reddish, greenish and yellowish were all dependent on both the type and concentration of iron added. From the values obtained, ferrous sulphate was more reactive than ferrous fumarate as far as colour is concerned.

Table 12 ANOVA summary table of colour (L*) of dehydrated and stored fermented maize dough samples

Source of variation	sum of squares	d.f	Mean square	F-ratio
Iron compound	0.22885	1	0.22885	0.076
fermentation time	176.3798	3	58.793	19.427*
Iron level	173.783	2	86.891	28.711*
Mode of addition	0.262	1	0.262	0.087
Storage	626.863	2	313.431	103.566*
Iron compound X iron level	489.152	2	244.576	80.814*

*significant ($p \leq 0.05$)

4.2.5 Water absorption index

Water absorption index measures the capacity of the component in the product to retain water. This is a desirable quality index because it provides body and viscosity to the final product when reconstituted in water (Saalia, 1995). Generally water absorption decreased with increase in fermentation time at both 30°C and 70°C for the maize dough. There was however, an increase after 48 hours of fermentation. This is likely to be due to the degradation of starch molecules resulting in lower molecular weight dextrans and sugars that have available hydrophilic groups as fermentation proceeds. Sefa-Dedeh and Ampadu (1991) reported on samples containing fermented maize dough generally having low water absorption properties.

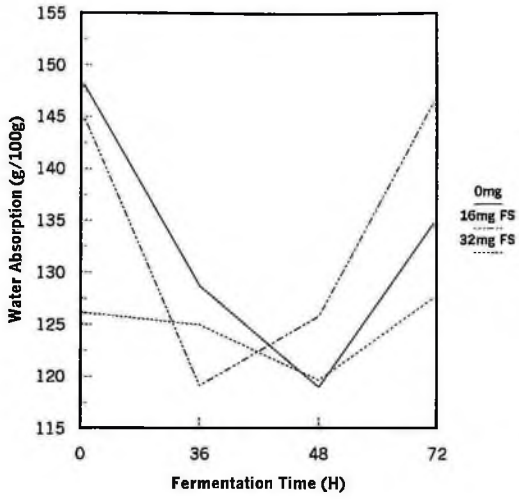
Addition of 16mg of ferrous sulphate before and after fermentation of maize dough did not change the trend. Water absorption however was stable during fermentation when 32mg ferrous sulphate was added to maize dough (Figure 9). Similar results were reported by Frimpong (1995) in fermented maize dough fortified with cowpea. This implies that, ferrous sulphate addition to fermented maize dough did not affect the food components responsible for water absorption.

Generally water absorption at 70°C was higher than 30°C. Proteins are primary sites for water absorption (Sefa-Dedeh, 1988) and the binding sites become more exposed on application of mild treatment resulting in increased water absorption (Phillips *et al.*, 1988). Similar observations were made by Frimpong (1995); Saalia (1995) and Ampadu (1994). A trend similar to that for ferrous sulphate occurred when ferrous fumarate was used to fortified fermented maize dough (Figure 10)

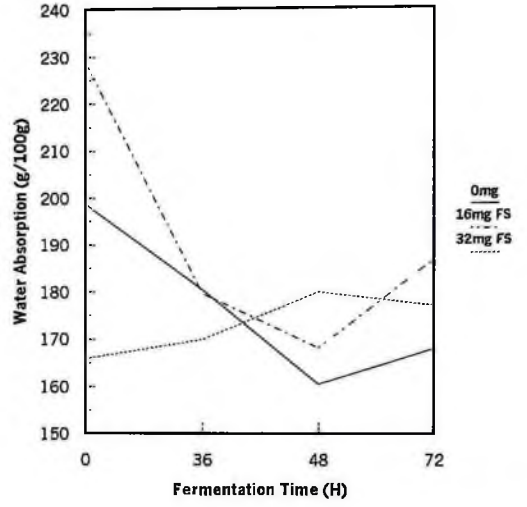
Analysis of variance on the data (Table 13) confirmed that fermentation time and iron level of fortification had significant effect on water absorption. An interaction between iron compound and concentration was found to influence water absorption. This implies that water absorption of the iron-fortified fermented maize dough for each iron source was dependent on the level of fortification. Multiple range analysis indicated that fermentation for 36 or 48 hours did not have any significantly different effects on water absorption. The 0 and 72 hours also have the same effect. In the case of iron fortification level, 0 and 16mg iron addition had the same effect on water absorption.

Figure 9 Changes in water absorption in ferrous sulphate (FS) fortified dehydrated maize dough before fermentation at 30°C (A) and 70°C (B) and after fermentation at 30°C (C) and 70°C (D)

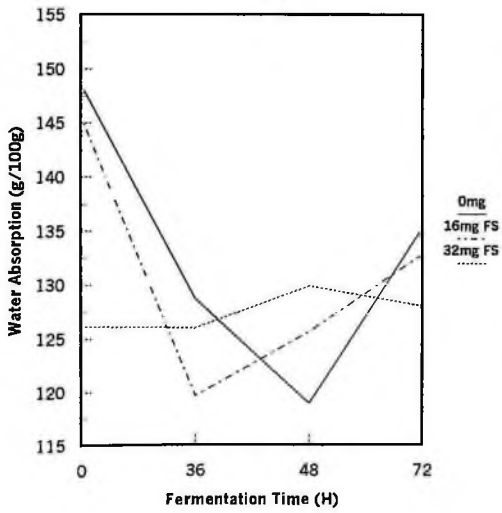
A



B



C



D

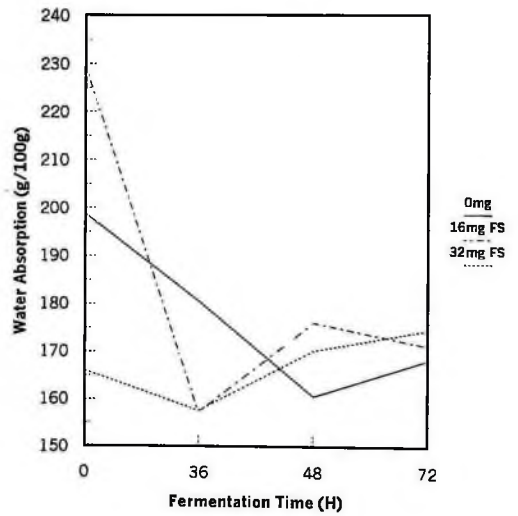
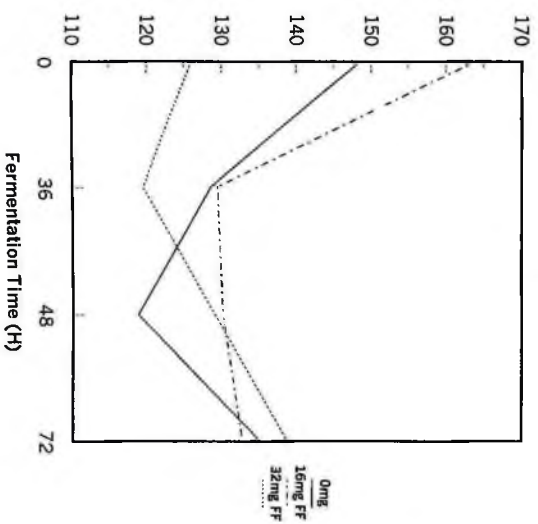
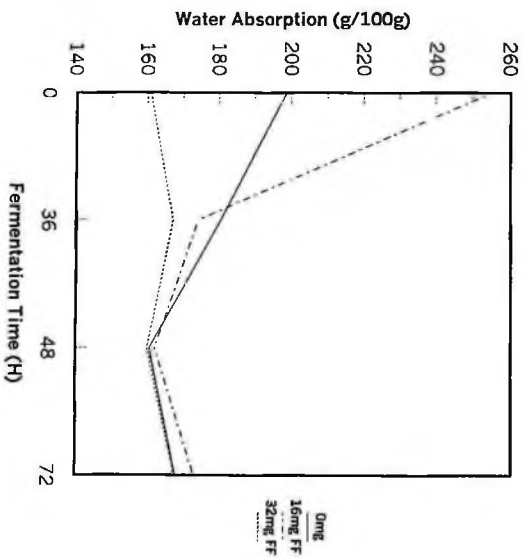


Figure 10 Changes in water absorption in ferrous fumarate (FF) fortified dehydrated maize dough before fermentation at 30°C (A) and 70°C (B) and after fermentation at 30°C (C) and 70°C (D)

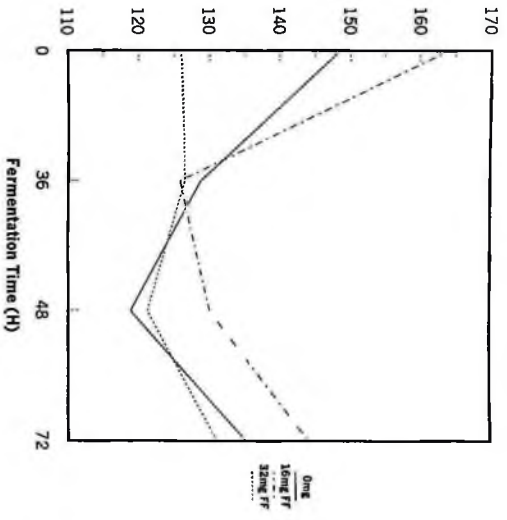
C



D



A



B

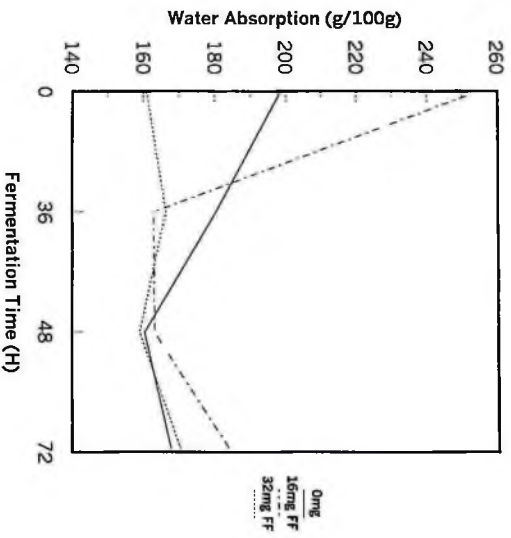


Table 13 ANOVA summary table of water absorption dehydrated and stored fermented maize dough samples

Source of variation	Sum of squares	d.f	Mean square	F-ratio
Iron compound	3.474	1	3.474	0.015
fermentation time	40472.873	3	13490.958	57.693*
Iron level	5020.377	2	2510.189	10.735*
Mode of addition	102.727	1	102.727	0.439
Storage	859.013	2	429.506	1.837
Iron compound X iron level	4179.367	2	2089.6834	8.936*
Fermentation time X iron level	8717.613	6	1452.9354	6.213*

* significant ($p \leq 0.05$)

4.2.6 Apparent Hot paste viscosity (90°C)

Consistency of the cooked paste was evaluated through measurements of the viscosity using the Brookfield viscometer. Viscosity is a reflection of the swelling of the starch paste affected by the starch concentration. Generally high starch concentration leads to a low pasting temperature. The viscosity of the iron-fortified fermented maize dough was determined to find out the effect of iron on the swelling property of starch. The apparent hot paste viscosity refers to the paste viscosity of 6% solid concentration of iron-fortified fermented maize dough flour at 90°C.

Generally an increase in viscosity of fermented maize dough as fermentation progressed was observed. Akpapunam and Sefa-Dedeh (1995) had earlier also reported increase in viscosity with fermentation in maize dough.

Fortification with 16mg iron salt led to high hot paste viscosity than the 32mg iron fortified samples. The values for non-fortified products were similar to that of the 16mg addition of iron salts. This occurred irrespective of how the iron was added (Figure 11A and

B). The increase in hot paste viscosity suggests that dough fermentation apart from flavour development also contributes to improve the rate and degree of starch gelatinisation and therefore promotes ease of cooking (Sefa-Dedeh, 1988). Similar observations were made on cowpea fortification of fermented maize dough by Akpapunam and Sefa-Dedeh (1995), Ampadu (1994) and Frimpong (1995). This implies that addition of 16mg of iron salts does not interfere with starch gelatinisation.

It was reported that lactic fermentation did not cause a reduction of the viscosity in boiled product on contrary; the acidity causes a viscosity increase (Lorri, 1993). Addition of 32mg of iron reduces hot paste viscosity and this might be due to the decrease in acidity as more ferrous salts are provided to react with the acids produced.

Generally low hot paste viscosity was recorded for both non-fortified and iron-fortified products. Highest values recorded were 262 and 316 cP for non-fortified and iron-fortified samples respectively. For weaning foods, a low hot paste viscosity is desirable (Desikacher, 1980).

Analysis of variance (Table 14) showed that hot paste viscosity was influenced by fermentation time and iron level. A significant interaction between fermentation time and iron concentration implied that, at each fermentation time, viscosity of the iron-fortified dough was dependent on the iron level added. Multiple range analysis revealed that fermentation at 36 and 48 hours have the same effect on viscosity. Fermentation at 48 and 72 hours also has the same effect on viscosity. The unfermented dough was entirely different from the fermented products. There was no significant difference between the non-fortified and the iron fortified products as far as viscosity is concerned. The higher iron fortification level (32mg) was however different in its effect on viscosity.

Figure 11 Hot paste viscosity changes in dehydrated maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation

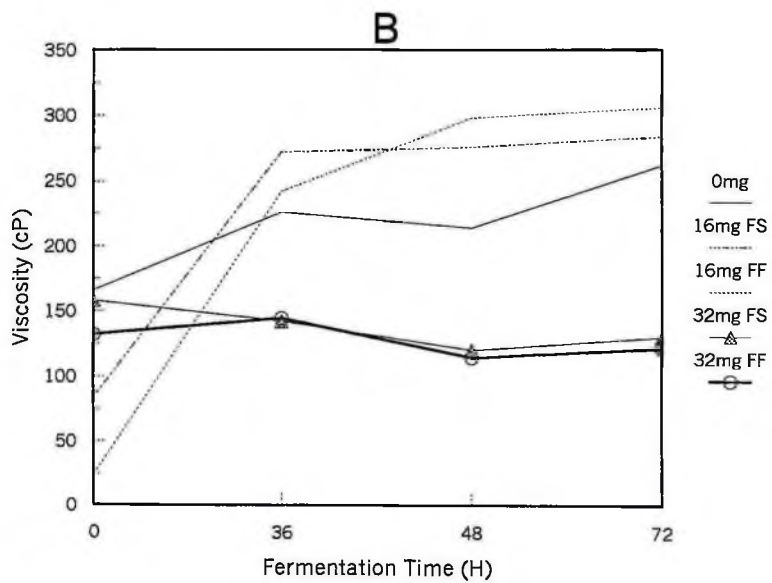
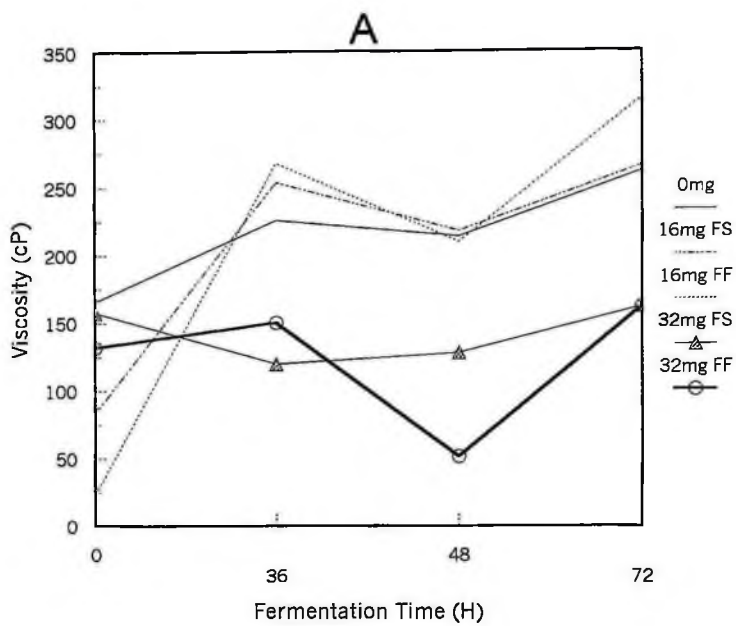


Table 14 ANOVA summary table of viscosity of dehydrated fermented maize dough samples

Source of variation	Sum of squares	d.f	Mean square	F-ratio
Iron compound	1113.8	1	1113.8	0.942
fermentation time	269914.1	3	89971.37	76.077*
Iron level	99434.3	2	49717.16	42.039*
Mode of addition	290.5	1	290.51	0.246
Fermentation time X iron level	136026.1	6	22671.017	19.17*

* significant ($p \leq 0.05$)

4.2.7 Apparent cooled paste viscosity (50°C)

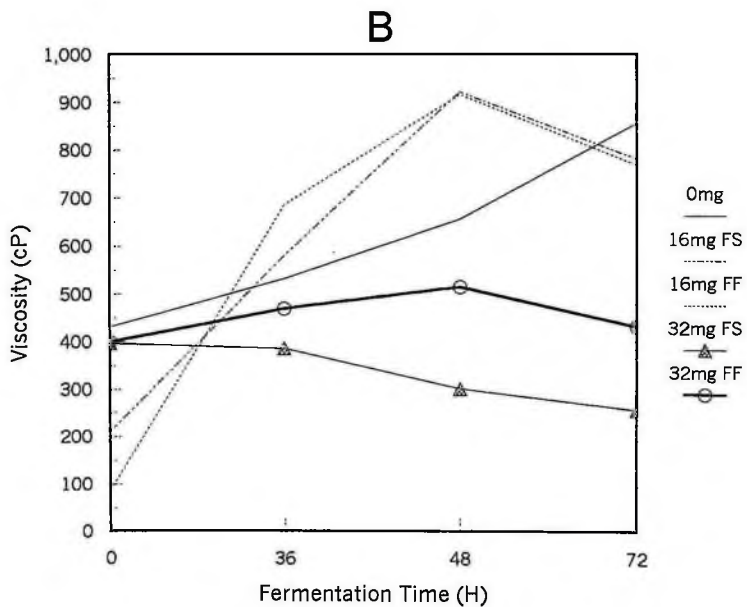
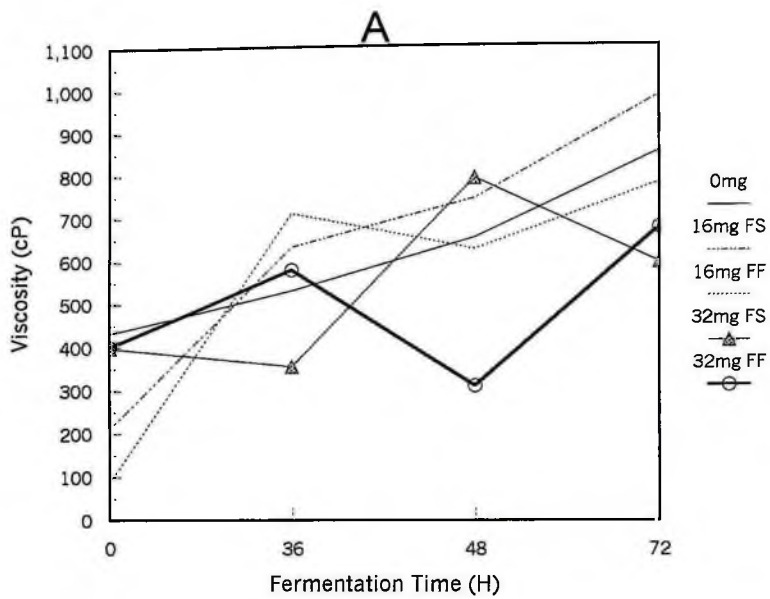
The apparent cooled paste viscosity refers to the viscosity of the cooked product after the hot paste was cooled to 50°C. It represents the eating characteristic of the product.

The addition of 32mg of ferrous salts slightly lowered viscosity as compared to the 0 and 16mg iron fortification. The trend in cooled paste viscosity is similar to that of hot paste viscosity.

Generally the values obtained for cooled paste viscosity were higher than that for the hot paste viscosity (Figure 12A and B). According to Hoover and Sosulski (1990), when pastes are cooled, the consistency increases as molecular association forms a cross-linked network that increases the paste resistance to deformation. This is consistent with trends reported by Kluitse (1995).

Highest values recorded were 856 and 986 cP for non-fortified and iron-fortified products respectively. This is below the consistency (1,000-3,000 cP), which makes weaning food an easy-to-swallow semi-liquid as reported by Motarjemi, *et al.*, (1993).

Figure 12 Cooled paste viscosity changes in dehydrated maize dough fortified with ferrous sulphate (FS) and ferrous fumarate (FF) before (A) and after (B) fermentation



4.2.8 Energy content

The emphasis for weaning food requirements is on a high caloric density per unit volume of food (Desikachar, 1980). The energy values measured for fermented maize dough was between 470.2 and 523.4 kcal/100g, whilst that for the iron-fortified products recorded between 421.0 and 523.4 kcal/100g (Table 15). Generally, fermentation has influenced the energy value of all the samples.

In the case of iron-fortified products, iron fortification level seems to have influenced energy values. Energy values for ferrous fumarate addition were relatively lower than when ferrous sulphate was added to the fermented samples. The values of 16mg ferrous salt fortified products recorded relatively higher energy values than those with 32mg ferrous salt fortified ones. Addition of ferrous salt to fermented maize dough before fermentation generally recorded higher energy values than the addition after fermentation. Analysis of variance in Table 16 indicated that, all the variables; the type of iron, its concentration and the mode of addition as well as fermentation time had significant effect on the energy content of the products. This implies that energy content of both non-fortified and iron-fortified maize dough are susceptible to change in any of the factors studied.

The range of energy values obtained for the products was higher than that of a recommended weaning formula by KIT, a leading Netherlands NGO (Dijkhuizen, 1989). Lorri (1993) reported energy density of lactic acid-fermented product which had increased three-fold in flour concentration as compared to 0.4 kcal/g for a non-fermented gruel prepared to the same consistency. Quarshie (1988) recorded an energy value of 461.3 kcal/100g sample of a cereal-legume blend used in Ghana. The energy values obtained for both non-fortified and iron-fortified maize dough were higher than the 375 kcal/100g recommended as minimum for high quality weaning foods (PAG, 1975).

Table 15 Effect of fermentation and iron fortification on energy* content of dehydrated maize dough (kcal/100g)

Method of fortification	Iron level mg/100g	Ferrous Sulphate				Ferrous fumarate			
		Fermentation Time (H)							
		0	36	48	72	0	36	48	72
Before fermentation	0	470.2	520.8	523.4	521.2	470.2	520.8	523.4	521.2
	16	501.8	481.2	523.4	476.4	441.4	421.4	462.4	428
	32	462	476.4	496.4	487.4	470.6	491.2	505.4	472.8
After fermentation	16	521.8	469.4	500.2	462.8	441.4	441.4	459.4	440.2
	32	462	430.6	455.6	469.8	470.6	443.8	452	460.2

*: 1. Average of two determinations
 2. g/100g sample on dry matter basis

Table 16 ANOVA summary table of energy on dehydrated fermented maize dough samples

Source of variation	Sum of squares	d.f	Mean square	F-ratio
Iron compound	6.453333	1	6.453333	28.024*
fermentation time	10.7431	3	3.581033	15.551*
Iron level	45.74495	2	22.872475	99.324*
Mode of addition	3.00	1	3.00	13.028*
Iron compound X iron level	16.028467	2	8.0142333	34.802*
Fermentation time X iron level	18.75585	6	3.125975	13.575*
Iron level X mode of addition	4.4768	2	2.2384	9.72*

* significant (p≤0.05)

Duncan multiple range analysis indicated that the effect of 0 and 36 hours fermentation was the same on energy content. The effect of 36 and 48 hours of fermentation was also the same. Effect of 72 hours of fermentation however was different from all the

fermentation time. The effect of iron fortification irrespective of levels was different from the non-fortified product. The effect of iron addition before fermentation was different from the addition after fermentation.

4.2.9 Total iron content

The total iron content determined for raw dried maize was 12.4mg/100g. When maize was soaked in water for 24 hours, total iron content increased to 15.8mg/100g. The increase in iron content may be due to the activity of the microorganisms. Halm *et al.*, (1993) reported that during steeping of maize about 10^6 cfu/g of microbes were enumerated.

The amount of ferrous salt added was 16 and 32mg/100g sample. This however means that based on at least the iron content for the raw dried maize (12.4mg/100g), the estimated total iron content of iron-fortified products should be 28.4 and 44.4mg/100g for the addition of 16mg and 32mg ferrous salt respectively.

Generally an increase in total iron content was observed during fermentation. This might be due to microbial activity. Values are also relatively higher when ferrous salts were added to fermented dough after fermentation as compared to the addition before fermentation. When 16mg of ferrous salt was added before fermentation of maize dough, the determined total iron values were lower than the estimated values. However, addition of 32mg ferrous salt increases iron content as expected (Table 17). The decrease in total iron content might be due to the bacterial nutrition where iron serves as a constituent of cytochromes and other haeme and nonhaeme proteins. It was reported by Smith (1983) that, iron retention response was linear to the iron dose used.

Table 17 Effect of fermentation and iron fortification on total iron content* of dehydrated maize dough (mg/100g)

Method of fortification	Iron level mg/100g	Ferrous Sulphate Fermentation Time (H)				Ferrous fumarate Fermentation Time (H)			
		0	36	48	72	0	36	48	72
Before fermentation	0	15.8	14.5	18	19	15.8	14.5	18	19
	16	19.4	22	26.5	22.1	18.1	20.3	26.4	22.5
	32	43.4	41.2	39.7	54.3	37.1	49.3	48.9	37.8
After fermentation	16	19.8	33.9	26.8	25.5	18.1	24.7	27.5	27.4
	32	43.4	40.3	48.4	50.8	37.1	45.4	40.1	40.4

*: Average of two determinations

Analysis of variance showed that, fermentation time and iron level had a statistically significant effect on total iron content (Table 18). In addition to increasing total iron content fermentation process also provides optimal pH condition for degradation of phytate, which inhibits iron absorption. This process can therefore make more iron available in the maize system.

Duncan multiple range analysis indicated 0 and 36 hours fermentation had the same effect on total iron content, whilst that of 36, 48 and 72 hours also had the same effect. Different levels of iron fortification affected total iron content of the fortified fermented maize dough.

Table 18 ANOVA summary table of total iron content of dehydrated fermented maize dough samples

Source of variation	Sum of squares	d.f	Mean square	F-ratio
Iron compound	0.27	1	0.27	1.827
Fermentation time	1.830742	3	0.610247	4.129*
Iron level	61.755317	2	30.877658	208.925*
Mode of addition	0.085008	1	0.085008	0.575

* significant ($p \leq 0.05$)

4.2.10 Microbiological Evaluation

Bacteria are mainly responsible for the fermentation of cereal and animal products. The major types of bacteria important in cereal fermentation are those that produce lactic acid from available sugar. Iron-fortified maize dough was analysed microbiologically alongside the non-fortified one.

There was an increase in viable microbial counts for all the systems, that is, iron-fortified and non-fortified during fermentation. The increase was from 10^4 to 10^8 cfu/g at the end of fermentation at 72 hours (Table 19). It was reported by Halm *et al.* (1993), that even at the start of steeping, a mixed group of microorganisms including lactic acid bacteria, catalase-positive, Gram-positive bacteria, Gram-negative bacteria, yeast and moulds were found in maize.

Lactic acid bacteria counts were increased by 10^4 and 10^5 cfu/g for non-fortified and iron-fortified products respectively (Table 20). This implies that in spite of the concentration of iron; lactic acid bacteria were able to grow. Nche *et al.* (1994) enumerated similar number of lactic acid bacteria for all fermented maize and cowpea-supplemented maize dough. Nout *et al.* (1989) reported that due to the dominance of lactic acid bacteria and rapid acidification,

Enterobacteriaceae were absent in the fermented material, even though they were introduced with each new batch of cereal and legume meal.

Yeast and mould growth was also seen with maximum numbers of colony forming units in the range of 10^5 cfu/g after 24-48 hours, which was followed by a reduction (Table 21). This was observed for both fortified and non-fortified products.

Table 19 Effect of fermentation on total bacterial count for microorganisms of iron-fortified and non-fortified maize dough

Fermentation Time (H)	Ferrous sulphate (cfu/g)			Ferrous fumarate (cfu/g)	
	0	16	32	16	32
0	3.2×10^4	3.3×10^4	2.8×10^4	3.8×10^4	6.2×10^4
24	3.5×10^6	8.2×10^6	2.3×10^6	4.1×10^6	1.0×10^7
36	4.6×10^6	3.2×10^7	2.1×10^7	6.3×10^6	5.2×10^7
48	4.9×10^8	5.2×10^7	5.4×10^7	8.3×10^7	6.2×10^7
72	5.2×10^8	1.2×10^8	8.1×10^8	3.8×10^8	5.4×10^8

Table 20 Effect of fermentation on lactic acid bacteria count of iron-fortified and non-fortified maize dough

Fermentation Time (H)	Ferrous sulphate (cfu/g)			Ferrous fumarate (cfu/g)	
	0	16	32	16	32
0	8.1×10^3	4.3×10^3	5.2×10^3	4.3×10^3	6.8×10^3
24	2.0×10^3	2.6×10^4	1.8×10^4	8.8×10^3	5.0×10^4
36	4.2×10^6	2.1×10^7	6.2×10^6	2.7×10^6	6.8×10^6
48	4.8×10^7	3.6×10^7	2.2×10^6	6.6×10^7	6.3×10^7
72	6.3×10^7	1.4×10^8	2.3×10^8	2.4×10^8	4.4×10^8

Table 21 Effect of fermentation on yeast and mould count of iron-fortified and non-fortified maize dough

Fermentation Time (H)	Ferrous sulphate (cfu/g)			Ferrous fumarate (cfu/g)	
	0	16	32	16	32
0	2.8×10^3	4.2×10^3	2.3×10^3	4.2×10^3	2.1×10^3
24	5.9×10^3	6.9×10^3	5.2×10^3	6.6×10^3	4.8×10^3
36	2.8×10^5	4.2×10^6	2.5×10^5	1.1×10^6	5.4×10^6
48	4.6×10^5	5.4×10^5	4.3×10^5	4.2×10^5	3.2×10^5
72	9.8×10^3	9.2×10^3	3.2×10^4	1.0×10^4	8.4×10^3

Even though the organisms responsible for fermentation in the systems studied were not isolated and identified, work done by Halm *et al.* (1993) on fermented maize dough revealed that more than 96% of the organisms were obligatively heterofermentative lactobacilli. These were related to *Lactobacillus fermentum* and *Lactobacillus reuteri* in their pattern of carbohydrate fermentation. A dominant role of *Lactobacillus fermentum* in fermented maize dough was observed by Christian (1970).

Yeasts were seen in high levels in all samples investigated, and it cannot be ruled out that they influence the aroma of the final product, as suggested by Akinrele (1970). He found that *Saccharomyces cerevisiae* and *Candida mycoderma* contributed to flavour acceptability of "ogi", a traditional African fermented maize product. In agreement with the previous investigation, the aroma compounds were dominated by organic acids, that is, lactic acid, acetic, butyric and propionic acids (Plahar and Leung, 1982). Yeast and mould counts were reduced after the second day of fermentation in the study. Fields *et al.* (1981) found that viable coliforms, yeast and moulds disappeared from the fermentation of corn meal during the second day. These findings have been attributed to antimicrobial properties in the fermenting maize dough. Enteric pathogens have been found to be inactivated in these fermented foods (Mensah *et al.*, 1991; Simango and Rukure, 1992).

The study revealed that iron fortification at level used did not interfere with the fermenting medium for microbial growth. The ability of iron-fortified maize dough system to ferment and produce acids comparable to the traditional maize dough system is beneficial. In addition, the fortified maize has antimicrobial properties and high iron content.

4.3 Storage stability of iron-fortified and non-fortified dehydrated fermented maize dough

Minerals are often chemically reactive and their bioavailability will often be greatly affected by interactions with food components when added, or during processing and storage. Such reactivity will, of course, vary with the mineral used, the food, the process and storage time and conditions. The dehydrated fermented maize dough flour was stored for 4 months at room temperature.

4.3.1 pH and Titratable acidity

An increase in pH was observed when samples were stored for 2 months at room temperature. However, a decrease after 4 months of storage was recorded. Increase recorded by the non-fortified sample at 2 months storage was greater than that of the iron-fortified products. pHs of the iron-fortified products were almost the same as those for the fresh samples. The non-fortified and the maize dough fortified with 16mg iron after fermentation had pHs lower than 4 (Appendix 3 and 4). Analysis of variance results showed significant effect of storage on pH (Table 22). This implies that pH of both fortified and non-fortified fermented maize samples were influenced by storage.

Multiple range tests showed that at 2 months of storage, effect on pH was different from that of 0 and 4 months. Even though storage had effect on of both fortified and non-fortified products, it is in the range that can inhibit the growth of pathogenic organisms and also increase iron bioavailability as already reported. On the other hand, there was no significant effect of storage on titratable acidity.

Table 22 ANOVA summary table of pH of dehydrated and stored fermented maize dough samples

Source of variation	Sum of squares	d.f	Mean square	F-ratio
Iron compound	0.00028	1	0.00028	0.002
Fermentation time	41.2931	3	13.76436	104.875*
Iron level	0.39777	2	0.198885	1.515
Mode of addition	1.0177	1	1.0177	7.754*
Storage	10.226277	2	5.113139	38.958*
Iron compound X iron level	2.10877	2	1.054387	8.034*
Fermentation time X iron level	1.9976	6	0.3329386	2.537*

* significant ($p \leq 0.05$)

4.3.2 Fat acidity

Ferrous sulphate is an unstable iron source because of its tendency to cause rancidity with prolonged storage. There was an increase in fat acidity for both fortified and non-fortified samples during storage at room temperature. There was about 100% increase in fat acidity for non-fortified and maize dough fortified with 16mg iron after fermentation, whilst those of higher iron fortification level were rather below 50% (Appendix 5). High fat acidity values were recorded initially when 32mg iron was added. Fat acidity increased with lower fortification level addition and non-fortified products occurred with time of storage.

Analysis of variance results had indicated that storage had significant effect on fat acidity (Table 9). Multiple range analysis indicated that storage time of 0, 2 and 4 had different effect on fat acidity.

4.3.3 Colour

There was decrease in all the colour components over storage. The lightness of colour was reduced over the 4 month storage for all samples (Appendix 6). The effect of storage on colour of both fortified and non-fortified samples was found to be significant as indicated by analysis of variance results (Table 12). The time of storage had different effect according to Duncan multiple range analysis. This might due the reaction of tannins and phenolic compounds with iron.

4.3.4 Water absorption index

Increase in water absorption was observed over the 4 month storage for the lower fortification level. Reduction of water absorption was recorded for both 32mg fortified and the non-fortified samples (Appendix 7 and 8).

However, storage did not have a significant effect on water absorption of the samples (Table 13). Plahar (1983) reported a similar observation on air-dried fermented maize dough. He observed that total soluble carbohydrate increase during storage when the dough was air-dried. He also reported changes in pasting properties occurring during storage.

4.4 Optimisation of concentration of legume and iron in roasting weaning blend

Another weaning food with roasted maize flour as the base was also investigated for the effects of fortification with cowpea and iron on the system. Iron and legume concentration were optimised using the Central Composite Rotatable Design to relate to the characteristics

of roasted weaning food such as colour, fat acidity, water absorption, viscosity, energy and total iron content.

Models were developed to relate legume and iron concentration and roasting temperature to the characteristics. Analysis of variance for the full regression of each model and the calculated F-values of lack of fit are also shown in Table 23. These two indicators were used to determine the adequacy of the models. Models with significant analysis of variance and no lack of fit were considered adequate to predict the cowpea and ferrous salt fortified products.

Table 23 Analysis of variance for the full regression of the models and lack of fit (F-values)

Variable	Iron compound	Model	Lack of fit
Colour	Ferrous sulphate	152.74*	117.77*
	Ferrous fumarate	146.36*	3.52
Fat acidity	Ferrous sulphate	3.93*	0.31
	Ferrous fumarate	1.53	2.19
Water absorption at 30°C	Ferrous sulphate	14.22*	0.58
	Ferrous fumarate	2.81	1.69
Water absorption at 70°C	Ferrous sulphate	25.76*	1.87
	Ferrous fumarate	3.30*	0.63
Hot paste viscosity	Ferrous sulphate	7.08*	5.72*
	Ferrous fumarate	11.54*	0.19
Cooled paste viscosity	Ferrous sulphate	4.03*	0.44
	Ferrous fumarate	7.24*	1.6
Energy	Ferrous sulphate	2.58	0.17
	Ferrous fumarate	3.45*	0.64
Total iron content	Ferrous sulphate	4.03*	0.5
	Ferrous fumarate	8.60*	0.57
4 months storage			
Colour	Ferrous sulphate	117.06*	86.38*
	Ferrous fumarate	131.25*	5.04*
Fat acidity	Ferrous sulphate	0.56	1.37
	Ferrous fumarate	15.36*	0.57
Water absorption at 30°C	Ferrous sulphate	12.07*	0.89
	Ferrous fumarate	4.86*	1.52
Water absorption at 70°C	Ferrous sulphate	27.86*	2.44
	Ferrous fumarate	13.02*	0.72

*Significant ($p \leq 0.05$)

4.4.1 Colour

The colour of the roasted weaning blend was visibly brown and free flowing. Lightness of colour was used as an index for the model. The model was adequate only to predict the ferrous fumarate fortified product (Table 23). Colour was affected by the addition of legume (combination of cowpea and groundnut) in the same proportion.

Reduction of the lightness of colour was observed as legume concentration was increased. This is shown in the response surface plot generated in Figure 14. Akpapunam and Sefa-Dedeh (1995) reported a similar colour change when cowpea was added to fermented maize dough. At high legume concentration, there was reduction of lightness of colour when iron was added. The role of roasting temperature in the development of colour was prominent since roasting temperature and its quadratic term had significant effect on lightness of colour.

4.4.2 Fat acidity

Fat acidity has been described as important determinant for product's shelf life since it can lead to rancidity. Values obtained for ferrous sulphate fortification was between 13.8 and 19.1mg KOH/100g sample as compared to 13.1 and 22.3mg KOH/100g sample for ferrous fumarate fortified products. The model was adequate only to predict the ferrous sulphate fortified product (Table 23).

Figure 13 Response surface plot for colour (L*) at 175°C roasting temperature for ferrous fumarate roasted blend

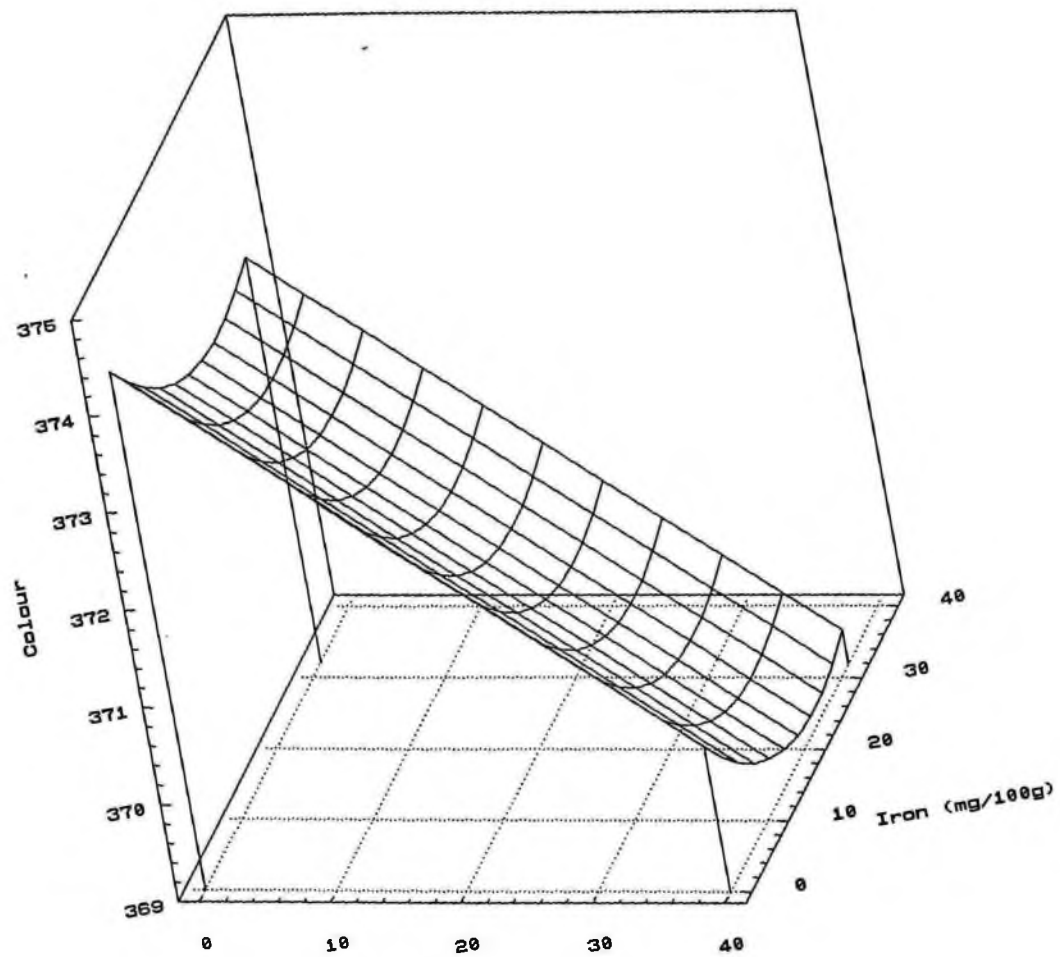
$$Z = -10.35555 - 0.508168X_1 - 0.114408X_2 + 1.344915X_3 + 0.002354X_1X_3 + 0.002451X_2^2 - 0.004887X_3^2$$

$$R^2 = 0.979$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature



Legume (%)

The response surface graph in Figure 14 showed sharp rise in fat acidity as iron concentration was increased. This means that increase in iron concentration increases the breakdown of triglycerides yielding free fatty acids. However, legume addition had decreasing effect on fat acidity. This implies that, reduction of the breakdown of triglycerides in the products could be achieved by the addition of high level of roasted cowpea and groundnut.

Analysis of variance showed that only legume addition had significant effect on fat acidity of roasted the products.

4.4.3 Water absorption index

Water absorption characteristics of cereal/legume blends are important quality attributes, which can affect their behaviour in the food systems. Starch, a major carbohydrate in cereals and legumes has the ability to absorb water when suspension of granules is heated above gelatinisation temperature. The model was adequate only to predict ferrous sulphate fortified roasted blend for water absorption at 30°C (Table 23). However it was adequate to predict for both ferrous salts fortified products when water absorption was measured at 70°C (Table 23).

Generally, water absorption was observed to be higher when temperature was rising. Also, the values obtained for ferrous fumarate were relatively higher as compared with that of ferrous sulphate fortified products.

Figure 14 Response surface plot for fat acidity at 175°C roasting temperature for ferrous sulphate roasted blend

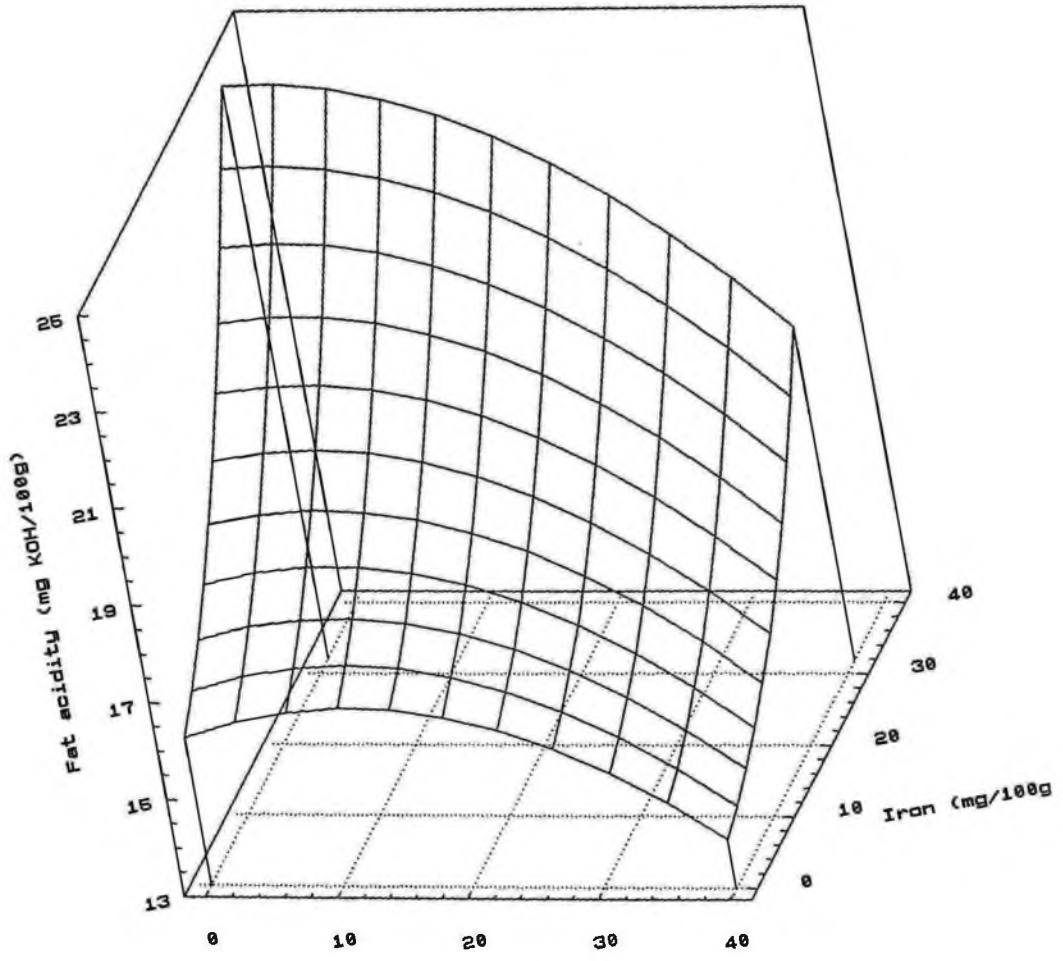
$$Z = 16.042347 + 0.095206X_1 + 0.146103X_2 - 0.00234X_1X_2 - 0.003657X_1^2 - 0.004046X_2^2$$

$$R^2 = 0.435$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature



Legume (%)

The response surface plot generated for water absorption at 30°C is as shown in Figure 15. Water absorbed at room temperature by the roasted blend was reduced as legume concentration was increased. This suggests that roasting of maize and cowpea/groundnut decreases the water absorption of the blends. This decrease may be attributed to denaturation and possible reduction in water binding sites in both protein and carbohydrates as a result of Maillard reaction. Sefa-Dedeh and Ampadu, (1991) reported the same observation in roasted maize/cowpea blend. Iron concentration however, had no effect on water absorption. A similar trend occurred for water absorption at 70°C. However there was increase when ferrous sulphate addition was increase at low legume whilst the converse occurs at high legume concentration (Figure 16). Legume concentration had significant effect on water absorption at room temperature, whilst legume concentration and its interaction with iron and roasting temperature affected water absorption at 70°C.

4.4.4 Apparent paste viscosity

The model was only adequate to predict hot paste viscosity for ferrous fumarate fortified product (Table 23). Cooled paste viscosity had all the models adequate that is for both ferrous salts fortified product (Table 23).

Roasting has been noted to have a slight viscosity-reducing effect on some cereals, but negligible in cereal-pulse mixtures (Nout, 1993).

Low viscosity was observed at both hot and cooled paste, which implies that roasting had effect on the cooked paste characteristics. Generally the longer the sample is roasted, the lower its cooked paste viscosity. This suggests that it is more difficult to cause gelling in the samples which have already receive a relatively longer heat treatment during roasting.

Figure 15 Response surface plot for water absorption at 30°C at 175°C roasting temperature for ferrous sulphate roasted blend.

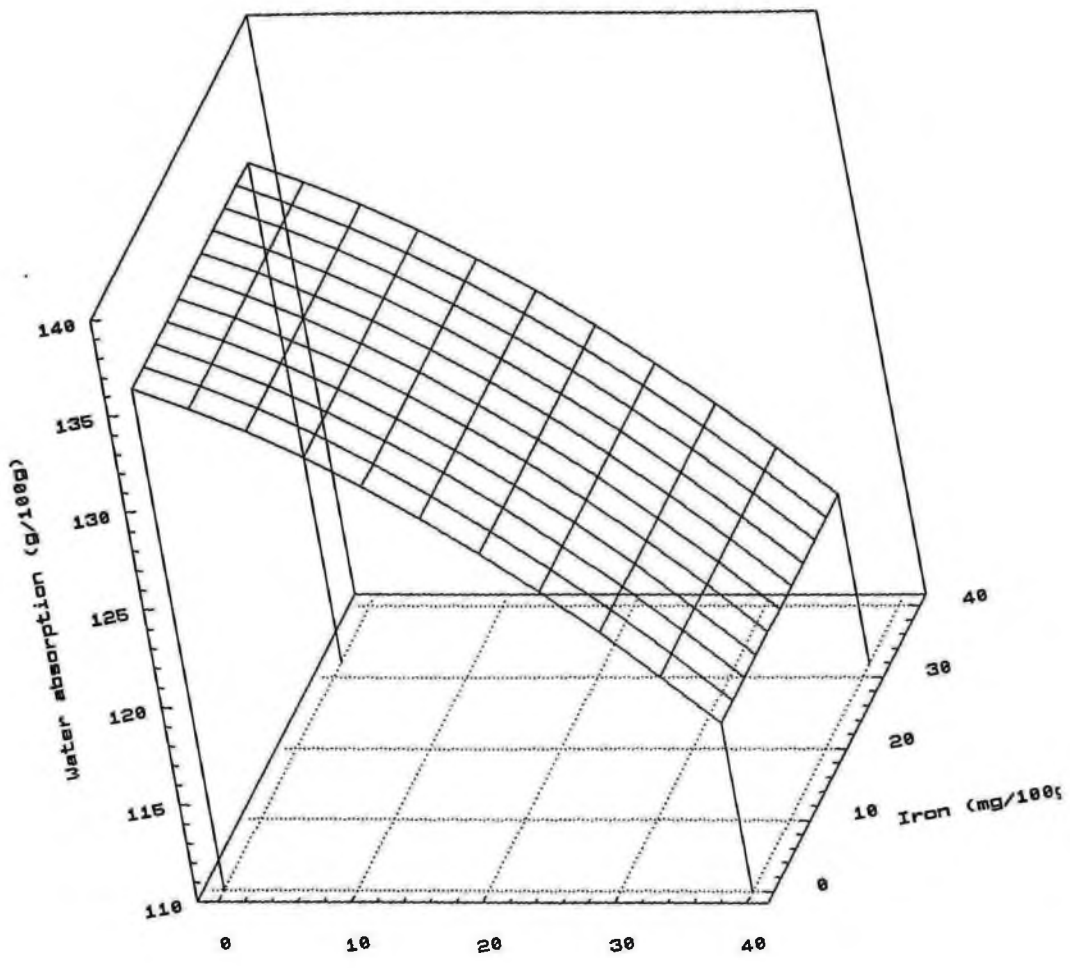
$$Z = 99.68414 + 0.799871X_1 + 0.20647X_3 - 0.005951X_1X_3 - 0.004694X_1^2$$

$$R^2 = 0.736$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature



Legume (%)

Figure 16 Response surface plot for water absorption at 70°C at 175°C roasting temperature for ferrous sulphate roasted blend.

A = Ferrous sulphate addition

$$Z = 150.108142 - 0.202676X_1 - 0.000882X_2X_3 - 0.000105X_1X_2X_3 + 0.012807X_2^2$$

$$R^2 = 0.839$$

B = Ferrous fumarate addition

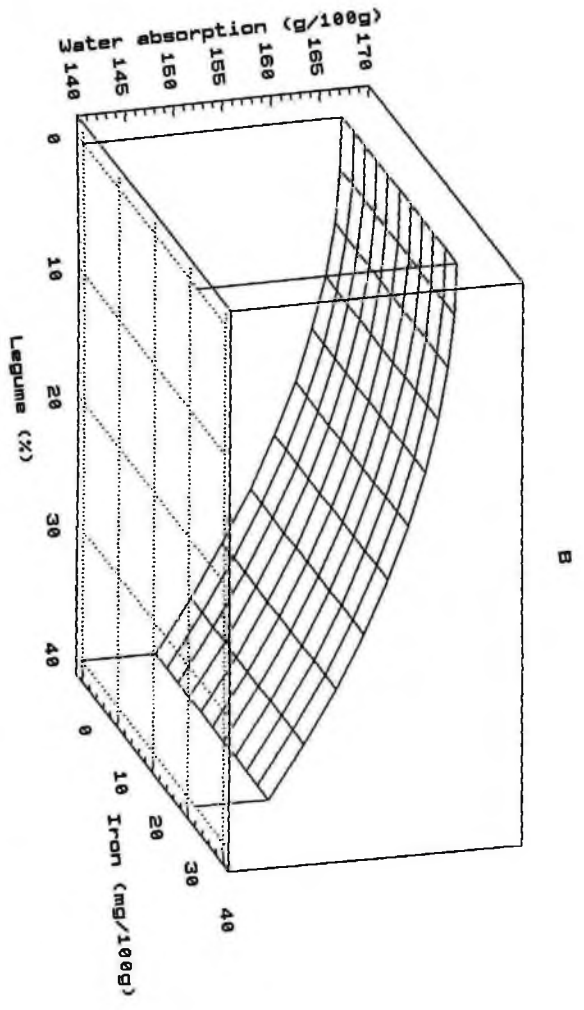
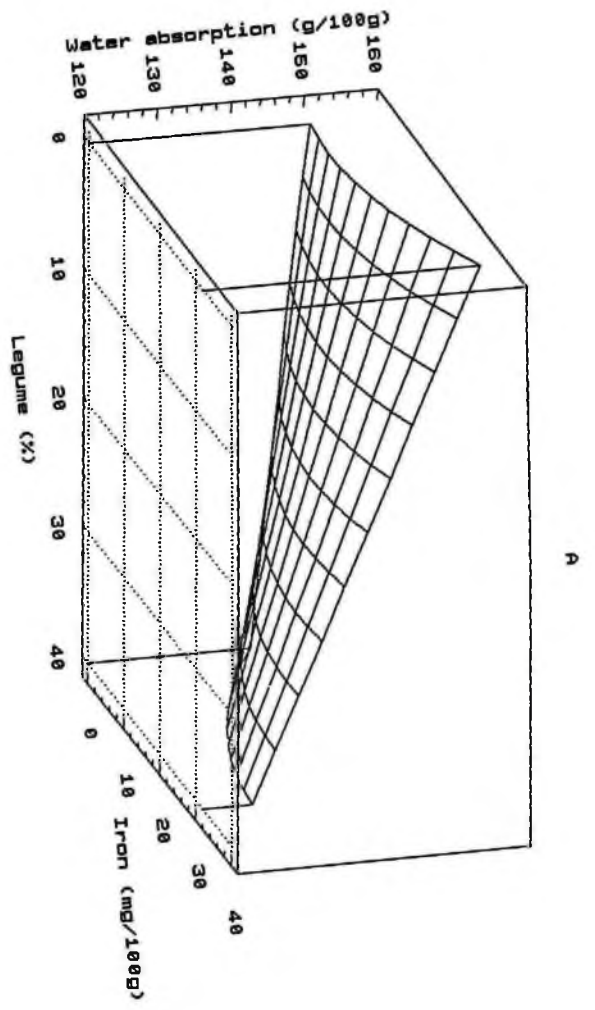
$$Z = -248.196408 + 4.436184X_3 - 0.011954X_1^2 - 0.011802X_3^2$$

$$R^2 = 0.267$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature



This was reported by Sefa-Dedeh and Ampadu, (1991). The effect of iron was dependent on the amount of legume added. Viscosity was higher when the paste was cooled to 50°C as compared to the hot paste at 90°C. This is due to the gelatinisation of the protein of the cowpea and the retrogradation of starch molecules.

Response surface plot in Figure 17 and 18 showed reduction with increase in legume concentration. Apparent paste viscosity was significantly influenced by legume concentration and roasting temperature. The interaction between legume and roasting temperature also had significant effect.

4.4.5 Energy content

The model was adequate to predict energy content for only ferrous fumarate fortified products (Table 23). Values obtained were between 469.6 and 518.6 kcal/100g and 448.0 and 518 kcal/100g for ferrous sulphate and ferrous fumarate fortified products respectively. The energy content of the products was higher than the recommended value for a quality weaning food (PAG, 1975). Quarshie, (1988) recorded an energy value of 461.3 kcal/100g sample for a similar product.

Figure 17 Response surface plot for apparent hot paste viscosity at 175°C roasting temperature for ferrous fumarate roasted blend.

$$Z = 76.009035 - 0.02048X_1X_3 + 0.062448X_1^2 + 0.008023X_2^2$$

$$R^2 = 0.625$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature

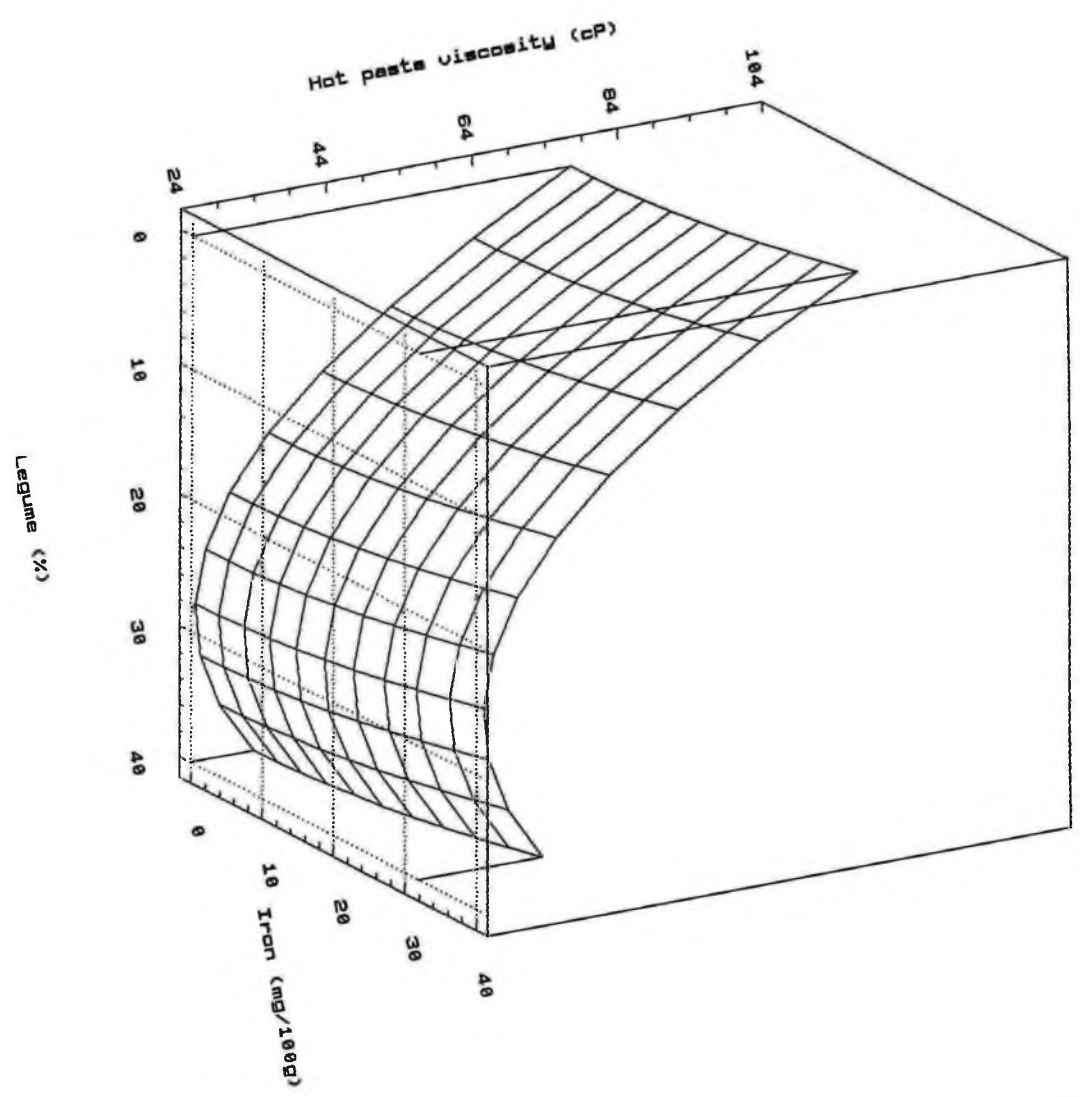


Figure 18 Response surface plot for apparent cooled paste viscosity at 175°C roasting temperature.

A = Ferrous sulphate addition

$$Z = -4641.068973 - 0.44.16124X_1 + 65.527756X_3 + 0.234132X_1X_3 - 0.000553X_1X_2X_3 - 0.208339X_3^2$$

$$R^2 = 0.444$$

B = Ferrous fumarate addition

$$Z = 364.925486 - 3.951138X_2 - 0.059773X_1X_3 + 0.18186X_1^2 - 0.167784X_2^2$$

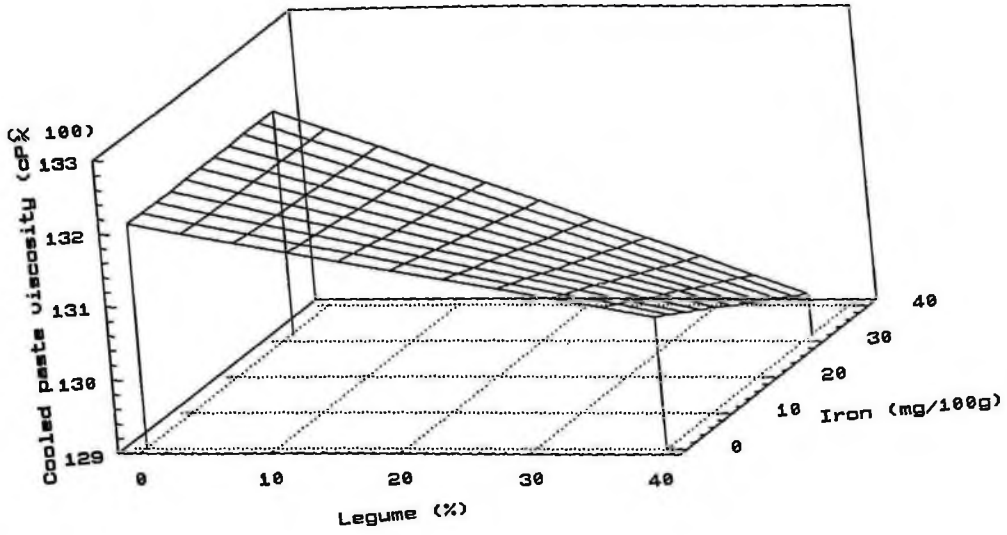
$$R^2 = 0.568$$

X_1 = Legume concentration

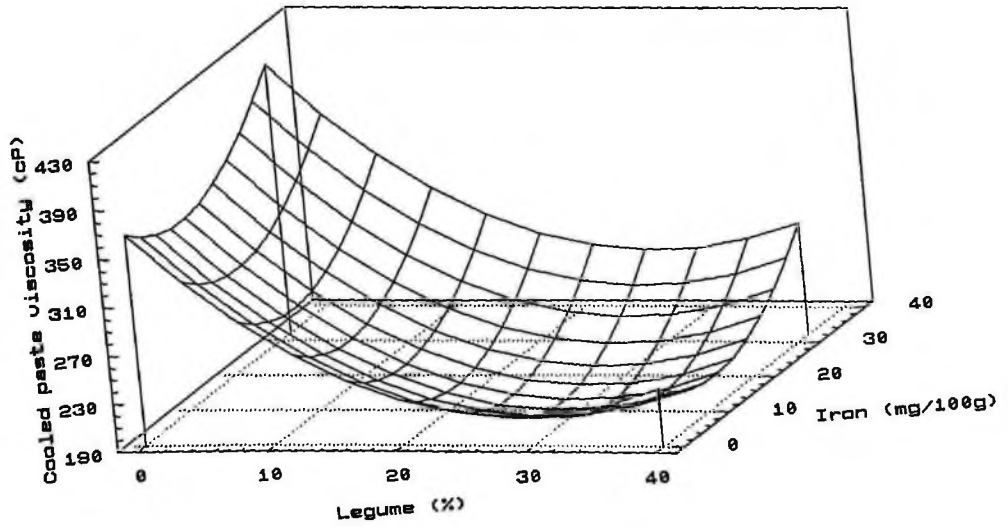
X_2 = Iron concentration

X_3 = Roasting temperature

A



B



Response surface plot generated in Figure 19 shown an increase in energy content as legume concentration was increased. This might be due to the inclusion of groundnut for its high oil content as the total legume concentration for the roasted blend. The energy content was significantly affected by legume concentration as observed on the graph and also roasting temperature. The effect of iron concentration however was not significant.

4.4.6 Total iron content

Addition of the ferrous salts was to increase the total iron content of the roasted blend. A work done by Quarshie (1988) indicated that the total iron content of a similar product was between 27 and 34.7mg/100g. Addition of the ferrous salt (0-32mg/100g) raised total iron content to between 67.3 and 78.5mg/100g as the highest values obtained for these products.

The models developed were adequate to predict total iron for all the samples (Table 23).

From the response surface plot shown in Figure 20, iron addition increases the total iron content. Addition of legume generally affected total iron negatively. This was more affected when ferrous sulphate was used as shown in Figure 20A. This implies that higher increase in total iron can be achieved at lower legume concentration with iron addition.

Iron and interaction between iron and roasting temperature had significant influence on total iron content of fortified roasted blend.

Figure 19 Response surface plot for energy content at 175°C roasting temperature for ferrous fumarate roasted blend

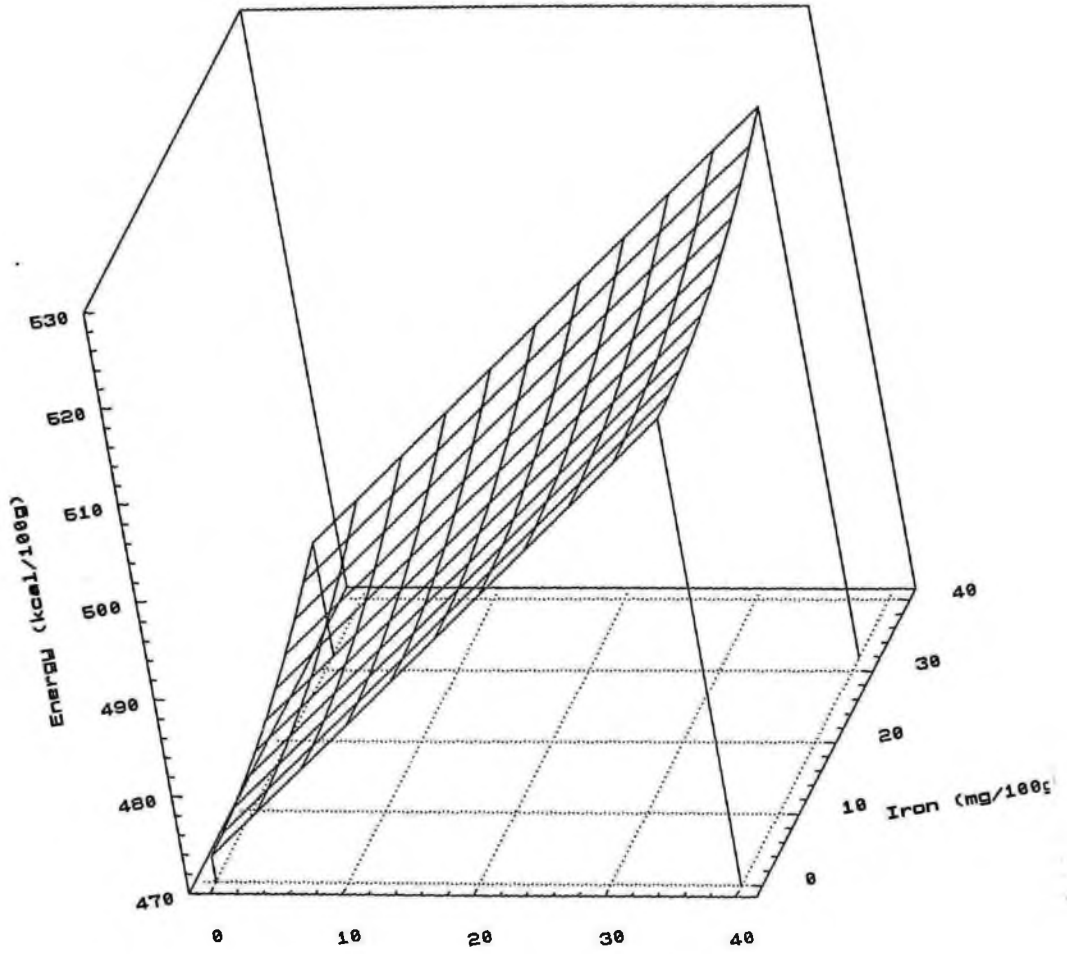
$$Z = -485.788805 + 10.683608X_3 + 0.006448X_1X_3 + 0.008586X_2^2 - 0.029752X_3^2$$

$$R^2 = 0.34$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature



Legume (%)

Figure 20 Response surface plot for total iron content at 175°C roasting temperature

A = Ferrous sulphate addition

$$Z = 13.04124 - 0.693073X_1 + 2.566109X_2 - 0.029354X_1X_2 \\ - 0.004587X_2X_3$$

$$R^2 = 0.389$$

B = Ferrous fumarate addition

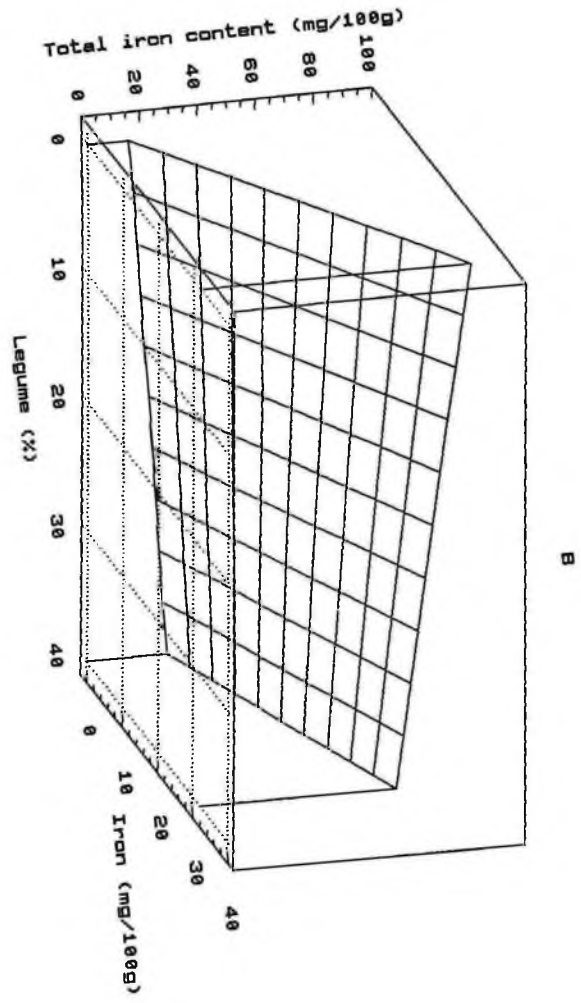
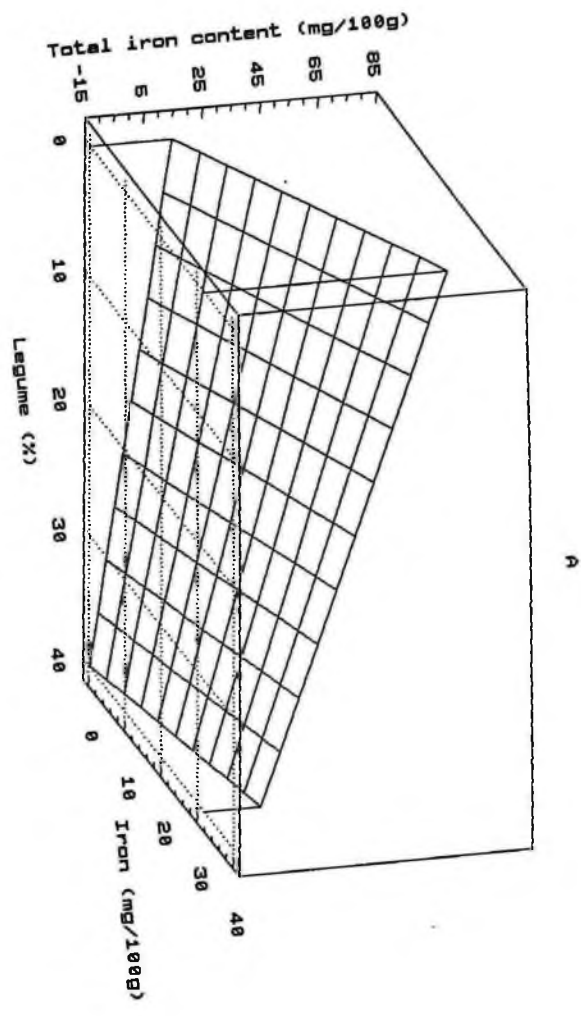
$$Z = 31.297448 + 0.329971X_1 - 0.097477X_3 + 0.013986X_2X_3 \\ - 0.00017X_1X_2X_3$$

$$R^2 = 0.615$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature



4.5 **Storage stability of iron-fortified roasted blend**

The products were stored for 4 months to investigate the effect of legume and iron concentration on the characteristic of the roasted blend.

4.5.1 **Fat acidity**

The model could be used to predict fat acidity for ferrous fumarate fortified products (Table 23). During storage, fat acidity decreases with increase in legume concentration. This is shown a response surface plot in Figure 21.

The quadratic term of the legume concentration had significant effect on fat acidity during storage. Also, an interaction between legume and iron on one hand and legume and roasting temperature on the other hand had significant effect on fat acidity.

4.5.2 **Water absorption index**

The models were adequate for water absorption at 30°C and 70°C for all the ferrous salt fortified products (Table 23).

From the response surface plot generated, increase in legume concentration reduces water absorption during storage (Figures 22 and 23).

Water absorption was significantly influenced by legume, its quadratic term and interaction with roasting temperature. This implies that even during storage iron addition has no role as far water absorption index is concerned for a roasted weaning blend.

Figure 21 Response surface plot for fat acidity at 175°C roasting temperature for ferrous fumarate roasted blend at 4 months storage

$$Z = 39.250941 - 0.24124X_3 - 0.002227X_1X_2 - 0.001485X_1X_3 + 0.004158X_1^2 + 0.000771X_3^2$$

$$R^2 = 0.791$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature

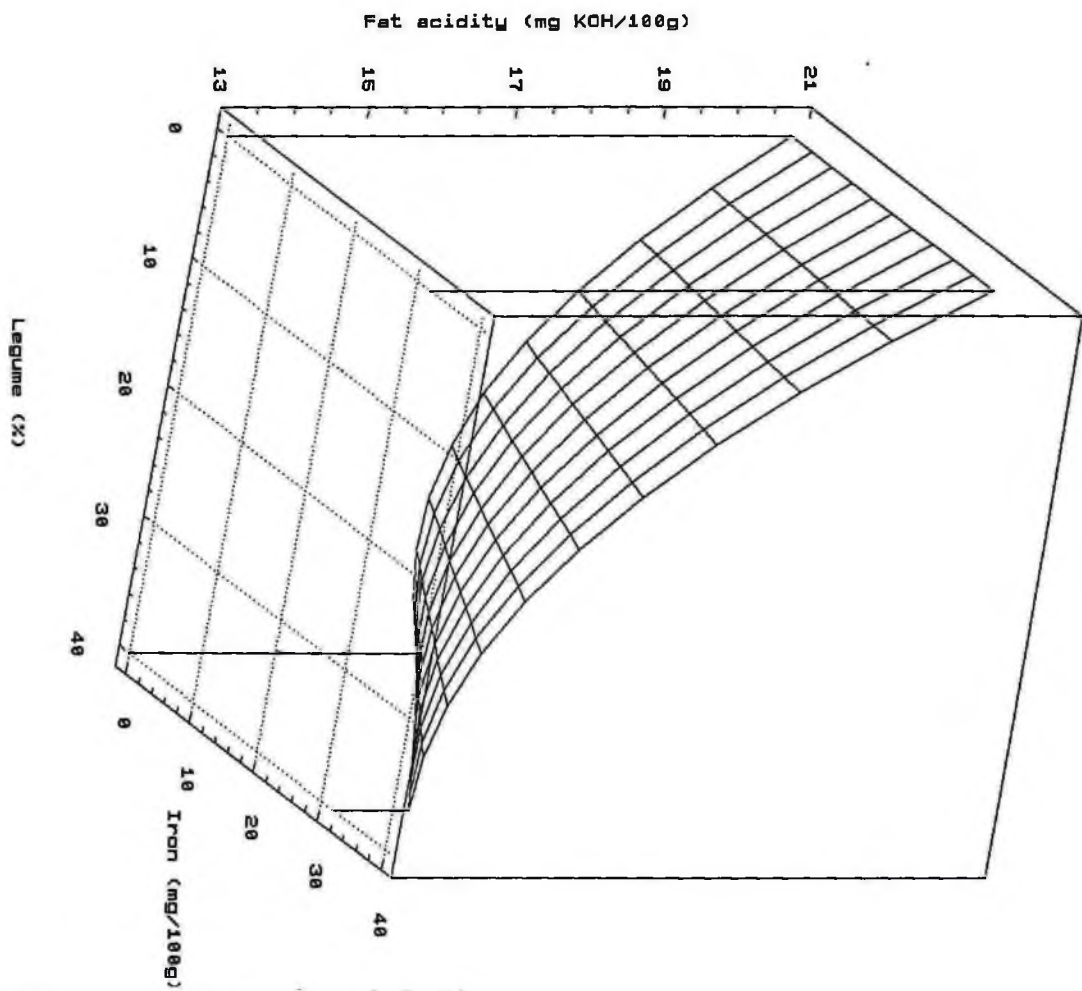


Figure 22 Response surface plot for water absorption at 30°C at 175°C roasting temperature at 4 months storage

A = Ferrous sulphate addition

$$Z = 126.255789 - 0.43182X_1 + 0.054082X_3 + 0.004319X_2^2$$

$$R^2 = 0.636$$

B = Ferrous fumarate addition

$$Z = 131.11459 + 0.001657X_1X_3 - 0.014218X_1^2 - 0.001754X_2^2$$

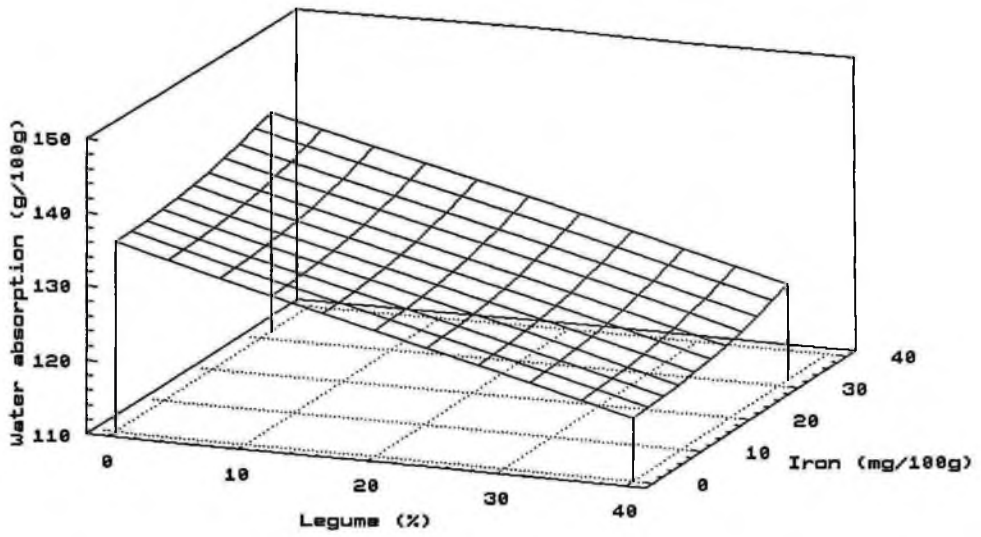
$$R^2 = 0.379$$

X_1 = Legume concentration

X_2 = Iron concentration

X_3 = Roasting temperature

A



B

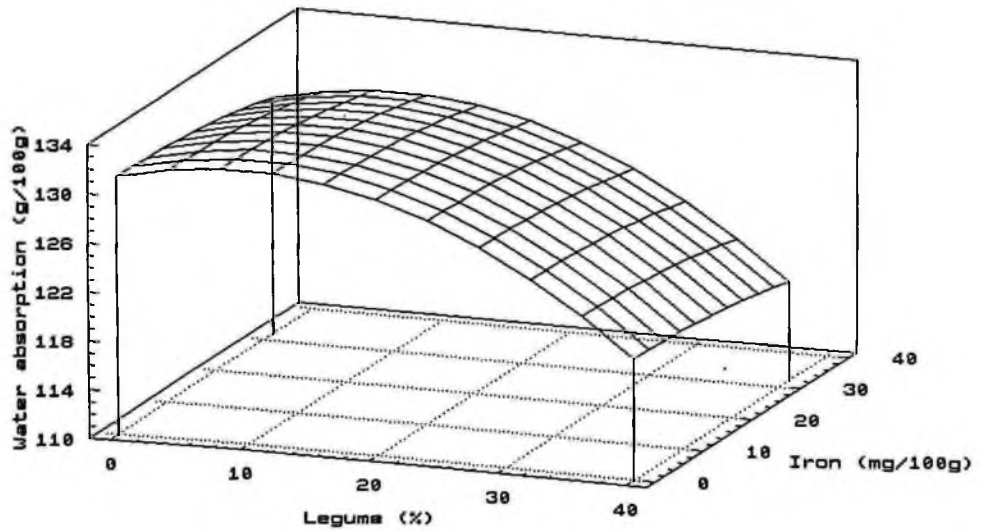


Figure 23 Response surface plot for water absorption at 70°C at 175°C roasting temperature at 4 months storage

A = Ferrous sulphate addition

$$Z = 8.026456 + 1.694923X_3 - 0.002734X_1X_3 - 0.000012X_1X_2X_3 - 0.004974X_3^2$$

$$R^2 = 0.849$$

B = Ferrous fumarate addition

$$Z = 161.378734 - 0.236212X_1 - 0.755716X_2 - 0.001951X_1X_3 - 0.016516X_2^2$$

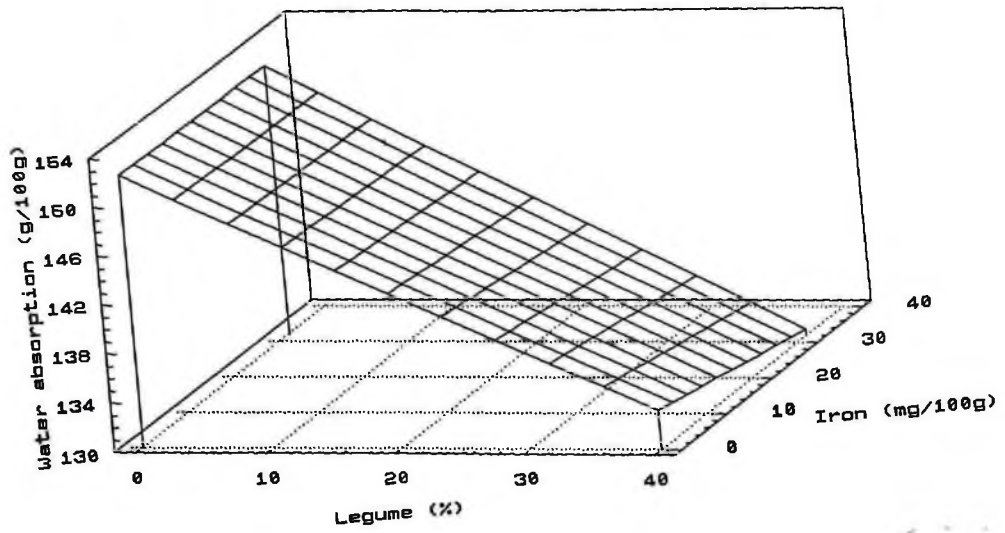
$$R^2 = 0.717$$

X_1 = Legume concentration

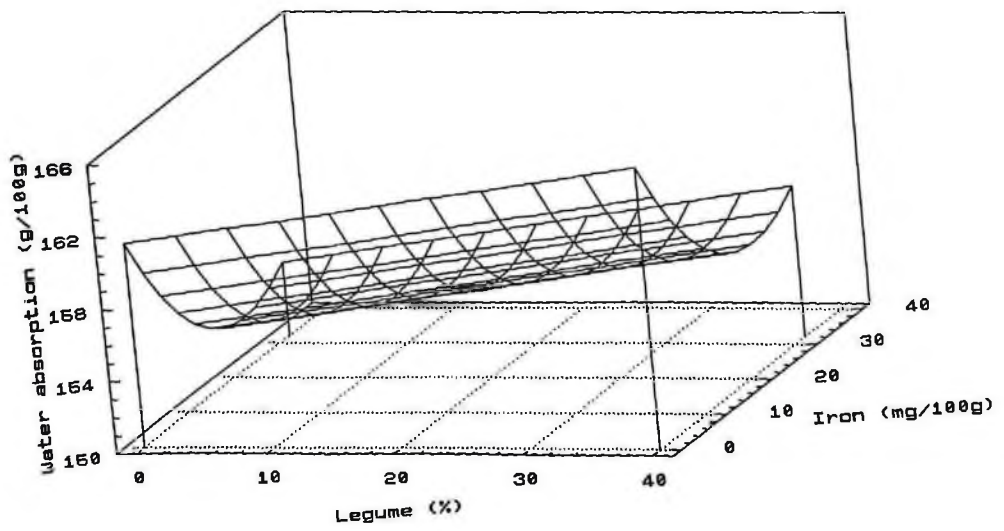
X_2 = Iron concentration

X_3 = Roasting temperature

A



B



4.6 Sensory evaluation through field trials

4.6.1 Background data on respondents

A total of seventy-two mothers participated in the evaluation of the iron-fortified and non-fortified products. These mothers were drawn from five communities in the Central region namely: Cape Coast, Abandze, Taido Anomabo, Pomaase and Assin Jakai. These communities were used because the investigator was involved in the national anaemia survey and developmental projects initiated by The Hunger Project, Ghana in these communities. This made community entry relatively easy. It was proposed to undertake this project as community-based; hence four rural and one urban communities were used. Mothers at the Child Welfare Clinic (CWC) in Cape Coast and the communities were used for this exercise because they are infant-food-purchasing population.

About 45% of the respondents had at least basic education, whilst 24% had only primary and 28% completely illiterate (Figure 24A). Twenty-five percent of the mothers were under 20 years, whilst about 57% were under 25 years of age. Only 4% of them were above 35 years (Figure 24B).

The primary occupation of the respondents was mainly farming (34.7%) and trading (27.8). Almost all the respondents were self-employed with only 1.4% as salary workers (Figure 24C). The economic activities of a mother are important in child nutrition. This is because the primary responsibility of a child lies on the woman. Besides, the educational and economic activities of mothers mostly influence the choice of food even for their children. Most mothers (80%) used in the evaluation had a child less than one year old (Table 24). For a weaning food that is targeted at children, then this group of mothers was most suitable.

Table 24 Age of last child of mothers

Age group (months)	No. of mothers	% of mothers
0-5	26	36.1
6-12	32	44.4
13-18	3	4.2
19-24	7	9.2
>24	4	5.6
Total	72	100

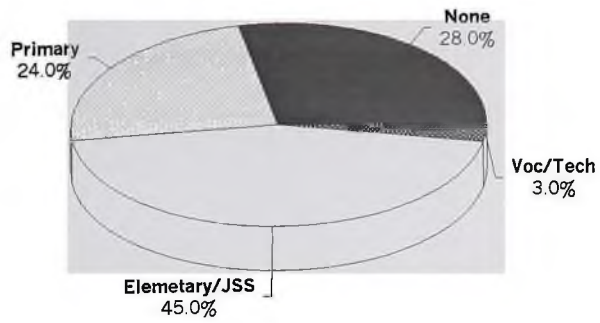
4.6.2 Sensory evaluation of some quality attributes

This was aimed at finding out whether mothers could differentiate between the iron-fortified and non-fortified samples in attributes such as colour, taste, flavour as well as the general acceptability. Products were evaluated against some proprietary weaning foods such as *Frisocreme*, compared to maize and maize/cowpea dough and *Cerelac*, compared to roasted maize/cowpea/groundnut blend because of their colour. Friedmann's table was used for the analysis.

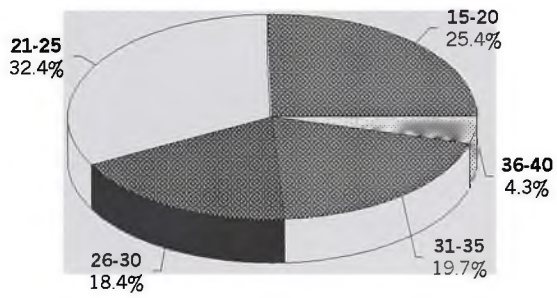
Many biases due to the difference in the ages of the children of the mothers evaluating the products for desirable consistency were noted. This was corrected by preparing the porridge at a constant consistency throughout the evaluation.

Figure 24 The profile of respondents: Educational background (A), age group distribution (B) and primary occupation (C)

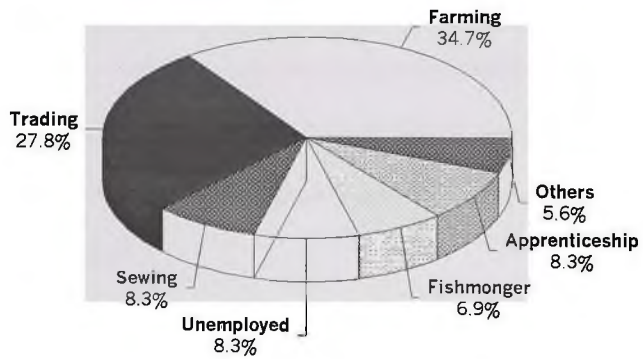
A



B



C



Analysis revealed that the proprietary weaning foods, that is, Frisocrema and Cerelac were the most preferred for all the attributes, that is, colour, flavour and taste (Tables 25, 26 and 27). They were also the most accepted as against the test samples. This may be due to perhaps ingredients such as milk, some sugar and some flavour they might contain and also the particle size.

(a) Fermented maize dough

In terms of colour, the iron-fortified products were preferred to the non-fortified one (Table 25). Taste followed the same trend as colour. Even though the fortified were more preferred, there was no difference between the non-fortified and the 16mg ferrous fumarate addition detected as far as flavour was concerned.

In general, the iron-fortified fermented dough was accepted as compared to the non-fortified product. However, there was no differences between the non-fortified and 16mg ferrous salt fortified product with respect to general acceptability. Rank sums are shown in Table 25.

(b) Fermented maize/cowpea dough

Non-fortified product was ranked third in terms of colour against the test samples. Meanwhile, differences could not be found between the non-fortified and 16mg ferrous fortified product. The position of non-fortified in terms of flavour was the same as colour. There were no differences in all the samples as far as flavour and taste are concerned and therefore acceptability (Table 26).

Table 25 Rank sums of sensory evaluation on fortified and non-fortified fermented maize dough porridges

Attributes	Samples					
	Fri	OF	16FS	16FF	32FS	32FF
Colour	87	284 ^a	259 ^a	263 ^a	249 ^a	223
Flavour	79	296 ^b	249	277 ^b	241	223
Taste	81	290 ^c	264 ^c	248 ^c	247 ^c	233
General Acceptability	89	296 ^d	262 ^d	261 ^d	230	227

*Rank sums of the same letter are not significantly different

Table 26 Rank sums of sensory evaluation on fortified and non-fortified fermented maize/cowpea dough porridges

Attributes	Samples					
	Fri	OF	16FS	16FF	32FS	32FF
Colour	110	269 ^a	316	281 ^a	265 ^a	263 ^a
Flavour	93	290 ^b	304 ^b	275 ^b	278 ^b	272 ^b
Taste	94	284 ^c	275 ^c	293 ^c	284 ^c	282 ^c
General Acceptability	101	281 ^d	287 ^d	278 ^d	292 ^d	281 ^d

*Rank sums of the same letter are not significantly different

Fri = Frisocrema

OF = Non-fortified products

16FS = 16mg ferrous sulphate fortified products

16FF = 16mg ferrous fumarate fortified products

32FS = 32mg ferrous sulphate fortified products

32FF = 32mg ferrous fumarate fortified products

Table 25 Rank sums of sensory evaluation on fortified and non-fortified fermented maize dough porridges

Attributes	Samples					
	Fri	OF	16FS	16FF	32FS	32FF
Colour	87	284 <i>a</i>	259 <i>a</i>	263 <i>a</i>	249 <i>a</i>	223
Flavour	79	296 <i>b</i>	249	277 <i>b</i>	241	223
Taste	81	290 <i>c</i>	264 <i>c</i>	248 <i>c</i>	247 <i>c</i>	233
General Acceptability	89	296 <i>d</i>	262 <i>d</i>	261 <i>d</i>	230	227

*Rank sums of the same letter are not significantly different

Table 26 Rank sums of sensory evaluation on fortified and non-fortified fermented maize/cowpea dough porridges

Attributes	Samples					
	Fri	OF	16FS	16FF	32FS	32FF
Colour	110	269 <i>a</i>	316	281 <i>a</i>	265 <i>a</i>	263 <i>a</i>
Flavour	93	290 <i>b</i>	304 <i>b</i>	275 <i>b</i>	278 <i>b</i>	272 <i>b</i>
Taste	94	284 <i>c</i>	275 <i>c</i>	293 <i>c</i>	284 <i>c</i>	282 <i>c</i>
General Acceptability	101	281 <i>d</i>	287 <i>d</i>	278 <i>d</i>	292 <i>d</i>	281 <i>d</i>

*Rank sums of the same letter are not significantly different

Fri = Frisocrema

OF = Non-fortified products

16FS = 16mg ferrous sulphate fortified products

16FF = 16mg ferrous fumarate fortified products

32FS = 32mg ferrous sulphate fortified products

32FF = 32mg ferrous fumarate fortified products

(c) Roasted maize/cowpea/groundnut blend

Panellists could not find a difference between the non-fortified product and those fortified with ferrous fumarate irrespective of the fortification level. The roasted blend had brownish colour, likewise the ferrous fumarate compound and therefore this could be the reason why differences could not be made (Table 27)

Table 27 Rank sums of sensory evaluation on fortified and non-fortified roasted maize/cowpea/groundnut blend porridges

Attributes	Samples					
	Cer	OF	16FS	16FF	32FS	32FF
Colour	133	241 <i>a</i>	330	237 <i>a</i>	306	265 <i>a</i>
Flavour	105	249 <i>b</i>	320	279 <i>b</i>	295	258 <i>b</i>
Taste	99	254 <i>c</i>	322	269 <i>c</i>	331	253 <i>c</i>
General Acceptability	101	251 <i>d</i>	329	267 <i>d</i>	298	272 <i>d</i>

*Rank sums of the same letter are not significantly different

Cer = Cerelac

OF = Non-fortified products

16FS = 16mg ferrous sulphate fortified products

16FF = 16mg ferrous fumarate fortified products

32FS = 32mg ferrous sulphate fortified products

32FF = 32mg ferrous fumarate fortified products

When respondents were asked to describe the products, about 72% could liken both the iron-fortified and the non-fortified products of the fermented maize and fermented maize/cowpea dough. Work done by Kluvitse (1995) revealed that as many as 69% of mothers fed their infants on traditional fermented maize dough porridge during the weaning period. This means

that, since most of the mothers are used to fermented maize dough as a weaning food, their assertion that both iron-fortified and the non-fortified products were the same as the traditional fermented maize porridge could not be challenged.

About 72% of the mothers could liken the iron-fortified products of the roasted blend to the cereal-legume blend “weanimix” being promoted by the Ministry of Health.

4.6.3 Cost

Respondents gave various prices they could afford to pay for the test products. An equivalent amount of 250g of product based on the quantity of product measured with a 450g margarine tin was used since most mothers are used to this measure. Nearly 40% of the respondents could pay ₦500.00 for each of the three products tested, whilst about 15% mentioned ₦1,000.00 (Table 28). Most mothers found it difficult pricing the products since in their opinion, a lot would have to be taken into consideration. It could be deduced from their expressions that a 250g packaged product could attract ₦1,000.00 or more.

The production cost for a 250g sample was estimated to be about ₦925.00. It is possible that a large-scale production of these products may reduce the cost further.

Table 28 Estimated price for 250g of product by respondents

Price (¢)	Dehydrated fermented maize dough		Dehydrated fermented maize/cowpea dough		Roasted maize/cowpea/groundnut blend	
	Number	%	Number	%	Number	%
200	13	18.1	9	12.5	12	16.7
300	5	6.9	8	11.1	4	5.6
400	8	11.1	7	9.7	7	9.7
500	28	38.9	29	40.3	27	37.5
600	1	1.4	1	1.4	4	5.6
700	-	-	-	-	1	1.4
800	2	2.8	3	4.2	1	1.4
1000	14	19.4	12	16.7	11	15.3
1200	-	-	-	-	1	1.4
1500	1	1.4	3	4.2	4	5.6

5.0 CONCLUSIONS

5.1 **Effect of fermentation and drying on iron fortification of fermented maize dough**

Iron fortification and drying brought about relative increase in pH. Lowest pH for non-fortified sample was 3.34 whilst 16mg and 32mg iron fortification recorded 3.51 and 4.11 respectively. There was however, no significant difference between the pH of iron-fortified and non-fortified fermented maize dough. These are levels believed to be optimal for cereal phytase. Using pH as a factor for iron availability in iron-fortified products, iron can be incorporated into products fermented between 36 and 60 hours irrespective of the iron compounds or the state of the dough. The 16mg iron fortification is suitable as far as pH is concerned.

Rancidity was not a problem, even though fat acidity increases with fermentation in both iron-fortified and non-fortified products and also when ferrous salts were added after fermentation. Fermentation and iron addition increased iron content of the products. Energy of at least 460 kcal/100g was recorded for all samples. This is higher than the recommended energy value for a quality weaning food. Fermentation and addition of iron had lowering effect on water absorption and viscosity.

5.2 **Microbiological evaluation**

Iron levels used in the fermenting medium did not interfere with the microbial growth. This signifies that the levels added were not toxic to the microorganisms. The microorganisms were predominantly lactic acid bacteria, yeast and moulds.

The ability of iron-fortified maize dough system to ferment and produce acids comparable to the traditional maize dough system is beneficial. In addition, the fortified maize has antimicrobial properties and high iron content and therefore is a product, which may address diarrhoeal diseases and iron deficiency anaemia.

5.3 Response surface methodology study on roasted maize/cowpea/groundnut blend

Regression models developed to predict product characteristics of roasted weaning food were influenced by the process variables. These were mainly legume concentration and roasting temperature. Iron addition however, affected the characteristics when legume concentration was high.

Reduction of lightness of colour was mainly due to increase in legume concentration rather than the iron at levels used. Increase in fat acidity and therefore breakdown of triglycerides, yielding free fatty acids was due to iron concentration, whilst a decreasing effect was observed at higher legume concentration. Viscosity was lowered with addition of roasted legume. Energy content was increased by the addition of legume (cowpea/groundnut). Total iron content was significantly lowered by increase in legume concentration, even at higher iron concentration.

5.4 Shelf stability

Increase in pH was recorded over the period of storage. There was 100% increase in fat acidity for both iron-fortified and non-fortified fermented maize dough over the storage period of 4 months. Lightness of colour was also reduced over storage. There was no

significant effect of storage on water absorption. Storage generally affected most of the characteristics for both iron-fortified and non-fortified fermented maize dough, however, not to the detriment of product quality. Characteristics of the roasted maize/cowpea/groundnut blend at storage were significantly affected by legume concentration and roasting temperature.

5.5 Sensory evaluation/consumer acceptability

Consumer acceptability was performed on 48 hours fermented cooked maize and maize/cowpea dough, and roasted maize/cowpea/groundnut blend (175°C). About 72% of respondents could not differentiate between the iron-fortified and non-fortified products.

Iron-fortified fermented maize dough was most preferred. All the products of fermented maize/cowpea dough prepared with different ferrous salts and levels were accepted on equal strength. Panelists did not detect any difference in the roasted maize/cowpea/groundnut blend. The iron-fortified samples of all the three products presented were generally acceptable to mothers in the five communities in the Central region.

The price quoted for a 250g sample was five hundred cedis (¢500.00) for all the products by the panelists.

Further works

1. The efficacy of the iron-fortified products can be tested by feeding children on the products against the protection of iron deficiency anaemia.
2. Bioavailability studies can be carried out on the iron-fortified products.

REFERENCES

AACC. 1983. Approved methods of the American Association of Cereal Chemists. 8th Edition

Abbey, B. W. and Nkanga, U. B. 1988. Production of high quality of weaning products from maize-cowpea-crayfish mixture. *Nutr. Rep. Intern.* 37: 951-955

Ackom-Quayson, G. and Sefa-Dedeh, S. 1993. Optimisation of conditions for the fermentation of maize dough using response surface methodology presented at 13th National Maize And Legumes Workshop. Kumasi, Ghana. 30th March-1st April

Akinrele, I. A. and Bassir, O. 1967. The nutritive value of Ogi, a Nigerian infant food. *J. Trop. Med. Hyg.* 70: 279-280

Akinrele, I. A. 1970. Fermentation studies on maize during the preparation of a traditional African starch cake food. *J. Sci. Agric.*, 21; 619-625

Akinyele, I. O., Love, M. H. and Kinge, M. 1988. Nutrient composition of extruded cowpea products. *Int. J. Food Sci. Technol.* 23: 297-301

Akpanunam, M. A. and Sefa-Dedeh, S. 1995. Traditional lactic acid fermentation, malt addition and quality development in maize-cowpea weaning blends. *Food Nutr. Bull.* 16(1): 75-80

Ali, M. R. and Willis, R. G. H. 1983. Milling and cooking of hybrid sorghum grains. Qual. Plant Foods Hum. Nutr. 32:59

Ampadu, E. 1991. Soybean flour processing and incorporation into traditional food product: Case study - Maasa. M. Phil. Miniproject, Dept. of Nutrition and Food Science, University of Ghana, Legon

Ampadu, E. W. 1994. Soybean (*Glycine Max*) processing and performance in fermented maize dough system. M. Phil. Thesis. Dept. of Nutrition and Food Science, University of Ghana.

Anderson, R. 1985. Breakfast cereals and dry milled corn products. In: Iron fortification of foods. eds. Clydesdale, F. and Wiemer, K.L. Academic Press, New York

Anim, M. 1991. Effects of pre-fermentation conditions on characteristics of cereal-legume mixes. B.Sc. Project. Dept. of Nutrition and Food Science, Univ. of Ghana.

AOAC. 1990. Approved Official Methods of Analysis. 15th edition. Association of Official Analytical Chemists. Washington, D. C.

APHA (1976). Compendium of methods for the microbiological examination of foods. Byrd Press Inc.

Ashworth, A. and Draper, A. 1992. The potential of traditional technologies for increasing the energy density of weaning foods. A critical review of existing knowledge with particular reference to malting and fermentation. WHO/CBD/EDP/92.4.

Asibey-Berko, E. 1995. Workshop Report and Recommendation. In: Proceedings of the National Workshop on Iodine Deficiency Disorders in Ghana. Asibey-Berko, E. and Orraca-Tetteh, R. (eds)

Barampama, Z. and Simard, R. E. 1994. Oligosaccharides, antinutritional factors and protein digestibility of dry beans as affected by processing. *J. Food Sci.* 59(4): 833-838

Barret, F. and Ranum, P. 1985. Wheat flour and cereal based products. in: Iron fortification of foods. Eds: Clydesdale, F. M. and Wiemer, K.L. Academic Press, New York

Baynes, R. D. and Bothwell, T.H. 1990. Iron deficiency. *Annual Review of Nutrition* 10: 133-148

Borejszo, Z. and Khan, K. 1992. Reduction of flatulence-causing sugars by high temperature extrusion of pinto bean high starch fractions. *J. Food Sci.* 57(3): 771-772

Bothwell, T.H., Charlton, R. W., Cook, J. and Finch, C. A. (1979). *Iron Metabolism in man.* Blackwell Scientific Publication, Oxford.

Bouis, H. and Slack, A. 1996. Food Policy Perspectives in evaluating alternative Interventions for reducing micronutrient malnutrition. In: Food-Based Approaches to preventing micronutrient malnutrition, Combs, G.F., Welch, R.M., Duxbury, J.M., Uphoff, N. T. and Nesheim, M.C. (eds) Ithaca, New York

Bressani, R. 1985. In: Cowpea Research, Production and Utilisation. Singh, S. R. (ed). John Wiley and Sons, Chichester pp 353-359

Bressani, R. and Elias, L. G. 1983. Guidelines for the development of processed and packaged weaning foods. *UNU Food Nutr. Bull.* 5(3), 57-64

Brown, K. H. 1991. The importance of dietary quantity versus quality for weanlings in less developed countries: A framework for discussion. *Food Nutr. Bull.* 13(2): 86-94

Cameron, N. and Hofvander, Y. 1971. Manual on feeding infants and young children. Advisory group of the UN systems

Chaberek, S. and Martell, A.E. 1959. Organic Sequestering Agents. John Wiley and Sons, Inc., New York

Christian, W.F.K. 1970. Lactic acid bacteria in fermenting maize dough. *Ghana J. Sci.* 10: 22-28

Clydesdale, F.M. 1991. Mineral Additives in: Nutrients Additions to food Nutritional, Technological and regulatory Aspects ed: Bauernfeind, J.C. and Lachance, P.A. Food and Nutrition Press, Inc,

Clydesdale, F.M. 1989. The release of mineral chemistry to bioavailability. Nutr. Today 24(2) 23-30

Cocodrilli, G.J. and Shah, N. 1985. Beverages. in: Iron fortification of foods. eds. Clydesdale, F. and Wiemer, K.L. Academic Press, New York

Cochran, W.G. and Cox, G.M. 1957. in: Experimental Design. 2nd edition. Publ. John Wiley and Sons, Inc., N.Y

Combs, G.F., Welch, R.M., Duxbury, J.M., Uphoff, N.T. and Nesheim, M.C. (eds) , 1996. Food-Based Approaches to preventing micronutrient malnutrition: An International Research agenda. Ithaca, New York

Committee on Food Protection, Food and Nutrition Board. 1975. Technology of Fortification of foods. Proc. of a Workshop of the Subcommittee on Food Technology. National Academy of Sciences. Washington

Cook, J. D. And Reusser, M. E. (1983). Iron fortification: An Update. Am. J. Clin. Nutr. 38, 648-659.

Desikacher, H. S. R. 1980. Development of weaning foods with high caloric density and low paste viscosity using traditional technologies. *Food and Nutr. Bull.* 2: 21-23

Dijkhuizen, P. 1989. Weaning food: a formula for success. *Dev. J.* 1: 33-35.

Dovlo, F. E., Williams, C. E. and Zoaka, L. 1976. Cowpeas: Home preparation and use in West Africa. International Development Research Centre (IDRC-055E), Ottawa, Canada

Draper, A. 1996. Street foods in developing countries. The potential for micronutrient fortification. USAID 5122 - C - 00 - 3025 - 00

Duszkiewicz-Reinhard, W., Gujska, E. and Khan, K. 1994. Reduction of stachyose in legume flours by lactic acid bacteria. *J. Food Sci.* 59(1): 115-117

Eyeson, K. K. And Plahar, W. A. (1988). Available Resources for weaning food formulation in Ghana. A paper prepared for presentation at a seminar on the development of infant and weaning food based on cereals and legumes at ITA, Dakar, Senegal.

FAO(Food and Agriculture Organisation/World Health Organisation). (1988). Requirements of Vitamin A, Iron, folate and Vitamin B12. FAO Food and Nutrition Series 32, FAO, Rome, Italy.

FAO/WHO.. Expert Committee on Nutrition. 1971. Food fortification; Protein-calorie malnutrition. 8th Report. WHO, Geneva. pp 8-33

FAO/WHO. 1992. International Conference of Nutrition: Final Report of the Conference, Rome:

Fields, M.L., Hamed, A. M. and Smith, D. .K. 1981. Natural lactic acid fermentation of corn meal. *J. Food Sci.* 46: 900-902

Frimpong, K. 1995. Cowpea fortification of traditional foods. M. Phil. Thesis. Dept. of Nutrition and Food Science, University of Ghana.

Gavin, M. W., McCarthy, D. M. and Garry, P. J. 1994. Evidence that iron stores regulate iron absorption - a setpoint. *Am. J. Clin. Nutr.*, 59. 1376-1380

Ghana Statistical Service (GSS) and Macro International Inc. (MI). 1994. Ghana Demographic and Household Survey, 1993. Calverton, Maryland: GSS and MI

Ghana VAST Study Team. 1993. Vitamin A supplementation in Northern Ghana: Effects on clinic attendance's, hospital admissions, and child mortality. *Lancet* 342: 7-12

Gibson, R. S. 1996. Monitoring the progress and evaluating the impact of food-based strategies for combating micronutrient deficiencies. In: *Food-Based Approaches to preventing micronutrient malnutrition*, Combs, G.F., Welch, R.M., Duxbury, J.M., Uphoff, N.T. and Nesheim, M.C. (eds) Ithaca, New York

Hallberg , L. 1992. Iron nutrition and food iron fortification. *Sem. Hematol.* 19: 31-41

Hallberg, L., Sandstrom, B. and Aggett, P.J. 1993. Iron, zinc and other trace elements. in: *Human nutrition and dietetics*. Ed: Passmore, R. and Eastwood. M. A. 8th Ed. Churchill Livingstone

Halm, M., Lillie, A., Sorensen, A. K. and Jakobsen, M. 1993. Microbiological and aromatic characteristics of fermented maize doughs for kenkey production in Ghana. *Int. J. Food Microbiol.* 19: 135-143

Hans, I. K., Kim, S. W., Chae, B. J., Ha, J. K., Park, Y. J. and Lee, Y. S. 1988. Bioavailability of amino acids in cereals and legumes. In proceedings, Fifth Asian Congress of Nutrition, Osaka, Japan, 26-29 Oct. 1987. Eds. Yasumoto, K., Hokawa, Y. Koishia, H. and Sanno, Y. pp. 351-354. Tokyo Centre for Academic Publications

Hathcock, J.N. 1985. Quantitative evaluation of vitamin safety. *Pharmacy Times*, 51(5), 104-113

Hiel, A. M. M. 1984. Young children and supplementary feeding programmes: a field guide for the use of local foods and dried skim milk in Africa. Wageningen: The Netherlands Nutrition Foundation

Hoover, R. and Sosulski, F. W. 1990. Composition, structure , functionality and chemical modification of legume and starches; a review. *Canadian Journal of Physiology and Pharmacology* 69: 79-92

Hurrell, R. F. 1990. Combating iron deficiency anaemia through food fortification technology. International Nutrition Anaemia Consultative Group (INACG) Conference, Washington, D. C

Hurrell, R.F. (1992). Prospects for improving the iron fortification of foods. In: Nutritional anaemias. Fomon, S. J. and Zlotkin, S. H. (eds).. New York: Raven Press.

Ihekoronye, A. I. And Ngoddy, P. O. 1985. Cereal crops. In: *Integrated Food Science and Technology for the Tropics*. MacMillan Publishers, London, pp. 236-265

JECFA (Joint FAO/WHO Expert Committee on Food Additives). 1993. Summary and conclusions. 41st Meeting, Geneva, 9-18 February

Jellinek, G. 1985. *Sensory Evaluation of food. Theory and Practice*. International Publishers in Science and Technology

Kivisto, B., Andersson, H., Cederblad, G., Sandberg, A.S. and Sandstrom, B. 1986. Extrusion cooking of high-fibre cereal product. *Br. J. Nutr.* 55, 255-260

Klavitse, Y. M. 1995. Development of cowpea-fortified weaning food: Consumer expectations, functional and chemical properties. M. Phil. Thesis. Dept. of Nutrition and Food Science, University of Ghana.

Lachance, P.A. and Bauernfeind, J.C. 1991. Concepts and Practices of Nutrifying foods: In: Nutrients Additions to food Nutritional, Technological and regulatory Aspects ed: Bauernfeind, J.C. and Lachance, P.A. Food and Nutrition Press, Inc,

Lee, K. and Clydesdale, F.M. 1980. Effect of baking on the forms of iron-enriched flour. J. Food Sci. 45; 1500-1504

Lee, K. and Clydesdale, F.M. 1981. The effect of thermal processing on the endogenous and added iron in canned spinach. J. Food Sci. 46, 1064-1068

Lofti, M., Mannar, M. G. V., Merx, R. J. H. M. And Naber-van den Heuvel, P. 1996. Micronutrient Fortification of foods, Current practices, research and opportunities. MI, IAC.

Lopez, Y., Gordon, D. T. and Fields, M. L. 1983. Release of phosphorus from phytate by natural lactic fermentation. J. Food Sci. 48: 953-954

Lorri, W.S.M. 1993. Nutritional and Microbiological Evaluation of Fermented cereal weaning foods. Chalmers Biblioteks ReproService, Goteborg

Lupiem, J.R. 1996. Implementing the ICN World Plan of Action for Nutrition: Focus on Food and Agriculture-Based Approaches to improve Nutrition and Preventing micronutrient deficiencies: In: Food-Based Approaches to preventing micronutrient malnutrition, Combs, G.F., Welch, R.M., Duxbury, J.M., Uphoff, N.T. and Nesheim, M.C. (eds) Ithaca, New York

MacPhail, A. P. and Bothwell, T. H., 1992. The prevalence and causes of nutritional iron deficiency anaemia. In: Nutritional anaemias Eds. Fomon, S. J. and Zlotkin, S. Nestle Nutrition Workshop Series 30: 1-12, Raven Press, New York.

Maxwell, P.E. 1990. Cost - control implications of nutrient fortification. *Pre. Foods* 11 (2):: 87-93

Meija, L. A. (1994). Fortification of foods: Historical development and current practices. *Food and Nutr.* 15: 278-281.

Mensah, E. O. and Sefa-Dedeh, S. 1991. Traditional food processing technology and high protein food production. *Food and Nutrition Bulletin*, 1(1): 43-49

Mensah, P. A., Tomkims, A. M., Drasar, B. S. and Harrison, T. J. 1988. Effect of fermentation of Ghanaian maize dough on the survival and proliferation of 4 strains of *Shigella flexneri*. *Trans. Roy. Soc. Trop. Med. Hyg.* 82: 635-656

Mensah, P. A., Tomkims, A. M., Drasar, B. S. and Harrison, T. J. 1991. Antimicrobial effect of fermented Ghanaian maize dough. *J. Appl. Bact.* 7: 203-210

Mensah, P. P. A. 1990. Microbiological studies of fermented Ghanaian maize dough.

PhD. Thesis. London School of Hygiene and Tropical Medicine

Ministry of Health (Nutrition Unit). 1998. Dissemination of Results National Anaemia and Vitamin A Surveys. In press.

Motarjemi, Y. and Nout, M. J. R. 1996. Food fermentation: a safety and nutritional assessment. *Bulletin of the World Health Organisation.* 74(6): 553-559

Motarjemi, Y., Kaferstein, F., Moy, G. and Quevedo, F. 1993. Contaminated weaning food: a major risk factor for diarrhoea and associated malnutrition. *Bull. of the WHO,* 71(1): 79-92

Muller, H. G. 1970. Traditional cereal processing in Nigeria and Ghana. *Ghana J.Agric.Sci.* 3:187-195

Nawar, W.W. 1985. Lipids. In: *Food chemistry*, 2nd ed., Fennema. O.R. ed. Marcel Dekker, New York

Nche, P. F., Nout, M. J. R. and Rombouts, F. M. 1994. The effect of cowpea supplementation on the quality of kenkey, a traditional Ghanaian fermented maize food. *J. of Cereal Sci.* 19: 191-197

Nielsen, S.S. 1994. Fat characterisation. In: Introduction to the chemical analysis of food. Publ. Jones and Bartlets pp 195-205

Nogueira, N.N., Colli, C. and Cozzolino, S.M.F. 1992. Control of iron deficiency anaemia in preschool children by fortification of food with bovine haemoglobin concentrate (preliminary study). *Cadernos de Saude Publica* 8: 459-465

Nout, M. J. R, Rombouts, F.M. and Havelaar, A. 1989: Effect of accelerated natural lactic fermentation of infant food ingredients on some pathogenic microorganisms. *Int. J. Food Microbiol.* 8; 351-361

Nout, M. J. R. (1993). Processed weaning foods for Tropical climates. *Intern. J. of Food Sciences and Nutr.* 43: 213-221.

Obiri-Danso, K. 1994. Microbiological studies of corn dough fermentation. *Cereal Chem.* 71(2): 186-188

Okoree, J. 1993. Cowpea in solid state fermentation in maize dough. BSc. Project. Dept. of Nutrition and Food Science, Univ. of Ghana.

Orraca-Tetteh, R. 1972. The place of legumes in nutrition, 9th International Congress of Nutrition, Mexico, *Summaria.* 152

Osa-Mensah, A. 1991. The effect of process treatment and functional properties and chemical characteristics of sorghum and millet. BSc. Project. Dept. of Nutrition and Food Science, Univ. of Ghana.

Osei, A. K. and Sefa-Dedeh, S. 1993. Performance of steam treated cowpea in solid state fermentation of cereal dough "Aboloo" system. Paper presented at the 13th National Maize and Legume workshop. Ghana grains development project.

Paredes-Lopez, O. and Mora-Escobedo, R. 1983. Influence of storage on the quality of maize meal for tortilla making. *J. Food Technol.* 18:53-60

Patrick, J., Jr. 1985. Elemental sources. In: Iron fortification of foods. Clydesdale, F. M. and Wiemer, K. L. (eds). Academic Press, New York.

Phillips, R. D. 1991. Weaning foods. In: Sustain Notes. Sustain National Cooperative Business Assoc., NY, 44: 195-211

Phillips, R. D., Chinnam, M. S., Branch, A. L., Miller, J. and McWatters, K. H. 1988. Effects of pretreatment on functional and nutritional properties of cowpea malt. *J. Food Sci.* 53(3): 805-809

Pizarro, F., Yip, R., Dallman, P. R., Olivares, M., Hertrampf, E. and Walter, T. 1991. Iron status with different infant feeding regimens: relevance to screening and prevention of iron deficiency. *J. Pediatr.* 118: 687-692

Plahar, W. A. and Leung, H. K. 1982. Effect of moisture content on the development of carboxylic acids in traditional maize dough fermentation. *J. Sci. Food Agric.* 33: 555

Pour-EL, A. 1981. Protein functionality, classification, definition and methodology. In: *Protein functionality in foods*. Cheny, J. P. (ed). ACS Symposium Series No. 147, American Chemical Society. Washington. D. C.

Protein-Calorie Advisory Group of the United Nations Systems: Marketing protein-rich foods in developing countries. PAG Guideline No. In: *Developing countries in: Compendium 14/10*: G. New York: John Wiley and Sons, 1975: 405-500

Purvis, G.A. 1985. Supplementation of infant products. in: *Iron fortification of foods*. eds. Clydesdale, F. and Wiemer, K.L. Academic Press, New York

Quarshie, N. B. 1988. Nutrient contribution of weanimix to the food intake of Ghanaian infants. BSc. Project. Dept. of Nutrition and Food Science, Univ. of Ghana.

Rasper, 1982. Theoretical aspects of Amylographology. In: *The Amylograph Handbook*. Shuey, W. C. (ed), Tipples, K. H. American Association of Cereal Chemists

Renner, E. (ed) 1989. *Micronutrients in milk and milk-based food products*. London: Elsevier Applied Sci. Publishers, London.

Roman, A. V. B., Bender, A. E. and Norton, I. D. 1987. Formulation and processing of a weaning food based on rice, cowpea and skim milk powder. *Hum. Nutr. Food Sci. & Nutr.* 41F: 15-22

Saalia, F.K. 1995. Physical modification of maize and cowpea and their application in traditional foods. M. Phil. Thesis. Dept. of Nutrition and Food Science, University of Ghana.

Sandberg, A-S, Anderson, H., Carlson, N-G. and Sandstrom, B. 1987. Degradation products of bran phytate formed during digestion in the human small intestine. effect of Extrusion cooking on digestibility. *American Institute of Nutrition*

Scheon, H. N. 1977. Functional properties of proteins and their measurement In: *Food proteins*. Whitaker, J. R. and Tannenbaum, S. R. AVI Publishing Company Inc.

Sefa-Dedeh, S. 1988. Interim report research on maize varieties grown in Ghana, University of Ghana, Legon. (Unpublished)

Sefa-Dedeh, S. 1989. Effects of particle size on some physicochemical characteristics of "agbelima" (Cassava dough) and corn dough. *Tropical Science* 29(1) 21-32

Sefa-Dedeh, S. 1991. (ed.) Proceedings of the AAU seminar on development of high protein-energy foods from grain legumes. 5-7

Sefa-Dedeh, S. 1993. Traditional Food Technology. In: Encyclopaedia of Food Science, Food Technology and Nutrition. R. Macrae, R. Robinson and R. Sadler (eds.). Academic press pp 4600-4606

Sefa-Dedeh, S. and Ampadu, E. W. 1991. Chemical and functional properties of cereal /legume blends. In: Development of high protein-energy foods from grain legumes. Proc. of Int'l workshop, AAU Food and Nutrition Project. Sefa-Dedeh, S. (ed)

Sefa-Dedeh, S., Donkor, M., Lartey, A. and Asiedu, M. S. 1996. Infant feeding practices of hospitalised malnourished children and cowpea utilisation. UGL-CRSP Working Paper 96-4

Simango, C. and Rukure 1992. Survival of bacterial enteric pathogens in traditional fermented foods. J. Appl. Bacteriol. 73, 37-40

Simmons, W. K. and Gurney, J. M. 1980. Nutritional Anaemia in English-speaking Caribbean and Surinam, Kingston, Jamaica: Caribbean Food Nutr. Inst. J.- 46-80. PAHO

Sloan, A. E. and Stiedeman, M. K. (1996). Food Fortification: From Public-Health Solution to Contemporary Demand. Food Technol. 69 (6): 100-108.

Smith, K.T. 1983. Effects of chemical environment on iron bioavailability measurements. Food Technol. 37(10): 115

Svanberg, U., Lorri, W. and Sandberg, A.S. 1993. Lactic fermentation of non-tannin and high-tannin cereals: Effects on in vitro estimation of iron availability and phytate hydrolysis. *J. Food Sci.* 58: 408-412

Tien, M., Svingen, B.A. and Aust, S.D. 1981. Initiation of lipid peroxidation by preferred complex. In: *Oxygen and Oxy-Radicals in chemistry and biology*. eds: Rodgers, M.A.J. and Powers, E.I. Academic Press, New York

Tien, M., Svingen, B.A. and Aust, S.D. 1981. Superoxide dependent lipid peroxidation. *Fed. Proc. Fed. Am. Soc. Exp. Biol.* 40 (20), 179-182

Tien, M., Svingen, B.A. and Aust, S.D. 1982. An investigation into the role of hydroxyl radical in xanthine oxidase-dependent lipid peroxidation. *Arch. Biochem. Biophys.* 216, 142-151

Trowbridge, F. L., Harris, S. S., Cook, J., Dunn, J.T., Florention., R.F., Kodyat, B. .A., Mannar, M.G., Reddy, V., Tontisirin, K., Underwood, B. A. and Yip, R. 1993. Coordinated strategies for controlling micronutrient malnutrition: A technical Workshop. *J. Nutr.* 123: 775-787

UNICEF. 1998. *The State of the World's Children*. New York: Oxford Univ. Press.

Valysevi, A. and Dhanamitta, S. 1987. Development of supplementary foods at home and village level. *Asean Food J.* 3: 33-36

Van der Poel, A. F. B. 1990.: Effects of processing on bean (*Phaseolus vulgaris* L.) protein quality. PhD Thesis, Wageningen Agricultural University.

Waddel, J. 1973. The Bioavailability of iron sources and their utilisation in food enrichment. FASEB report for FDA. Bethesda, Maryland

Walter, T., Dallman, P. R., Pizarro, F., Velozo, L., Pena, G., Barhomey, S. J., Hertrampf, E., Olivares, M., Letelier, A. and Arredondo, M. 1993. Effectiveness of iron-fortified infant cereal in prevention of iron deficiency anaemia. *Paediatric*, 91: 876-982.

Wanink, J. F., Van Vliet, T. and Nout, M. J. R. 1994. Effect of roasting and fermentation on viscosity of cereal-legume based food formulas. *Plant Foods for Hum. Nutr.* 46: 117-126

Wien, E.M. and Schwartz, R. 1985. Dietary calcium exchangeability and bioavailability. In: nutritional Bioavailability of calcium. Kies, C. (ed) ACS Symposium series 275. American Chemical Society, Washington, D.C.

Yip, R. 1989. Improving iron nutrition status of U.S. children. In: Dietary iron: Birth to two years. Filer, J. (ed) pp 37-61, Raven Press, New York, NY.

APPENDICES

APPENDIX 1

SAMPLE CALCULATION FOR IRON ADDITION

- I. Energy for weanimix consumed in a day for 0-6 months
= 270.2 kcal (Quashie, 1988)
100g of weanimix = 410.7 kcal

Xg / day = 65.79g

5% iron bioavailability of diet containing cereal/legume for 0.25-1 year from the RDI (Table 2) is 21mg of iron

If 65.79g = 21mg

100g = 32mg

- II. For 7-12 months energy consumed = 289.5 kcal

31mg iron/day

Maximum iron content = 32mg/100g

CONSENT FORM/DECLARATION

UNIVERSITY OF GHANA/UNICEF/CRSP PROJECT
SENSORY EVALUATION OF IRON-FORTIFIED WEANING FOODS

Date..... Community..... District.....

Background Data

1. Age
2. Highest Educational level a)None b)Elementary c)JSS d)SSS
e)VOC/TECH f)Others.....
3. Occupation.....
4. Number of children in household.....
5. Age of last child.....

PRODUCT EVALUATION

A COOKED DRIED IRON-FORTIFIED FERMENTED MAIZE DOUGH

6. Please taste the cooked product and rank the coded samples in order of preference with respect to the following indices.

	1	2	3	4	5	6
COLOUR						
FLAVOUR						
TASTE						
GENERAL ACCEPTABILITY						

7. How much would you be willing to pay for a margarine tin full (450g) of the dried product?.....

8. Any comments?.....

B COOKED DRIED IRON-FORTIFIED FERMENTED MAIZE/COWPEA DOUGH

9. Please taste the cooked product and rank the coded samples in order of preference with respect to the following indices.

	1	2	3	4	5	6
COLOUR						
FLAVOUR						
TASTE						
GENERAL ACCEPTABILITY						

10. How much would you be willing to pay for a margarine tin full(450g) of the dried product?

11. Any comments?.....

C COOKED ROASTED IRON-FORTIFIED MAIZE/COWPEA/GROUNDNUT BLEND

12. Please taste the cooked product and rank the coded samples in order of preference with respect to the following indices.

	1	2	3	4	5	6
COLOUR						
FLAVOUR						
TASTE						
GENERAL ACCEPTABILITY						

13. How much would you be willing to pay for a margarine tin full(450g) of the dried product?

14. Any comments?.....

Appendix 3 Effect of fermentation, storage and type, concentration and mode of addition iron on pH* of dehydrated maize dough

Method of fortification	Iron level mg/100g	Storage time in months	Ferrous Sulphate Fermentation time/h				Ferrous fumarate Fermentation time /h			
			0	36	48	72	0	36	48	72
	0	0	5.26	3.45	3.34	3.58	5.26	3.45	3.34	3.58
		2	5.57	4.62	4.53	4.48	5.57	4.62	4.53	4.48
		4	5.40	3.58	3.50	3.52	5.40	3.58	3.50	3.52
Before	16	0	5.71	4.31	4.03	4.42	5.57	4.13	3.81	5.12
		2	6.27	5.11	4.48	4.92	6.28	5.22	4.93	5.74
		4	5.47	4.18	4.02	4.07	4.06	4.09	3.77	4.57
	32	0	5.06	4.23	4.17	4.22	5.09	4.24	4.25	4.39
		2	5.00	4.09	4.09	4.13	5.05	4.17	4.17	4.29
		4	5.06	4.18	4.18	4.23	5.09	4.23	4.25	4.39
After	16	0	5.71	3.72	3.62	3.84	5.27	3.61	3.51	3.78
		2	6.27	4.45	4.41	4.36	6.28	4.32	4.27	4.24
		4	5.47	3.64	3.59	3.55	4.06	3.60	3.54	3.56
	32	0	5.06	4.13	4.11	4.69	5.09	4.16	4.17	4.24
		2	5.00	4.13	4.10	4.14	5.05	4.12	4.16	4.17
		4	5.06	4.14	4.13	4.15	5.09	4.15	4.17	4.24

*: Average of two determinations

Appendix 4 Effect of fermentation, storage and type, concentration and mode of addition iron on titratable acidity* (in terms of lactic acid) of dehydrated maize dough

Method of fortification	Iron level mg/100g	Storage time in months	Ferrous Sulphate Fermentation time/h				Ferrous fumarate Fermentation time /h			
			0	36	48	72	0	36	48	72
	0	0	0.03	0.14	0.16	0.18	0.03	0.14	0.16	0.18
		2	0.035	0.143	0.163	0.18	0.035	0.143	0.163	0.18
		4	0.032	0.142	0.162	0.18	0.032	0.142	0.162	0.18
Before	16	0	0.035	0.04	0.06	0.05	0.03	0.065	0.085	0.04
		2	0.045	0.048	0.065	0.053	0.035	0.068	0.09	0.045
		4	0.045	0.058	0.065	0.053	0.035	0.068	0.087	0.048
	32	0	0.035	0.065	0.06	0.048	0.038	0.065	0.063	0.08
		2	0.03	0.055	0.06	0.04	0.03	0.06	0.058	0.075
		4	0.035	0.075	0.065	0.048	0.04	0.075	0.065	0.078
After	16	0	0.035	0.14	0.15	0.155	0.03	0.16	0.16	0.18
		2	0.045	0.15	0.15	0.16	0.038	0.17	0.165	0.18
		4	0.045	0.14	0.15	0.155	0.07	0.15	0.16	0.165
	32	0	0.035	0.065	0.065	0.05	0.04	0.065	0.065	0.049
		2	0.03	0.055	0.06	0.05	0.038	0.06	0.06	0.048
		4	0.035	0.075	0.065	0.05	0.048	0.075	0.075	0.05

*: 1. Average of two determinations

2. g/100g sample on dry matter basis

Appendix 5 Effect of fermentation, storage and type, concentration and mode of addition iron on fat acidity* of dehydrated maize dough

Method of fortification	Iron level mg/100g	Storage time in months	Ferrous Sulphate Fermentation time/h				Ferrous fumarate Fermentation time /h			
			0	36	48	72	0	36	48	72
	0	0	33.96	92.80	143.68	127.27	33.96	92.80	143.68	127.27
		2	54.60	136.23	197.62	200.22	54.60	136.23	197.62	200.22
		4	81.40	214.95	264.03	280.98	81.40	214.95	264.03	280.98
Before	16	0	14.64	101.58	182.69	135.38	13.96	124.19	133.40	130.81
		2	60.57	173.46	223.44	225.51	39.52	178.39	194.59	201.75
		4	60.44	134.08	112.17	141.55	97.89	173.09	101.00	105.33
	32	0	57.94	137.11	153.36	175.07	76.59	134.72	150.65	146.91
		2	52.12	116.72	136.97	153.06	64.71	124.61	144.03	148.13
		4	96.85	155.47	195.18	205.12	113.94	157.08	199.35	208.80
After	16	0	14.64	108.54	156.01	172.21	13.96	130.39	163.41	181.23
		2	60.57	178.03	246.89	201.27	39.52	170.21	217.54	226.46
		4	60.44	250.69	217.85	285.79	97.89	234.72	199.01	258.00
	32	0	57.94	123.89	140.89	171.87	76.59	108.67	181.18	150.81
		2	52.12	122.94	144.93	159.07	64.71	11.50	176.87	175.16
		4	96.85	162.73	203.43	212.05	113.94	156.44	221.43	238.05

*: Average of two determinations

Appendix 6 Effect of fermentation, storage and type, concentration and mode of addition iron on colour*(L*) of dehydrated maize dough

Method of fortification	Iron level mg/100g	Storage time in months	Ferrous Sulphate Fermentation time/h				Ferrous fumarate Fermentation time /h			
			0	36	48	72	0	36	48	72
	0	0	91.99	95.58	94.48	92.54	91.99	95.58	94.48	92.54
		2	90.05	93.17	93.10	92.56	90.05	93.17	93.10	92.56
		4	84.17	87.56	86.97	86.62	84.17	87.56	86.97	86.62
Before	16	0	91.32	95.06	93.89	92.44	90.60	94.69	93.68	92.10
		2	87.70	92.97	92.15	92.49	88.67	92.66	91.99	92.59
		4	84.02	86.83	86.09	86.62	82.98	86.66	86.09	86.66
	32	0	84.03	86.28	86.05	85.80	84.56	86.24	86.19	86.16
		2	84.81	86.46	86.21	86.18	84.94	86.37	86.48	85.66
		4	84.91	86.08	85.92	86.18	85.13	85.97	86.02	85.81
After	16	0	91.32	94.90	94.08	92.88	90.60	95.13	93.90	92.24
		2	87.70	93.36	93.29	92.46	88.67	93.20	93.04	92.74
		4	84.02	87.12	86.94	86.79	82.98	86.98	86.98	86.82
	32	0	84.03	86.21	86.06	85.82	84.56	85.99	85.61	86.09
		2	84.81	86.48	86.07	86.08	84.94	86.33	85.93	86.42
		4	84.91	85.72	85.78	85.89	85.13	86.23	85.92	86.19

*: Average of three determinations

Appendix 7 Effect of fermentation, storage and type, concentration and mode of addition iron on water absorption* at 30°C of dehydrated maize dough

Method of fortification	Iron level mg/100g	Storage time in months	Ferrous Sulphate Fermentation time/h				Ferrous fumarate Fermentation time /h			
			0	36	48	72	0	36	48	72
	0	0	148.54	128.76	118.98	135.33	148.54	128.76	118.98	135.33
		2	144.69	127.47	122.03	136.63	144.69	127.47	122.03	136.63
		4	150.15	124.18	119.07	138.09	150.15	124.18	119.07	138.09
Before	16	0	145.56	119.13	125.84	147.22	163.82	125.90	130.19	144.25
		2	149.04	121.65	126.90	141.19	167.41	128.57	134.00	140.71
		4	144.01	123.70	127.01	143.91	166.94	127.58	138.01	146.98
	32	0	126.13	125.00	119.65	127.87	125.99	126.59	121.33	131.20
		2	133.53	127.12	124.37	140.54	131.21	127.09	123.89	137.13
		4	132.00	122.17	118.68	128.62	125.40	121.72	120.18	128.12
After	16	0	145.56	119.69	125.71	132.88	163.82	129.66	130.42	133.00
		2	149.04	123.81	120.99	137.79	167.41	120.97	127.93	137.16
		4	144.01	126.68	124.43	131.94	166.94	129.75	134.12	137.90
	32	0	126.13	126.04	129.95	128.09	125.99	119.67	129.31	138.98
		2	133.53	123.88	128.76	142.96	131.21	120.60	133.33	140.19
		4	132.00	120.29	128.01	133.42	125.40	122.34	138.02	142.17

*: 1. Average of two determinations

2. g/100g sample on dry matter basis

Appendix 8 Effect of fermentation, storage and type, concentration and mode of addition iron on water absorption* at 70°C of dehydrated maize dough

Method of fortification	Iron level mg/100g	Storage time in months	Ferrous Sulphate Fermentation time/h				Ferrous fumarate Fermentation time /h			
			0	36	48	72	0	36	48	72
	0	0	198.74	180.49	160.58	168.11	198.74	180.49	160.58	168.11
		2	219.47	180.24	162.79	170.00	219.47	180.24	162.79	170.00
		4	225.24	182.24	169.68	171.69	225.24	182.24	169.68	171.69
Before	16	0	229.19	179.73	168.26	187.01	254.91	163.01	163.36	184.67
		2	242.30	173.91	169.32	180.59	250.78	165.09	166.63	181.06
		4	243.48	179.16	167.04	181.99	252.20	164.00	170.64	185.12
	32	0	165.96	170.01	179.97	177.12	160.94	166.42	159.12	170.92
		2	173.36	173.23	185.44	179.02	161.59	166.91	161.05	176.84
		4	177.27	165.34	164.20	180.68	169.34	164.80	161.19	183.99
After	16	0	229.19	157.19	175.83	171.14	254.91	174.12	162.14	173.14
		2	242.30	166.51	173.19	170.97	250.78	175.22	161.88	169.61
		4	243.48	165.91	172.19	171.03	252.20	180.01	164.13	167.99
	32	0	165.96	157.53	170.12	174.39	160.94	167.12	159.99	167.76
		2	173.36	164.47	170.19	178.28	161.59	168.69	160.18	170.21
		4	177.27	161.10	178.43	171.94	169.34	163.87	167.81	173.00

*: 1. Average of two determinations

2. g/100g sample on dry matter basis

Appendix 9 Effect of fermentation, drying and type, concentration and mode of addition iron on pH* of dehydrated maize dough

Method of fortification	Iron level mg/100g	State of sample	Ferrous Sulphate							Ferrous fumarate						
			Fermentation time/h							Fermentation time /h						
			0	12	24	36	48	60	72	0	12	24	36	48	60	72
	0	fresh	5.23	3.93	3.69	3.39	3.41	3.37	3.41	5.23	3.93	3.69	3.39	3.41	3.37	3.41
		dried	5.26	3.95	3.69	3.45	3.34	3.58	3.58	5.26	3.95	3.69	3.45	3.34	3.58	3.58
Before	16	fresh	5.35	4.02	4.00	3.72	4.26	3.81	3.95	5.50	4.06	4.13	4.00	4.35	5.00	4.78
		dried	5.71	4.27	4.10	4.31	4.03	4.23	4.42	5.57	4.36	4.11	4.13	3.81	4.98	5.12
	32	fresh	5.13	4.25	4.16	4.10	4.98	4.95	5.01	5.17	4.23	4.18	4.03	4.99	5.00	5.10
		dried	5.06	4.33	4.18	4.23	4.17	4.11	4.22	5.09	4.39	4.24	4.24	4.25	4.21	4.39
After	16	fresh	5.35	3.90	3.90	3.60	3.61	3.56	3.54	5.50	3.93	3.84	3.60	3.57	3.59	3.53
		dried	5.71	3.95	3.90	3.72	3.62	3.65	3.84	5.27	3.99	3.79	3.61	3.51	3.56	3.78
	32	fresh	5.13	4.24	4.10	4.03	4.97	4.90	5.00	5.17	4.25	4.17	4.10	4.99	5.00	5.09
		dried	5.06	4.23	4.15	4.13	4.11	4.16	4.69	5.09	4.24	4.17	4.16	4.17	4.14	4.24

*: 1. Average of two determinations

Appendix 10 Effect of fermentation, drying and type, concentration and mode of addition iron on titratable acidity* (in terms of lactic acid) of dehydrated maize dough

Method of fortification	Iron level mg/100g	Storage time in months	Ferrous Sulphate Fermentation time/h							Ferrous fumarate Fermentation time /h						
			0	12	24	36	48	60	72	0	12	24	36	48	60	72
	0	fresh	0.025	0.115	0.125	0.16	0.17	0.16	0.175	0.025	0.115	0.125	0.16	0.17	0.16	0.175
		dried	0.03	0.105	0.125	0.14	0.16	0.185	0.18	0.03	0.105	0.125	0.14	0.16	0.185	0.18
Before	16	fresh	0.025	0.09	0.085	0.105	0.055	0.085	0.07	0.025	0.085	0.075	0.085	0.045	0.025	0.025
		dried	0.035	0.06	0.075	0.04	0.06	0.05	0.05	0.03	0.085	0.09	0.065	0.085	0.04	0.04
	32	fresh	0.045	0.07	0.08	0.09	0.075	0.18	0.175	0.05	0.08	0.095	0.085	0.105	0.07	0.055
		dried	0.035	0.05	0.057	0.065	0.06	0.065	0.048	0.038	0.055	0.07	0.065	0.063	0.065	0.08
After	16	fresh	0.025	0.115	0.105	0.15	0.145	0.16	0.17	0.025	0.11	0.125	0.155	0.135	0.175	0.18
		dried	0.035	0.115	0.145	0.14	0.15	0.175	0.155	0.03	0.11	0.125	0.155	0.16	0.185	0.18
	32	fresh	0.045	0.08	0.085	0.09	0.075	0.18	0.175	0.05	0.08	0.08	0.075	0.085	0.065	0.05
		dried	0.035	0.055	0.07	0.065	0.065	0.065	0.05	0.038	0.08	0.06	0.065	0.065	0.065	0.05

*: 1. Average of two determinations

2. g/100g sample on dry matter basis