EFFICACY OF FERRIC SODIUM ETHYLENE DIAMINE TETRAACETIC ACID SUPPLEMENT IN REDUCING IRON DEFICIENCY ANAEMIA AMONG UNDER FIVE CHILDREN IN MOROGORO

SARAH JOHNSON SHALUWA

A DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR MASTER DEGREE IN HUMAN NUTRITION OF SOKOINE UNIVERSITY OF AGRICULTURE. MOROGORO, TANZANIA.

2013

ABSTRACT

Use of high dose of iron (10 mg) supplement to control Iron Deficiency Anaemia (IDA) as recommended by WHO/FAO has been reported to increase severity of malaria in malaria endemic areas. This study was conducted in Mvomero district, one of malaria endemic areas in Tanzania to determine the efficacy of a low dose ferric sodium ethylene diamine tetraacetic acid (Fe Na-EDTA) supplement to reduce anaemia among children aged 6-36 months. A total of 1021 children were screened for their weights, heights and blood haemoglobin concentrations. Out of 1021 children, 217 met the eligibility criteria of being mildly anaemic with blood hemoglobin concentration ranging from ≥7 to ≤ 9g/dl. At baseline, children in the study group were screened for Hb and malaria parasites and all children received anti-helminthes drug- 600 mg of melbendazol (except children under the age of one year). Study children were divided into two groups of 108 each. Group one received the low-dose iron (5 mg of 2.5 mg Fe-lactate and 2.5 mg Fe-Na-EDTA) while the other group received the high dose of iron (10 mg Iron lactate). The children received the supplement for a period of 8 weeks. Results showed that, there was a significant ($p<0.05$) improvement in the Hb concentration for both groups receiving low and high doses of iron. Likewise, there was no significant difference ($p > 0.05$) in the Hb concetrations between the group receiving 10 mg iron lactate and the one receiving 5 mg Na-EDTA (2.5 mg ferrous lactate and 2.5 mg EDTA). The average Hb concentration for the group receiving high dose iron was 11.1 ± 2.0 g/dl while for the group receiving low dose iron the mean Hb concentration was 11.0 ± 2.9 g/dl. It was concluded from this study that, consumption of low dose iron (combination of 2.5 mg sodium-ferric EDTA and 2.5 mg of iron lactate) was just as effective in controlling IDA as consuming high dose iron (10 mg of iron lactate). Low dose of Fe-containing EDTA may therefore be adopted in anaemia control programs especially in malaria endemic areas. It is

recommended that, in order for the iron deficiency control programs to be effective, there must be a concurrent control of malaria and helminthes.

DECLARATION

I, SARAH JOHNSON SHALUWA, do hereby declare to the Senate of Sokoine University of Agriculture that this dissertation is my own original work done within the period of registration and that it has neither been submitted nor being concurrently submitted in any other institution.

03-06-2013

Sarah Johnson Shaluwa
(MSc. Candidate)

Date

The above declaration is confirmed by:

03-06-2013

Prof. T.C.E. Mosha
(Supervisor)

Date

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AKNOWLEDGEMENTS

My profound gratitude is due to my supervisor Prof. T.C.E. Mosha for his guidance, suggestions, constructive criticisms and devotion to the work throughout the study. His readiness to assist has been a key for success of this work. Appreciation should also go to Prof. H. Laswai and Ms. Zahara S. Majili of the Department of Food Science and Technology for their various contributions to this study.

Grateful acknowledgements are also due to Dr. John Assey, of the Turiani District Hospital and all the village health attendants of Lusanga, Kichangani, Kilimanjaro and Manyinga villages for their valuable assistance during data collection. I am greatly indebted to all the mothers who generously accepted to participate with their children in this study, without them the study would not have been possible.

Special thanks are also extended to my daughter Theresia and my husband J. Kapwani for their moral and material support. Many thanks also to my young sister Hellen Shaluwa for taking care of my family during the whole period of the study.

Lastly, I would like to express my sincere thanks to my employer – the President's Office for providing financial support for this study. I wish also to acknowledge with thanks the Heinz Company Foundation for supporting me during data collection.

DEDICATION

This work is dedicated to my beloved parents, Mr. and Mrs Johnson Shaluwa Mange who laid the foundation of my education.

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LIST OF ABBREVIATIONS

ANOVA	Analysis of Variance
CI	Confidence Interval
DNA	Deoxyribonucleic acid
HACCP	Hazard Analysis Critical Control Point
HAZ	Height-for-Age z-scores
Hb	Haemoglobin
IDA	Iron Deficiency Anaemia
GIT	Gastrointestinal Tract
Mo	Months of the child
Na-EDTA	Sodium Ethylene Diamine Tetraacetic Acid
NAS	National Academy Sciences
NBS	National Bureau of Statistics
RCH	Reproductive and child health clinic
TDHS	Tanzania Demographic and Health Survey
TNP	Tanzania Nutritional Profile
UKUMTA	'Kumwendeleza Mtoto Tanzania'
URT	United Republic of Tanzania
WAZ	Weight-for-Age z-scores
WHO	World Health Organization
WHZ	Weight-for- height z-scores

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background Information

Iron deficiency and iron deficiency anaemia (IDA) are widespread in most developing countries and IDA is considered the most significant nutrition problem worldwide (Chaparro, 2008 and Borwankar *et al.*, 2007). IDA during the critical development period of 6-24 months of age can result in the failure of infants to achieve full genetic potential for physical and mental development (Pollit, 1993 and Beard, 2008). Although the biological consequences of IDA have been known for decades, relatively little progress has been made in eradicating or even reducing this nutritional problem in resource poor countries. Tanzania, like most sub–Saharan countries in Africa, suffers from widespread IDA. In Tanzania, prevalence of IDA among children under the age of five years in some parts of the country is as high as 100% (Kavishe, 1991, UKUMTA, 1996).

Data from the Tanzania Demographic and Health Survey (NBS, 2011) indicated that, prevalence of IDA among children under the age of five years ranged between 42 % - 72 % with the most severe cases observed among children aged 6 – 24 months. Lindi, Mwanza, Shinyanga, Mtwara, Tabora, Morogoro and Coast are among the regions of Tanzania with the highest prevalence of iron deficiency anaemia (NBS, 2011). A study on community prevalence of anaemia underfive years of age children in Southern region of Ruvuma revealed that, the prevalence of anaemia among children under the age of five years was 87% with annual incidence of severe anaemia of 0.6 episodes per infant (Mennendez *et al.*, 1997, Schelleberg, 2003). In light of the foregoing, IDA is a serious public health problem in Tanzania, especially among children under the age of five years (Mwanri *et al.*, 2001).

Preventing/correcting IDA is in theory very simple; provide a source of biologically available iron. However, in countries where the diet is primarily plant based and there is no widespread iron fortification program, delivery of adequate amounts of absorbable iron via the normal diet is particularly difficult during infancy (6 – 24 months of age), onset of menstruation in adolescent females and pregnancy. Infants born in countries subsisting on plant foods with no national fortification program are generally born with low stores of iron in their livers because of inadequate maternal supplies of iron during pregnancy. Since breast milk is a poor source of iron, an infant's liver stores of iron are often depleted by six months of age. If the infant is not provided with an adequate source of absorbable iron in solid food or a supplement, the infant is likely to become anaemic and unable to achieve full cognitive and physical development (Christofides *et al.*, 2006, Giovannin *et al.*, 2006).

Numerous approaches to provide sufficient amounts of absorbable iron to infants in remote, rural and, resource – poor communities have been devised. A proven effective approach in these settings has been home-fortification of supplementary foods (Christofides *et al.*, 2006, Giovannin *et al.*, 2006). A daily dose of iron and other micronutrients is provided in easy to use sachets. The content of a sachet is sprinkled onto a prepared supplementary food just prior to consumption to provide micronutrients not available in the food. Heinz Nuture-Mate™ is one such home – fortification product suitable for infants in developing countries.

Malaria and hookworms increase the needs for iron. However, there is currently a controversy regarding the use of iron supplements in malaria endemic areas because a study conducted in Zanzibar reported an increase in severity of malaria caused morbidity with the use of iron supplements (Sazawal *et al.*, 2006). The study design, data interpretation, and recommendations of the study are often criticized and have led to considerable scientific debate (Prentice, 2008). Experts in this area of public health suggest that, iron supplementation is a potential problem in malaria endemic regions only in iron replete (non anaemic) individuals (Prentice, 2008). For individuals suffering from IDA, iron supplementation has been effective in correcting the anaemia with no adverse consequences. The current recommended approach for treating IDA is to prevent/treat malaria and parasites in addition to providing iron supplements (Prentice, 2008 and Ouedraogo *et al.*, 2008).

1.2 Problem Statement and Justification

Anaemia is among the serious nutritional problems in Tanzania with highest prevalence among children aged 6-59 months. According to NBS (2011), prevalence of iron deficiency anaemia among males is 60.9% and 56.4 % among female children of the same age. A number of interventions have been put in place to address anaemia in children. These include home-fortification of supplementary foods (Christofides *et al.*, 2006, Giovannin *et al.*, 2006). Promotion of use of insecticide-treated mosquito nets by children under age 5 years (to prevent malaria parasites) and deworming (to prevent worms infestation) these parasites feed on red blood cells and can lead to IDA. But, the prevalence of IDA especially in malaria endermic areas is still high at 59.2% in Morogoro (NBS, 2011). Study conducted in Zanzibar, Tanzania found that, the supplementation of high dose (10 mg iron lactate) in malaria endermic areas could help promote the growth of malarial parasites circulating in the blood (Ojukwu *et al.*, 2009). Significant increase of malaria severity prompted World Health Organization to issue to, institution and other stakeholders addressing IDA in children underfive necessary to device low dose iron. The low dose iron bound to EDTA (5mg Na-EDTA) increases iron absorption and does not increase malaria severity to children in malaria endermic areas. Interventions that aim at reducing the prevalence of IDA are thus essential. This study was conducted to i) verify the previous report that an iron supplement can effectively reduce IDA without increasing the incidence of malaria infection; and ii) determine if a supplement providing low dose (2.5 mg) of iron in the form of ferric Ethylene Diamine Tetraacetic Acid and (2.5 mg) of iron as ferrous lactate is as effective in correcting IDA as a supplement providing high dose 10 mg of iron as ferrous lactate. Then, the low dose of iron supplement (5mg Na-EDTA) would be recommended to provide an extra margin of safety in correcting IDA, especially in malaria endermic areas.

1.3 Objectives

1.3.1 General objective

To determine the efficacy of a low dose ferric sodium ethylene diamine tetraacetic acid (Fe Na-EDTA) supplement to reduce anaemia in malaria- endemic area of Turiani, Morogoro.

1.3.2 Specific objectives

i. To assess the nutrition status of children aged 6-59 months in the study area.
ii. To determine the effect of iron supplementation on the haemoglobin concentration.
iii. To compare the efficacy of low dose of ferric Na-EDTA with the normal dose
iv. To compare the anthropometric characteristics (WAZ, WHZ and HAZ) of the studied children before and after iron supplementation.
v. To evaluate dietary pattern of children in the study area.

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CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Meaning of Iron

Iron is the most abundant metal in the universe and in the earth crust. It is also one of the most useful both in technology and in biology. Iron compounds are involved in numerous oxidation–reduction reactions. Aerobic metabolism depends on iron because of its role in the functional groups of most of the Krebs cycle, as an electron carrier in cytochromes and as a means of oxygen and carbon-dioxide gases transport in hemoglobin (Shils *et al.*, 1994).

2.2 Forms of Iron

There are two (2) forms of dietary iron: heme and non heme iron. Heme iron is derived from hemoglobin, protein in red blood cells and myoglobin (pigment found in muscles). It is found in animal product and it is the most efficiently absorbed form of iron. Non-heme iron is found in plant foods such as grains and vegetables. Iron absorption depends on its form in the food and other factors such as the presence of vitamin C in the diet. The form of iron in food influences how much is absorbed. About 40% of the total iron in animal flesh is in the form of hemoglobin (the same form as in the red blood cells) and myoglobin. The heme iron is absorbed more efficiently than non-heme iron. Non heme iron is present in eggs, milk, vegetables and grains. It is also found in small amount in animal flesh (Wardlaw, 2003).

Consuming heme and non-heme iron together increases non-heme iron absorption. Eating meat with vegetables and grain products enhances the absorption of all non-heme iron present (Wardlaw, 2003). Vitamin C intake along with food increases non-heme iron absorption. Therefore when iron supplements is to be administered, one should take into account of vitamin C. Consuming more foods rich in vitamin C is particularly desirable if dietary iron is inadequate or blood iron is low. Iron use in the body is also aided by copper (Wardlaw, 2003). Manufactures add iron to certain foods such as wheat flour and bread, infant formula and other processed foods which are absorbed quite well (King and Burgess, 1993).

2.3 Functions and Importance of Iron to Health

Iron is involved in the function and synthesis of hemoglobin in red blood cells and myoglobin in muscle cells. Hemoglobin molecules in red blood cells transport oxygen from the lungs to cells and assist in the return of carbondioxide from cells to the lungs for excretion. Iron is used as part of many enzymes that are used in energy production. Iron is also involved in the synthesis of Deoxyribonucleic acid (DNA) and plays a role in the immune function and contributes to drug detoxification in the liver (Shills *et al.,* 1994). The health and vitality of human beings depend on a diet that includes adequate amounts of certain vitamins and minerals that promote effective functioning of physiologic processes, including reproduction, immune response, brain and other neural functions, and energy metabolism. The body needs relatively trace quantities of these elements measured in micrograms or milligrams (NAS, 1998). These elements are essential; they cannot be manufactured by the human body and must be obtained through diet. Deficiencies of most micronutrients are known to have devastating effects on health. They increase risk of overall morbidity and are associated with a variety of adverse health effects, including poor intellectual development and cognition, decreased immunity, and impaired work capacity. The adverse effects of micronutrient under nutrition are most severe for children, pregnant women, and the fetus. Therefore, the high prevalence of iron deficiency is of concern because of its adverse neurodevelopment outcome in children (Friel *et al.*, 2003).

2.4 Deficiency Signs and Symptoms

Iron deficiency anaemia is a type of anaemia due to lack of adequate iron. It may be caused by one or combination of inadequate dietary intake of iron, impaired absorption, blood loss due to parasites such as malaria parasites and worm infestation such as hookworms. It is most common in children below five years of age. IDA is typically diagnosed by low hemoglobin, accompanied by biochemical evidence of iron deficiency such as low serum ferritin concentration (Tatala and Mselle, 2006). Patients with iron deficiency anaemia may be unaware of being in ill health; however, they commonly experience symptomatic improvement once iron therapy is initiated. In symptomatic patients with moderate to severe degrees of anaemia, the signs and symptoms include weakness, fatigue, pallor, dyspnea on exertion, palpitation and the sense of being overly tired even when minor exercise is carried out. Other symptoms include malabsorption, Protein Energy Malnutrition (PEM), impaired cognition and anaemia (Shills *et al.*, 1994).

2.6 Causes of Iron Deficiency

Iron deficiency anaemia results from a variety of causes, including inadequate intake, high physiological demand in early childhood and pregnancy and iron losses from parasitic infections such as malaria and hookworm infestation which are the major non- nutritional risk factors for anaemia (Stoltzfus *et al.*, 1997). The situation is complicated further because anaemia in childhood can result not only from events in childhood but also from maternal iron deficiency and anaemia, which are associated with impaired fetal development and iron-deficient in anaemic babies. Socioeconomic status may also affect the risk of anaemia by affecting nutritional status, family size and birth interval, as well as intensifying problems of affordability and accessibility of preventive and curative measures (Schellenberg *et al.*, 2003). Additional causes include other nutritional deficiencies such as vitamins B-12, B-6, A, riboflavin and folic acid in the diet (Lutter, 2008). Also iron deficiency and anaemia are due to a negative iron balance in children. Despite the apparent simplicity of iron deficiency anaemia occurrence, few advances have been made to reduce iron deficiency anaemia prevalence. This could have been due to the fact that, the negative iron balance is the final outcome of a series of bio-socioeconomic and cultural factors acting on the child development (Almeida *et al.*, 2004).

2.7 Occurrence of Iron Deficiency Anaemia

The stages in the development of iron deficiency are the depletion of iron stores. Indicated by low plasma ferritin, interference with biochemical processes, indicated by low transferrin saturation and elevated free erythrocyte protoporphyrin and serum transferrin receptors and finally, anaemia, as indicated by low hemoglobin (Tatala and Mselle, 2006). It should be noted that, although transferrin receptors appear promising as an indicator, standard cutoffs and interpretation of values from different commercial assays are yet to be developed. Up to an anaemia prevalence of 50 percent, the proportion of individuals with biochemical iron deficiency is about double those with actual anaemia. The general diagnosis of anaemia should lead to a causal analysis. The necessary interventions and community participation toward the common aim of controlling iron deficiency and anaemia must be undertaken (NAS, 1998).

Children beyond 6 months of age need an additional source of iron beyond that provided by breast milk. A large body of evidence documents that, iron deficiency and anaemia in older infants and young children can be prevented by appropriate complementary feeding. When breast-feeding is not possible, iron-fortified milk preparations are needed (Tatala and Mselle, 2006). Another alternative after about six months of age is preventive iron supplementation. In this age group, once iron deficiency is present, anaemia develops quickly and therapy with oral iron is needed to rapidly improve the infant's hematological status and avoid possible permanent developmental deficits. The strategy of iron supplementation for this age group is often neglected by those who are unaware that the vast majority of infants live in poor households in the developing world, where the resources for preparing highly bioavailable, iron rich foods complementary to breast-feeding or for purchasing iron-fortified foods are nonexistent (NAS, 1998).

2.8 Consequences of Iron Deficiency

Iron deficiency adversely affects the cognitive performance, behavior, and physical growth of infants, preschool and school-aged children; the immune status and morbidity from infections of all age groups; and the use of energy sources by muscles and thus the physical capacity and work performance of adolescents and adults of all age groups is impaired (WHO, 2001). Specifically, iron deficiency anaemia during pregnancy increases prenatal risks for mothers and neonates Furthermore, it increases risk of pregnancy complications, including premature, fetal growth retardation and increases overall infant mortality. Moreover, iron-deficient animals and humans have impaired gastrointestinal functions and altered patterns of hormone production and metabolism. Others include impaired memory, increased lead and cadmium absorption (Stang and Story, 2005).

2.9 Iron Regulation and Metabolism

In considering iron regulation and metabolism, iron interactions with other nutrients is important as it affects the absorption and utilization of iron (Fernando, 1998). Copper is involved in oxidoreduction of iron in the process of absorption, transport, storage, and mobilization; folate and vitamin B-12 are involved in nucleic acid synthesis of all cells and clearly in erythropoiesis, thus modifying iron utilization. Vitamins B-6 and B-2 are specifically required in the process of haem synthesis; and amino acids are required for protein biosynthesis in general and in particular hemoglobin biosynthesis. Vitamin A is involved in mobilization of iron reserves, in hemoglobin biosynthesis, and appears to favor iron absorption in the presence of inhibitors. Zinc enhances iron absorption in the body (Fernando, 1998).

2.10 Importance of Iron Supplementation to Children

Supplementation is the method of choice when therapeutic treatment is necessary, that is, to address micronutrient deficiency. Supplementation is also an appropriate tool for preventive programs as long as the distribution system can be maintained and those receiving the supplements continue to consume (NAS, 1998). Therefore, Iron supplementation is the process of providing iron together with other minerals like zinc and vitamins such as B and C vitamins so as to enhance iron absorption in the body, the aim of supplementation is to correct iron deficiency. Study by Iannott *et al.* (2009) revealed that among iron-deficient or anaemic children, hemoglobin concentrations were improved significantly with iron supplementation. Another study by Ziegler *et al.* (2009) showed iron supplementation to be beneficial among anaemic children by improving their haemoglobin concentration. This suggested that, iron supplementation is important for better growth and development of children below the age of five years.

The global guidelines for iron supplementation in childhood need to be enhanced, with the exception of children of low birth weight, for whom iron supplements are recommended from two months of age. WHO/UNICEF (2006) recommends that, iron supplementation should commence only at 6 months; however, by this age anaemia has already affected a large proportion of children. A study by (Stolzfus, 1997) involving iron supplementation of children aged 2–6 months showed that there was 29% reduction in the incidence of severe anaemia (packed cell volume <25% (Hb <7 g/dl) in the first year of life.

Furthermore, considering the high prevalence of deficiencies of iron in malaria endemic areas and their negative effects, supplementation of young children with iron and other micronutrients of public health significance would be of great benefit because of the practical difficulties in improving the nutritional adequacy of traditional infant feeding pattern (Thu *et al.*, 1999). Children at high risk include those aged 6-24 months. These children can no longer depend on breast milk alone to supply their nutrition requirements. Also their complementary foods usually contains low amount of bioavailable iron and zinc. While containing high amounts of phytate, which inhibits the absorption of iron found in the food (Thu *et al.*, 1999).

2.10.1 Importance of iron-EDTA supplements

Ethylene diamine tetraacetic acid (EDTA) is a hexadentate chelator, which can combine with virtually every metal in the periodic table. Sodium iron ethylenediaminetetraacetic acid (Na-Fe EDTA), known as iron-EDTA, is a potentially valuable fortificant (WHO, 2001). Compared to other fortificants, it is better absorbed and not sensitive to many food iron inhibitors. Therefore, it is particularly important in populations whose staple foods are based on cereals and legumes. Since iron-EDTA is well absorbed and not reactive, it does not cause fat (e.g. in bread) to become rancid. Therefore, it is suitable for use in other (not previously fortified) foods (WHO, 2001).

It is also chemically stable, which allows for long storage of foods. Condiments such as curry powder in South Africa have been successfully fortified with iron-EDTA. In some industrialized countries, EDTA has been extensively used as a stabilizer. The Joint FAO/WHO Expert Committee on Food Additives (JECFA) examined the existing data on iron-EDTA and found no objection to its use at a level of 2.5 mg/kg of body weight per day (FAO/WHO, 1993).

Binding of EDTA with iron is favored in the acid environment of the stomach, but in the more alkaline medium of the duodenum the iron is exchanged, in part, with other metals. The iron released from EDTA is absorbed by the normal physiological mechanisms. When NaFeEDTA is present in a meal, the iron moiety exchanges with the intrinsic food iron and the EDTA partially protects the iron in this common non-heme iron pool from the effects of inhibitors of iron absorption, such as phytates and polyphenols (Bothwell and Macphail, 2004).

When iron is added as NaFeEDTA to an inhibitory meal, it is two to three times better absorbed than iron added as ferrous sulfate. It also has a similar effect on the intrinsic food iron in the meal. Fortification with NaFeEDTA is most efficacious when administered with cereal- and legume-based diets. Animal and human studies suggest that, NaFeEDTA has little or no effect on overall zinc metabolism (Bothwell and Macphail, 2004).

A study by Sun *et al.* (2007) suggested that, NaFeEDTA has the best efficacy in rectifying iron deficiency anaemia and improving iron status, and the effects were seen as early as two months after the beginning of the supplementation. NaFeEDTA when added at a lower level, it has better effects than FeSO4 and elemental iron in controlling iron deficiency anaemia and improving iron status in anaemic children while elemental iron is the least effective. Another study conducted in Kenya by Mutie *et al.* (2012) found the use of NaFeEDTA to be effective in improving iron status of children with iron deficiency anaemia.

2.10.2 Method of manufacture of FeNaEDTA

Ferric sodium ethylenediaminetetraacetate is a free-flowing yellow-brown powder with the chemical composition FeNa-EDTA.3H20. It is isolated *via* crystallization with a high level of chemical purity. The raw materials used in the manufacture of FeNaEDTA, ferric chloride ($FeCl_3$), tetrasodium EDTA (Na4-EDTA, and hydrochloric acid (HCI) are prepared internally at Azko Nobel Chemicals and are of technical grade quality. Although they are of technical grade quality, the levels of contaminating heavy metals are controlled by specification and are similar in levels to food grade materials (such as $FeCl_3$, <0.3 ppm Pb). FeNaEDTA is obtained by crystallization after adding an aqueous solution of ferric chloride to an aqueous solution of tetrasodium EDTA. Hydrochloric acid is used as a pH-adjusting agent. The production of FeNaEDTA is conducted in accordance with a Hazard Analysis Critical Control Points (HACCP) based food system (Sun *et al.*, 2007).

2.10.3 Safety assessment of iron EDTA

Sodium iron (Fe^{3+}) ethylenediaminetetraacetic acid (EDTA) has shown to have a significant beneficial effect on iron status by increasing iron bioavailability in human diets. The data collected and published over the past 20 to 30 years demonstrate that, iron EDTA is safe and effective for iron fortification of food products and meets the standards of "reasonable certainty of no harm". Based on these records, iron EDTA may be regarded as safe for the intended food uses and supplementing children with iron deficiency anaemia (Hembach *et al.*, 2000).

The safety of iron EDTA is based on the views of experts who are qualified by scientific training and experience to evaluate the safety of FeNaEDTA as a component of food. The safety of FeNaEDTA is supported by a number of published studies on ferric sodium EDTA and other sources of EDTA, including *in vitro* and *in vivo* mutagenicity/ genotoxicity studies, metabolic studies, acute, sub-chronic and chronic, and reproductive and developmental toxicity studies in experimental animals, as well as numerous clinical studies investigating the effects of food fortification with ferric sodium EDTA (Sun *et al.*, 2007).

Studies evaluating the nutritional safety of FeNaEDTA in both rats and humans have demonstrated that, FeNaEDTA does not adversely affect the absorption of other nutrients, including zinc, copper, and calcium. The results of toxicological and clinical studies of FeNaEDTA and other sources of EDTA and iron support the safe intake of FeNaEDTA by humans to a level of 2.5 mg/kg body weight/day (Heimbach *et al.*, 2000).

Significant improvement in iron status indicators were reported in the fortified groups compared to the controls, with no evidence of iron overload. The fastest and greatest responses to iron fortification were observed in individuals of the fortified and control groups who were iron-deficient at the start of the fortification study, compared to their iron-repleted peers (Sun *et al.*, 2007).

2.11 Parasitic Infection and Iron Deficiency Anaemia

Parasitic infection including *Plasmodium falciparum* and helminthes infestation largely contributes to iron deficiency and eventually anaemia because they feed on red blood cells leading to blood loss hence IDA.

2.11.1 Hook worms and iron deficiency anaemia

Parasitic worms contribute to iron-deficiency anaemia among children. Predominant species being the hookworms *Ancylostoma duodenale* and *Necator americanus* (which inhabit the gut) and *Schistosoma spp* (which inhabit the blood vessels surrounding the gut and bladder) (Guyatt *et al.*, 2001). Hookworm infestation in humans is caused by nematode parasites *Necator americanus* and *Ancylostoma duodenale* and is transmitted through contact with contaminated soil. It is one of the most common chronic infestations, with an estimated 740 million cases in areas of rural tropics and subtropics (Hotez *et al.*, 2004).

The amount of blood loss which leads to iron-deficiency anaemia depends on the intensity of infestation, the dietary intake of iron and the presence of other parasitic diseases that can cause haemolysis, such as malaria and *Trichuris trichiura* infestation (Guyatt *et al.*, 2001). The major clinical manifestation of hookworm infestation includes iron deficiency anaemia caused by intestinal blood loss. Iron-deficiency anaemia and hypoalbuminemia develops when blood loss exceeds iron intake and reserves. Depending on the status of host iron, hookworm burden (i.e. the intensity of infestation, or number of worms per person) of 40 to 160 worms is associated with hemoglobin levels below 11 g/dl (Hotez *et al.*, 2004).

School-age children are particularly vulnerable to iron-deficiency anaemia exacerbated by parasitic infestation. A recent analysis of the association between hookworm and anaemia in school-age children in Zanzibar suggested that, 25% of all anaemia cases, 35% of iron-deficiency anaemia cases, and 73% of severe anaemia cases could be attributed by hookworm infestation (Stoltzfus, 1997). Hookworm infestation has been shown to contribute to iron deficiency anaemia more than schistosomiasis or malaria in school-age children (Guyatt *et al.*, 2001)

Most of the physical signs of chronic hookworm infestation reflect the presence of iron-deficiency anaemia. The skin becomes waxy and acquires a sickly yellowish color (a feature of tropical chlorosis). For many common helminthic infestations, including ascariasis, trichuriasis and schistosomiasis, the intensity of infestation usually peaks during childhood and adolescence (Hotez *et al.*, 2004).

2.11.2 Malaria and iron deficiency anaemia

Malaria is an important cause of illness and death in many parts of the world, especially in sub-Saharan Africa. Recently, there has been a renewed emphasis on preventive measures at community and individual levels. Children should not be denied iron supplements, even if they are living in areas where malaria is prevalent. Iron is important for growth and development, and maintaining a healthy immune system (Lengeler, 2006). Guidelines recommends that, all children should be given iron supplements to help prevent iron deficiency and anaemia, which is a significant public health problem with an estimation of 726 000 childhood deaths each year in developing countries (Lengeler, 2006).

A recent large trial in Zanzibar, Tanzania and Nepal prompted the WHO to change its guidelines, which now recommend that routine iron supplements are withheld from children under two years in areas where malaria prevalence is high (Ojukwu *et al.*, 2009). The argument against giving iron is that, it could help promote the growth of malarial parasites circulating in the blood. Iron supplementation is recommended only for children with proven iron deficiency. If a screening system to detect iron deficiency is not available, supplementation is recommended only for children with clinical symptoms of severe anaemia. In response to this, cochrane researchers (Ojukwu *et al.*, 2009) reviewed data from 68 different trials involving 42 981 children. They concluded that iron did not increase the risk of malaria, as long as regular malaria surveillance and treatment services are available (Ojukwu *et al.*, 2009).

Iron supplementation does not increase the risk for malaria or death when given to children with anaemia. Although the benefits of giving iron are greater for children with anaemia, any decision to withhold iron supplements should need to be carefully considered. Any potential negative effects of giving iron have to be weighed against the serious implications of not giving it, considering its contribution to childhood infestation and death, especially in sub-Saharan Africa (Ojukwu *et al.*, 2009).

2.12 Assessment of Nutrition Status

Nutrition problems are complex in their etiology and the type of information that can be useful is the assessment of the nutrition status. Nutritional assessment refers to as interpretation of information obtained from dietary, biochemical, anthropometrics and clinical studies (Saito and Mark, 1999). The goal of nutritional assessment and intervention is to improve nutritional status, prevent further complications and boost the child's quality of life and survival. Nutritional assessment is important to gather information on the current nutrition status and the adequacy of the diet and helps to identify risk factors for developing nutritional complications. The earlier the intervention and more consistently can be done the better. The information gathered is usually interpreted to identify problems that lay children at an increased risk and to design the best intervention with caregivers. The assessment helps to capture information about changes in nutritional status i.e. weight for age (WAZ), weight for height (WHZ) height for age (HAZ). These three indices are expressed as standard deviation (SD) units from the median for the international reference population recommended for healthy populations (WHO, 2006).

2.13 Methods Used in Nutritional Assessment

Nutritional assessment systems utilize a variety of methods to characterize each stage in the development of a nutritional deficiency state. The methods are based on a series of dietary, biochemical, anthropometric and clinical assessment.

2.13.1 Dietary assessment methods

It involves measuring the quantity and quality of foods consumed, one to several days or assessing the pattern of food consumed during the previous days or months. The first stage of nutritional assessment involves dietary assessment. Dietary intake may appear to meet nutritional needs but conditioning factors (such as disease states) may interfere with the ingestion, digestion, absorption and/ or utilization of nutrients (Gibson, 1990).

2.13.2 Anthropometric measurement methods

Nutrition anthropometry is the measurement of the variation of physical dimensions and the gross composition of human body at different age levels and degree of nutrition (Gibson, 1990). In practice, anthropometry is the most useful tool for assessing nutritional status of children because disturbances in health and nutrition, regardless of etiology invariability affect children's growth (Joosje *et al.,* 1997). The anthropometric measurements that are most commonly used for assessing the nutritional status are growth parameters like weight, height, mid upper arm circumference, age and body composition measures which comprises of fat mass and fat free mass or lean body mass (WHO, 2006). Anthropometric indicators are globally used as the basis for assessing growth and nutritional status in children.

The anthropometric indices that are used as criteria for assessing nutritional status are weight-for-age (WAZ), height-for-age (HAZ), and weight-for-height (WHZ). These indices are compared with the recommended reference for healthy populations (WHO, 2006). Stunting (HAZ), wasting (WHZ) and underweight (HAZ) are common forms of under-nutrition. According to (Caulfield *et al.*, 2006) under- nutrition is generally characterized by comparing the weights or heights (or lengths) of children at a specific age and sex with the distribution of observed weights or heights in a reference population of presumed healthy children of the same age and sex and then calculating *z*-scores, that is, the difference between a child's weight or height and the median value at that age and sex in the reference population, divided by the standard deviation (SD) of the reference population.

A child whose height-for-age is less than −2 SD is considered stunted, because the chances of the child's height being normal are less than 3 percent. A child whose weight-for-age is less than −2 SD is considered underweight, and one whose weight-for-height is less than −2 SD is deemed wasted (Caulfield *et al.*, 2006). Therefore stunting results from chronic under-nutrition, which retards linear growth, whereas wasting results from inadequate nutrition over a shorter period, and underweight encompasses both stunting and wasting (Caulfield *et al.*, 2006).

2.13.2.1 Weight-for-age-z-scores (WAZ)

Weight-for-age is a composite index of height-for-age and weight-for-height. It takes into account both chronic and acute malnutrition. Children with weight-for-age below minus two standard deviations are classified as underweight. Children with weight-for-age below minus three standard deviations (-3 SD) are considered severely underweight (NBS, 2011). Weight-for-age is a useful tool in clinical settings for continuous assessment of nutritional progress and growth.

2.13.2.2 Weight –for- height – z-score (WHZ)

The weight-for-height index measures body mass in relation to body height or length and describes current nutritional status. Children with Z-scores below minus two standard deviations (-2SD) are considered thin (wasted) or acutely malnourished. Wasting represents the failure to receive adequate nutrition in the period immediately preceding the survey and may be the result of inadequate food intake or a recent episode of illness causing loss of weight and the onset of malnutrition (NBS, 2011). Children with a weight-for-height index below minus three standard deviations (-3 SD) are considered severely wasted. The weight-for-height index also provides data on overweight and obesity. Children more than two standard deviations (+2 SD) above the median weight-for-height are considered overweight, or obese (NBS, 2011).

2.13.2.3 Height-for-age-z-scores (HAZ)

The height-for-age index provides an indicator of linear growth retardation and cumulative growth deficits in children. Children whose height-for-age Z-score is below minus two standard deviations (-2 SD) from the median of the WHO reference population are considered short for their age (stunted), or chronically malnourished. Children who are below minus three standard deviations (-3 SD) are considered severely stunted. Stunting reflects failure to receive adequate nutrition over a long period of time and is affected by recurrent and chronic illness (NBS, 2011). Height-for-age, therefore, represents the long-term effects of malnutrition in a population and is not sensitive to recent, short term changes in dietary intake.

2.13.3 Biochemical methods

Biochemical methods measure the nutrient or its metabolite in the body fluids or variety of other components that have relation to nutritional status. Nutritional deficiencies may be detected due to either the reduction of its levels or its metabolites in certain tissues or body fluids (static tests) or by functional tests i.e. physiological or behavioral changes of functions dependent on specific nutrients (Gibson, 1990).

2.13.4 Clinical methods

According to Gibson (1990) clinical assessment methods uses physical examinations to detect signs and symptoms associated with under nutrition. Since signs and symptoms are non-specific and only develop during the advanced stages of nutritional depletion, diagnosis of nutrition deficiencies should not rely exclusively on clinical methods. The clinical assessment methods should be backed-up with either biochemical or anthropometric measurements.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Description of the Study Area

The study was conducted in Turiani Division, one of the malaria endemic areas in Mvomero District, Morogoro region, Tanzania. Turiani Division is located at latitude 6°9′ South and longitude 37°36′ East. The study was conducted in four villages namely Kilimanjaro, Kichangani, Lusanga and Manyinga. Mvomero District has a total population of 259 347, out of whom 130 552 are females and 128 795 are males. The average household size is 5 ±1.96 (range, 2 to 12) with the average number of children under the age of five years of 1.4 ± 0.5 (range, 1 to 3) per household (NBS, 2002). Most (82%) of the people are employed in the agricultural sector, with majority practicing subsistence farming (NBS, 2005). Out of the study villages, Kichangani and Lusanga have health centers, while Manyinga village receives health services from Turiani hospital, which is a designated Mvomero District hospital. Major ethnic groups found in Mvomero District include Waluguru and Wanguu.

3.2 Study Design

A longitudinal study was conducted in which data were collected at baseline, at four weeks and at eight weeks. The study was a community–based randomized double –blind intervention trial involving supplementation of anaemic children with high or low dose of iron with intention to treat iron deficiency anaemia.

3.2.1 Sampling frame

All children aged 6-36 months who had lived in the villages for at least 3 months were eligible for the study. Children who were mentally ill, those with HIV/ AIDS and those with iron-related diseases such as sickle cell disease were excluded from the study. Excluded also were children with normal Hb levels (Hb concentrations > 11g/dl), severe anaemia (Hb concentrations ≤ 6.9 g/dl), severely wasted children (WHZ < -3.0 SD), children not consuming solid foods and those <6 months of age and > 36 months of age.

3.2.2 Sampling technique

Subjects were recruited from the Reproductive and Child Health (RCH) clinics. Mothers with small children (≤ 5 years) are usually required to send their under-five children to the RCH clinics once every month to monitor their weight growth and to have them vaccinated as needed. Mothers whose children were aged 6 to 59 months of age were invited to have their children screened for anaemia. A total of 1021 children were screened for iron status. Out of these 766 children were found to be anaemic. From the list of anaemic children, children meeting the following criteria were selected for the study; children having mild anaemia (Hb values from 7-10.9g/dl), children consuming some solid foods, and children not severely wasted. By random sampling, 217 children were selected for the iron supplementation study. The selected group of 217 children was divided into two groups. One group received a low dose iron supplement while the second group received a high dose iron supplement. A third group comprised of children aged 49 to 60 months did not receive micronutrient supplement initially. The purpose of the older group was to serve as a control group to assess malarial pressure during the study. However, those children in the third group who were anaemic received the iron supplement at the end of the eight week study. The distribution of children for each of the participating villages was Kilimanjaro (36), Kichangani (49), Manyinga (64) and Lusanga (68).

3.2.3 Sample size

The sample size was determined by using statistical power analysis using a method for health studies (Lwanga and Lemeshow, 1991). The decrease in IDA was 25% with 95% power. A total 217 children were selected to complete the iron supplementation study. Minimum subjects per treatment were 108. The attrition rate was 20 %.

3.2.4 Micronutrient supplementation

Two types of micronutrient supplements were given to the study group. One supplement (Treatment A) contained 10 mg of Fe as ferrous lactate while the second supplement (Treatment B) contained 5 mg of iron in the form of 2.5 mg Fe Na-EDTA and 2.5 mg of Fe lactate. The composition of the two supplements is summarized in Table 1.

Table 1: Composition of the supplements

Nutrients	Unit	Per1g product	Per 1g product
Vitamin A	IU	1250	1250
Vitamin D	IU	200	200
Vitamin E	mg	9	9
Vitamin B1	mg	0.5	0.5
Vitamin B2	mg	0.5	0.5
Vitamin B6	mg	0.5	0.5
Vitamin B12	mg	0.9	0.9
Vitamin C	mg	30	30
Nicotinamide	mg	6	6
Folic acid	mg	0.15	0.15
Iron	mg	10*	5**
Zinc	mg	4.1	4.1

*Iron derived from iron lactate
**2.5 of iron derived from ferric EDTA and 2.5 mg of iron derived iron lactate.

A pack containing 30 satchet was provided to the care provider. Each child was supposed to take one sachet per day. The contents of the satchet were supposed to be mixed with the child's food or drink. The food or drink was then supposed to be taken within a day. When the next 30-days supply of satchets was given to the care providers, any unused satchets were collected and recorded to monitor the usage compliance.

In addition, side effects and health information regarding malarial occurrence, diarrhea, fever, coughing, and other sicknesses were also recorded. The satchets were not marked in any way that could identify the amount or source of iron contained in the package.

3.2.5 Malaria and helminthes treatment

All children enrolled in the study were screened for malaria at baseline and in the subsequent weeks up to the end of the study. Children who tested positive of malaria at baseline, 4 weeks or 8 weeks were treated with quinine phosphate. During the eight-weeks of the study, children with fever were referred to the local health centre or to the district hospital for treatment. Children aged 12-59 (study children and the siblings) months of age were given three doses of 600 mg of melbendazole to control for helminthes. Children aged 6-11 months were not treated for helminthes.

3.3 Data Collection

Data were collected on a standardized form that had provision for comments. Two village health attendants for each participating village served as compliance monitors and also recorded food intake for all children on a daily basis. Dietary intakes were collected on a daily basis to allow identification of the children who consume foods rich in iron more frequently. Consumption of the foods rich in iron was one of the confounders to the study. Care givers were asked to list all food items consumed by the child in the past 24 hours including snacks. A questionnaire was constructed to enable collection of information during screening. Questionnaire was pre-tested in Mzumbe Health Centre in Morogoro Region. The questionnaire was translated into Kiswahili to facilitate smooth communication between the care providers and the enumerators. The questionnaires used during screening (Appendix 3), baseline study (Appendix 4), four (midterm) and eight weeks (end of term) of the study (Appendix 5) and for dietary intake (Appendix 6). Measurement on height (cm) and weight (kg) were taken to enable assessment of nutritional status of the children.

3.3.1 Anthropometric measurement

Standard techniques and equipment (weighing scale for weight, length board and studiometer for height) was used for collecting anthropometric measurement. Age was determined from the growth monitoring cards.

3.3.1.1 Weight measurement

Weight was measured using a SECA digital scale. The weights were taken without shoes on and with minimum clothing. Weights were recorded to the nearest 0.1 g.

3.3.1.2 Height measurement

Length of children below two years was measured by using recumbent length board, while for children above two years the linear height was measured by a studiometer. Measurements of length/height were recorded to the nearest 0.1 cm.

3.3.2 Determination of hemoglobin concentration

Capillary blood samples were obtained via finger prick with a lancet. Hemoglobin concentrations were measured using a HemoCue® Photometer (HemoCue AB, Angelholm, Sweden) at base-line, and after consuming the supplements at four and eight weeks of the study. A piece of cotton wood was dipped in alcohol and used to sterilize the tip of the left hand ring finger. The sterilized finger was thereafter dried with cotton wool and pricked with a sterile lancet. The first drop of blood was wiped out using a sterile piece of cotton. The second drop of blood was collected into a microcuvette which was then inserted in the HemoCue photometer. The photometer read the concentration of Hb which was recorded to the nearest 0.1 g/dl.

Plate 1: Mothers brought their children for taking measurements

Plate 2: A researcher taking height measurement from a child

3.3.3 Determination of malaria parasites

At base-line, blood smears for all children were prepared for screening malaria parasites. A piece of cotton was dipped in alcohol and used to sterilize the tip of the left hand ring finger. The tip of ring finger was thereafter pricked with a lancet. A sterile glass slide was used to collect the blood smear. The slides with blood smear were dried in air before staining with Giemsa and covered with oil. The smears were stained with Giemsa and read under light microscope at the laboratories of the Turiani Hospital. Each slide was examined by an experienced laboratory technician at a magnification of ×100. Presence of trophozoites or rings on the blood film was taken as a laboratory evidence of malaria. Parasite counts were done using a standard WHO (1991) method. Occurrence of Plasimodium falcipurum *(PF)* infection in the three groups (receiving high, low doses of irons and the siblings) at baseline (malaria prevalence) was determined by dividing number of cases per health population at a given point in time. A smear with two or more trophozoite counts or ring forms was considered as positive for *PF* infection and hence presence of malaria.

3.4 Data Analysis

Data analysis was made by the use of Statistical Package for Social Science (SPSS) whereby descriptive statistics such as frequency; percentage, Paired-Sample t- test and Analysis of Variance (ANOVA) were used to analyze the data. Baseline Hb concentrations for the two groups were compared by the Paired-Sample t-test. Differences between base-line Hb concentrations and four week and eight week Hb concentrations within a group were compared by ANOVA. Comparison of Hb differences (8 week Hb concentration-baseline Hb concentration) between the two supplement treated groups were done by ANOVA. Anthropometric measurements height- for age (HAZ), weight-for age (WAZ), weight-for height (WHZ) z-scores was computed using the Anthro 2005 software.

Dietary intake information was computed by using SPSS in which frequencies of consumption of different foods were obtained. Occurrence of Plasimodium falcipurum was determined by dividing number of cases per health population at a given point in time. Linear regression was used to determine the association between Hb concentration and anthropometric characteristics. Significance level was set at 5%.

3.5 Ethical Considerations

The study received ethical approval from the National Institute for Medical Research Ethics Committee prior to commencement. Informed written consent was obtained from the children's caregivers before participation in the study. Permission was also obtained from the Mvomero District health authorities to conduct the study in the selected villages.

3.6 Confidentiality

All children were identified through their respective village health centres and were assigned identity numbers. These identity numbers were used during data collection, data analysis and reporting. Children were not identified by names.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Demographic and Socioeconomic Characteristics

Results of the screening showed that, out of the 1021 children under the age of five years, 522 (51.12%) were girls and 499 (48.87%) boys.

4.1.1 Age and sex distribution

Table 2 presents distribution of children who were screened according to age and sex. Distribution of males and females children ranged between 6 and 59 months (Table 2).

Table 2: Distribution of age and sex of the children during screening

Age (mo)	Males		Females		Total	
	n	%	n	%	n	%
6-17	196	19.2	215	21.0	411	40.2
18-29	184	18.0	191	18.7	375	36.8
30-41	112	11.0	109	10.7	221	21.6
42-53	7	0.7	6	0.6	13	1.3
54-59	0	0.0	1	0.1	1	0.1
Total	**499**	**48.9**	**522**	**51.1**	**1021**	**100**

4.1.2 Anthropometric characteristics of the screened children

The Table 3 summarizes the anthropometric characteristics of the children in the study area (N= 1021). More than 35% (95% CI 30%, 40%) of the screened children were stunted (HAZ < -2SD), 12.1% (95% CI 6.4%, 17.8%) were underweight (WAZ < -2SD) while 2.4% (95% CI 0, 8.4%) were wasted (WHZ < -2SD). More males were stunted (43.7%; 95% CI 39%, 48%) than their female counterparts (28%; 95% CI 24%, 32%). The result correlates with the study by Tanzania Demographic and Health Survey (NBS, 2011) which found the levels of stunting among children under the age of five years to be higher among boys (45.6%) than among girls (38.5%).

Likewise more males were underweight (13.4%; 95% CI 5%, 22%) compared to females of the same age (10.9%; 95% CI 3%, 19%). The proportion of wasting between males and females were almost similar (Table 3). The common forms of under-nutrition (stunting, underweight and wasting) begin at the age of six months, as children start consuming complementary foods that are often inadequate in micro-and macronutrients (Caulfield *et al.*, 2006).

Table 3: Anthropometric characteristics of the screened children

	Stunting (HAZ)< -2 SD		Wasting (WHZ) < -2 SD		Underweight (WAZ)< -2 SD		Normal >-2SD-2SD	
	n	%	n	%	n	%	n	%
All (1021)	364	35.70	25	2.40	124	12.10	508	49.8
Males (499)	218	43.70	11	2.20	67	13.40	203	20.0
Females (522)	146	28.00	14	2.70	57	10.90	305	29.8

4.1.3 Plasma haemoglobin concentration during screening

Mean plasma haemoglobin concentration of boys was 8.4 g/dl while for girls was 8.5g/dl (Table 4). The overall plasma haemoglobin concentration for all children was 8.4g/dl. According to Tanzania Demographic and Health Survey (NBS, 2011) anaemia is categorized into three groups namely severe, moderate and normal. Severe anaemia is classified as Hb < 7 g/dl, moderate anaemia as Hb 7-9.9 g/dl and normal iron as Hb >11g/dl. On the basis of this classification, all children involved in the study were moderately anaemic (Hb 7- 11 g/d). The most common cause of anaemia is nutritional anaemia resulting from inadequate dietary intake of nutrients necessary for synthesis of haemoglobin, such as iron, folate and vitamin B12. Anaemia also results from sickle cell disease, malaria, or parasitic infections (NBS, 2011).

Table 4: Mean plasma haemoglobin concentration of the screened children

	Number of children	Mean Hb (g/dl)
All	1021	8.4
Males	499	8.4
Females	522	8.5

4.2 Demographic Characteristics of Children Involved in the Iron Supplementation Study

4.2.1 Age distribution

Children who received high dose of 10 mg of ferrous lactate and low dose of 5 mg of 2.5 mg Na-Fe-EDTA and 2.5 mg of ferrous had age range between 6-36 months (Table 5).

Table 5: Age distribution of children involved in the iron supplementation study

Age of children (months)	Iron supplement 10 mg[1] n	%	5 mg[2] n	%
6	3	4.3	2	2.8
7 - 12	29	41.4	27	38.0
13 - 36	38	54.3	42	59.2

[1] - Iron supplement given as 10mg Ferrous Lactate

[2]- Iron supplement given as 2.5 mg of Na-Fe- EDTA and 2.5 mg of Ferrous Lactate

4.2.2 Distribution of children involved in the iron supplementation by gender

Result in Table 6 summarizes the distribution of children involved in the iron supplementation by gender. More than 26 % (95% CI 12%, 40%) and 29 % (95% CI 15%, 43% of males received 10 mg and 5 mg of iron supplement, respectively, while more than 23 % (95% CI 9%, 38%) and 21 % (95% CI 14%, 29%) of females received 10 mg and 5 mg of iron supplement, respectively (Table 6).

Table 6: Gender distribution of children involved in the iron supplementation study

	Treatment received			
Gender	**10 mg[1]**		**5 mg[2]**	
	n	**%**	**n**	**%**
Males	37	26.2	41	29.1
Females	33	23.4	30	21.3
Total	**70**	**49.6**	**71**	**50.4**

[1] - Iron supplement given as 10mg Ferrous Lactate

[2]- Iron supplement given as 2.5mg of Na-Fe-EDTA and 2.5mg of Ferrous Lactate

4.2.3 Weight and height distribution of the studied children

Children in the iron supplementation study had a mean weight of 9.3 kg for both males and females at baseline (range 6.2 to 14 kg) (Table 7). Mean height at baseline was 74.6 cm while the mean height at the end of the study period was 77.6 cm (range 61 to 94 cm). Average weight after the course of the study was 9.9 kg, with an increase of about 0.5 kg per child. The slight increase ($p>0.05$) in weight could be due to the effect of the supplements as some of the mothers and care providers reported that, their children's appetites and hence food intakes increased since they started using the iron supplement. The slightly improvement observed in weight of the children correlate with results of studies by Almeida *et al.* (2004) and Friel *et al.* (2003) who found no significant influence of iron supplementation on weight and height of the studied children.

Table 7: Mean weight and height of the studied children (N=141)

Weight or Height	Baseline		End of term	
(mean)	Males	Females	Males	Females
Mean weight (kg)	9.3	9.3	9.9	9.9
Mean height (cm)	74.6	74.6	77.6	77.6

4.3 Malaria Prevalence

The data in Table 8 showed that, malaria prevalence at baseline was 10.6% (95% CI 0%, 23%). Malaria incidences during the 4th and 8th weeks of iron supplementation were 9.7% (95% CI 0%, 23%) and 10.2% (95% CI 0%, 24%), respectively. This trend affirmed that, the prevalence of malaria remained almost constant for the entire study period. The prevalence rates of malaria observed in this study were slightly lower than those reported from other studies. According to NBS and ORC Macro (2008) reported malaria prevalence of 15.7 % in Morogoro. The variability could be due to the season in which the screening was done. Malaria is usually higher during rainy season and during warm and humid periods than at other times of the year. This study was done during cold season.

Table 8: Malaria[1] incidence before and during supplementation

Screening point	No of children screened	No of children with malaria	Incidence rate (%)
Baseline (0 mo)	217	23	10.6
Midterm (4 week)	195	19	9.7
End of term (8 week)	167	17	10.2

[1]- Malaria confirmed by presence of 2 or more trophozoites or ring forms

4.4 Variation in Plasma Hb Concentrations on the Course of the Study

Initially all children involved in the study were moderately anaemic (9.2 g/dl). After supplementation with 10 mg and 5 mg of iron, the average blood haemoglobin concentrations increased to 9.62 g/dl at midterm (4 weeks) and 11.1 g/dl at the end of the supplementation (8 weeks). The trend of increase in Hb concentrations was similar for children receiving low and high doses of iron supplement (Table 9). Improvement in Hb concentration was significant $p < 0.005$). A similar effect was reported in a study by Thu *et al.* (1999), in which daily and weekly supplementation of iron to children under the age 5 years reduced the prevalence of anaemia by > 50 % (daily) and > 10% (weekly).

Other studies by Iannott *et al.* (2009) and Sasawal *et al.* (2006), revealed that, supplementing children with 10 mg of iron resulted in a significant improvement in their Hb concentrations among iron-deficient or anaemic children so long as, there is treatment of clinical malaria or intermittent preventive treatment for malaria and worms infestation. Furthermore, (Sun *et al.*, 2007) indicated that, when NaFeEDTA was added to children's food it has better effects in controlling iron deficiency anaemia and improving iron status among anaemic children.

Table 9: Variation in average blood Hb concentration on the course of the study[1]

Treatment	Baseline	Midterm	End of term
10 mg[2]	9.2 ± 0.97	9.62 ± 1.34	11.1 ± 1.20
		P value = 0.001	P value = 0.001
5 mg[3]	9.2 ± 0.78	9.6 ± 1.40	11.0 ±1.29
		P value = 0.001	P value = 0.001

[1]-Average Hb concentrations ± standard deviation

[2]- Iron supplement given as 10mg Ferrous Lactate

[3]- Iron supplement given as 2.5mg of Na-Fe-EDTA and 2.5mg of Ferrous Lactate

4.5 Effect of Iron Supplementation on Plasma Haemoglobin Concetrations

4.5.1 Hb status of children receiving iron supplement by sex

The Table 10 summarizes the plasma haemoglobin concentrations of the children receiving high dose (10 mg) and low dose (5 mg) of iron at baseline, midterm and at the end of term by sex. Results of the study revealed that, 2.8% (95% CI 0%, 25 %.) of the children had severe anaemia at baseline. The proportion of children with severe anaemia was reduced to zero during the study period for both supplements. The proportion of moderately anaemic children at baseline was 98.6% (95% CI 96%, 100%) for both groups receiving high and low doses of iron.

Table 10: Hb status of children receiving iron supplement by sex[1]

Duration/Hb status[2]	Iron supplement					
	10 mg Fe lactate (n=70)			5 mg Fe Na-EDTA (n=71)		
	Male	Female	Total	Male	Female	Total
Baseline						
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	36(51.4)	33(47.1)	69(98.6)	40(56.3)	30(42.2)	70(98.6)
Severe	1(1.4)	0(0)	1(1.4)	1(1.4)	0(0)	1(1.4)
Midterm						
Normal	6(8.6)	7(9.8)	13(18.6)	8(11.3)	7(9.8)	15(21.1)
Moderate	31(44.3)	24(33.8)	55(78.6)	31(43.7)	23(32.4)	54(76.0)
Severe	0(0)	2(2.8)	2(2.8)	2(2.8)	0(0)	2(2.8)
End of term						
Normal	23(32.8)	19(27.1)	42(60.0)	17(23.9)	21(29.6)	38(53.5)
Moderate	14(20.0)	14(20.0)	28(40.0)	24(33.8)	9(12.7)	33(46.5)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)

[1]Values are presented as number (percent)
[2] Normal Hb = > 11.0 g/dl, Moderate anaemia, Hb= 7-10.9g/dl and Severe anaemia, Hb = < 7g/dl

During the midterm evaluation, moderately anaemic children were 78.0 % (95% CI 67%, 88%) for those receiving 10 mg of iron lactate supplement and 76.0% (95% CI 65%, 87%) for those receiving 5 mg of iron Na-EDTA. At the end of term, the proportion of moderately anaemic children was 40.0% (95% CI 22%, 58%) for those receiving 10 mg of iron lactate and 46.5% (95% CI 29.5%, 63.5%) for the group receiving 5 mg of iron Na-EDTA. The trend in the prevalence of moderate anaemia was decreasing for both groups receiving high and low dose of iron supplements, indicating improvement in the level of anaemia.

There were no children with normal haemoglobin concentration at baseline. However, the proportion of children with normal haemoglobin concentration at the end of the study was 60.0% (95% CI 45%, 75%) for those receiving 10 mg iron lactate and 53.5% (95% CI 37.7%, 69.3%) for those receiving 5 mg iron Na-EDTA. This indicated that, consumption of the supplements resulted in significant ($p<0.05$) improvement in the haemoglobin concentration. Also, presence of other micronutrients in the supplements particularly vitamin A, B12 and folic acid had contributed to the increased Hb due to their roles in iron absorption and utilization in the body (Mutie *et al.*, 2012). More males were anaemic at baseline (1.4%; 95% CI 0%, 24%) than their female counterparts (0%). The proportion of anaemic male children was reduced to zero at the end of the study period. Also, more males were moderately anaemic at baseline (51.4%; 95% CI 35%, 68%) compared to females (47.1%; 95% CI 30%, 64%) for the group receiving 10 mg iron lactate.

Likewise, more males (56.3%; 95% CI 41%, 72%) were moderately anaemic compared to the females (42.2%; 95% CI 25%, 60%) for the group receiving 5 mg of iron Na-EDTA supplement. For the group receiving 10 mg iron lactate, the proportion of moderately anaemic children was reduced to 44.3% (95% CI 27%, 62%) and 33.8% (95% CI 15%, 53%) for males and females, respectively, at mid-term evaluation and decreased further to 20% (95% CI 20%, 22%) for both males and females at the end of the study period. For the group receiving 5 mg of iron Na-EDTA, the proportion of moderately anaemic males was reduced to 43.7% (95% CI 26%, 50%) at midterm evaluation while the proportion of moderately anaemic females was reduced to 32.4% (95% CI 13.3%, 52%). Evaluation at the end of the intervention showed that, the proportion of moderately anaemic children were 33.8% (95% CI 15%, 53%) and 12.7 (95% CI 0%, 34%) for males and females, respectively, for children receiving 5 mg iron Na-EDTA supplement.

The proportion of moderately anaemic children receiving the higher dose of iron (10 mg iron lactate) was slightly higher ($p>0.05$) than that of children receiving a low dose of iron (5 mg iron Na-EDTA). At baseline, no children with normal haemoglobin concentration were observed. However, the proportion of normal children increased significantly ($p<0.05$) by midterm evaluation.

For the group receiving high dose of iron supplement (10 mg) the proportion of children with normal Hb increased from 0.0% to 8.6% (95% CI 0%, 31%) and 9.8% (95% CI 0%, 32%) for males and females, respectively. For the group receiving the low dose of iron (5 mg), the proportion of children with normal levels of haemoglobin concentration increased by 11.3% (95% CI 0%, 33%) and 9.8% (95% CI 0%, 32%) for males and females, respectively. At the end of the intervention, the proportion of children with normal Hb increased to 32.8% (95% CI 14%, 52%) (males) and 27.1% (95% CI 7%, 47%) (females) for the group receiving high iron dose and 23.9% (95% CI 4%, 44%) (males) and 29.6% (95% CI 10%, 49%) (females) for the group receiving low iron dose. Supplementation is the method of choice when therapeutic treatment is necessary to address severe micronutrient deficiencies. It is also an appropriate approach for preventive programs as long as the distribution system can be maintained and those receiving the supplements continue to consume (NAS, 1998). The iron supplement given to the children was therefore appropriate as it improved their haemoglobin concentrations. The study indicated that, receiving both high and low doses of iron supplementation resulted in significant improvement ($p<0.05$) in the Hb concentrations both at midterm (4 weeks) and at the end of the study (8 weeks).

These findings concur with those of Thu *et al.* (1999), in which daily supplementation of iron to children under the age five of years reduced the prevalence of anaemia. Also, the results by Sun *et al.* (2007) indicated that when NaFeEDTA was added to children's food it has better effects in controlling iron deficiency anaemia and improving iron status in anaemic children. Furthermore, a study conducted in Kenya by Mutie *et al.* (2012) found the use of NaFeEDTA to be effective in improving iron status of children with iron deficiency anaemia.

4.5.2 Hb status of children receiving iron supplement by age

The results in Table 11 indicates that, for children receiving the high dose of iron (10 mg) only 1.4% (95% CI 0%, 24%) of the children (at 6 months) were moderately anaemic at baseline but these children had their haemoglobin concentrations changed to normal at the end of the supplementation period. This trend was also similar for the same age (6 mo) children receiving the low dose (5 mg) of iron supplement. For children aged 7-12 months, 42.8% (95% CI 25%, 61%) of the children were moderately anaemic at baseline (high iron dose), but this proportion decreased ($p>0.05$) to 37.1% (95% CI 19%, 56%) at midterm evaluation and to 12.8% (95% CI 0%, 35%) at the end of supplementation. Their peers on low iron dose (5 mg), 42.2% (95% CI 25%, 60%) were moderately anaemic at baseline but this proportion decreased to 28.2% (95% CI 9%, 48%) at midterm and to 18.3% (95%

CI 0 %, 39%) at the end of the supplementation study. The supplement therefore, improved the Hb concentrations significantly ($p < 0.05$) and reduced the proportion of moderately anaemic children by 24-30 % in this age group (7-12 months).

For children aged 13-36 months, 57.7% (95% CI 43%, 73%) were moderately anaemic at baseline (high iron dose) but this proportion decreased to 46.5% (95% CI 30%, 64%) at midterm and to 28.2% (95% CI 9%, 48%) at the end of the supplementation period. For their counterparts receiving low iron dose (5 mg), 54.3% (95% CI 39%, 70%) were moderately anaemic at baseline, but this proportion decreased to 37.1% (95% CI 19%, 56%) at midterm and to 25.7% (95% CI 6%, 46%) at the end of the study. For this age group (13-36 months), supplementation with low and high doses of iron resulted in significant improvement in the Hb status of the children.

Iron supplementation reduced the proportion of moderately anaemic children in this age group by 29-30%. These data suggested that, prevalence of anaemia was higher among the older children groups (13-36 months) than the lower age groups (6 months and 7-12 months). Supplementation with both low and high doses of iron therefore appeared to have a greater improvement among the younger groups (100%) (6 months group) and 30- 40% (7-12 months group) than among the older group (29-30%, 13-36 months group).

Table 11: Hb status of children receiving iron supplement by age[1]

Age group/ Hb status[2]	Iron supplementation								
	Baseline			Midterm			End of term		
	Male	Female	Total	Male	Female	Total	Male	Female	Total
Children receiving 10mg Fe lactate (n=70)									
6 mo									
Normal	0 (0)	0(0)	0(0)	1(1.4)	0(0)	1(1.4)	1(1.4)	0(0)	1(1.4)
Moderate	1(1.4)	0(0)	1(1.4)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7-12 mo									
Normal	0(0)	0(0)	0(0)	0(0)	2(2.8)	2(2.8)	11(15.7)	9(12.8)	20(28.6)
Moderate	16(22.8)	14(20.0)	30(42.8)	17(24.3)	9(12.8)	26(37.1)	6(8.5)	3(4.3)	9(12.8)
Severe	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)	1(1.4)	0(0)	0(0)	0(0)
13-36 mo									
Normal	0(0)	0(0)	0(0)	6(8.6)	5(7.1)	11(15.7)	11(15.7)	9(12.8)	20(28.6)
Moderate	19(27.1)	19(27.1)	38(54.3)	13(18.6)	13(18.6)	26(37.1)	8(11.4)	10(14.3)	18(25.7)
Severe	0(0)	0(0)	0(0)	0(0)	1(1.4)	1(1.4)	0(0)	0(0)	0(0)
Children receiving 5mg Fe lactate(2.5mg Fe Na EDTA and 2.5 mg Fe lactate) (n=71)									
6 mo									
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	1(1.4)	0(0)	1(1.4)	1(1.4)	0(0)	1(1.4)	1(1.4)	0(0)	1(1.4)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7-12 mo									
Normal	0(0)	0(0)	0(0)	5(7.0)	1(1.4)	6(8.4)	7(9.8)	7(9.8)	14(19.7)
Moderate	19(26.8)	10(14.1)	30(42.2)	11(15.5)	9(12.7)	20(28.2)	10(14.1)	3(4.2)	13(18.3)
Severe	0(0)	0(0)	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	0(0)	0(0)
13-36mo									
Normal	0(0)	0(0)	0(0)	3(4.2)	6(8.4)	9(12.7)	10(14.1)	13(18.3)	23(32.3)
Moderate	22(30.9)	19(26.7)	41(57.7)	20(28.2)	13(18.3)	33(46.5)	13(18.3)	7(9.8)	20(28.2)
Severe	1(1.4)	0(0)	1(1.4)	0(0)	0(n0)	0(0)	0(0)	0(0)	0(0)

[1]Values are presented as number (percent)
[2]Normal Hb ≥ 11.0 g/dl, Moderate anaemia, Hb= 7-10.9g/dl and Severe anaemia, Hb < 7g/dl

4.6 Comparison of Hb Concentration between the Low and High Dose Groups

Data in Table 12 revealed that, for the two groups receiving 10 mg iron lactate and 5 mg iron Na-EDTA (2.5 mg Na-EDTA and 2.5 mg ferrous lactate) there were no significant differences ($p>0.05$) in Hb concentration among the children receiving the low or high dose of iron supplement. Between groups (that received different supplement) and within the groups (those received the same supplement) there were no significant differences ($p>0.05$) in terms of Hb concentration improvement after the consumption of the supplements for eight weeks.

Table 12: Comparison of the low and high dose of iron supplements between[1] and within[2] groups

Duration	Groups	Sum of Squares	Mean Square	F	P value
4 weeks	Between	.026	.026	.014	.906
	Within	262.147	1.886		
	Total	**262.173**			
8 weeks	Between	.579	.579	.370	.544
	Within	217.474	1.565		
	Total	**218.053**			

[1] - Comparison between low – high dose groups
[2]- Comparison for those received low-low and high-high dose

This suggested that, both high (10 mg) and low dose (5 mg) iron supplements were equally effective in improving the Hb concentrations of the studied children. The findings correlates those reported by Sun *et al.* (2007) who found NaFeEDTA has best efficacy in rectifying iron deficiency anaemia and improving iron status. Also, other studies by Lannott *et al.* (2009), Sasawal *et al.* (2006) revealed that, supplementing children with 10 mg (normal iron dose) resulted in significant improvement in their Hb concentrations among iron-deficient so long as, there is treatment of clinical malaria or intermittent preventive treatment for malaria and worms.

4.7 Nutrition Status of Children

Three standard indicators of growth were used, height-for-age; weight-for-height; and weight-for-age.

4.7.1 The Weight-for Age- Z-score (WAZ)

There were no severely underweight children in the study groups (Table 13). This was because children who were severely underweight were excluded from the study. At baseline, the proportion of moderately underweight children in the group receiving 10 mg of iron was 52.8% (95% CI 37%, 69%) and 42.8% (95% CI 25%, 60%) for males and females, respectively.

The proportions of underweight children for those receiving 5 mg of iron supplement at baseline were 42.2% (95% CI 25%, 59%) (males) and 35.2% (95% CI 18%, 52%) (females). The proportions of mildly underweight children at baseline were zero (males) and 1.4% (95% CI 0%, 24%) (females) (10 mg group) and 11.3% (95% CI 0%, 33%) (males) and (5.6%; 95% CI 0%, 28%) (females) for the 5 mg supplementation group.

Table 13: Changes in WAZ scores and Hb status of the studied children before and after iron supplementation by sex[1]

WAZ-score[3]/ Duration	Hb status[2]							
	Male				Female			
	Severe	Moderate	Normal	Total	Severe	Moderate	normal	Total
Children receiving 10mg Fe lactate (n=70)								
Baseline								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	2(2.8)	0(0)	2(2.8)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	1(1.4)	36(51.4)	0(0)	37(52.8)	0(0)	30(42.85)	0(0)	30(42.8)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Midterm								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	1(1.4)	2(2.8)
Mild	0(0)	2(2.8)	0(0)	2(2.8)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	0(0)	29(41.4)	6(8.6)	35(50.0)	2(2.8)	22(32.4)	6(8.6)	30(42.8)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
End of term								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	1(1.4)	2(2.8)
Mild	0(0)	1(1.4)	2(2.8)	3(4.3)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	0(0)	13(18.6)	21(30.0)	34(48.6)	0(0)	12(17.1)	18(25.7)	30(42.8)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Children receiving 5mg Fe lactate(2.5mg Fe Na EDTA and 2.5 mg Fe lactate) (n=71)								
Baseline								
Normal	0(0)	3(4.2)	0(0)	3(4.2)	0(0)	1(1.4)	0(0)	1(1.4)
Mild	0(0)	8(11.3)	0(0)	8(11.3)	0(0)	4(5.6)	0(0)	4(5.6)
Moderate	1(1.4)	29(40.8)	0(0)	30(42.2)	0(0)	25(35.2)	0(0)	25(35.2)
Severe	0(0)	0 (0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Midterm								
Normal	0(0)	2(2.8)	1(1.4)	3(4.2)	0(0)	1(1.4)	0(0)	1(1.4)
Mild	0(0)	7(9.8)	1(1.4)	8(11.3)	0(0)	2(2.8)	2(2.8)	4(5.6)
Moderate	2(2.8)	22(31.0)	6(8.4)	30(42.2)	0(0)	20(28.2)	5(7.0)	25(35.2)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
End of term								
Normal	0(0)	4(5.6)	0(0)	4(5.6)	0(0)	1(1.4)	0(0)	1(1.4)
Mild	0(0)	5(7.0)	5(7.0)	10(14.1)	0(0)	2(2.8)	2(2.8)	4(5.6)
Moderate	0(0)	15(21.1)	12(16.9)	27(38.0)	0(0)	6(8.4)	19(26.8)	25(35.2)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)

[1] Values are presented as number (percent)
[2]Normal Hb = > 11.0 g/dl, Moderate anaemia, Hb= 7-10.9g/dl and Severe anaemia, Hb = < 7g/dl
[3]Normal weight for age= SD>-1.0, Mild underweight=SD-1-- -1.9, Moderate underweight=-2-- -2.9 and Severe underweight= SD < -3.0

Generally, iron supplementation did not improve significantly (p>0.05) the nutrition status of the children as only 2.8% of the males receiving 10 mg of iron had their WAZ scores improved from moderate to mild malnutrition, while there was no change of the WAZ scores for the females receiving the same supplement. The results correlates with the study by Kounnavong *et al.* (2010) who found improvement in the weight for age z-score was very small and not statistically significant.

4.7.2 Height-for-age z-scores (HAZ)

A child who is below -2 SD from the median of the reference population in terms of height-for-age is considered to be stunted or short for his/her age. Table 14 summarizes the nutritional status of the studied children using height-for age z-scores before and after iron supplementation.

The effect of iron supplementation did not result in consistent effects on the HAZ in the groups receiving low and high doses of iron supplementation. For the group receiving 10 mg of iron supplement, the prevalence of moderate and mild stunting at baseline was 24.3% (95% CI 4%, 45%) and 15.7% (95% CI 0%, 37%) (males) and 28.6% 95% CI 9%, 48%) and 10.0% (95% CI 0%, 32%) (females), respectively. After the 8 weeks of iron supplementation, the moderate and mild stunting levels changed slightly (p>0.05) to 21.4% (95% CI 1%, 42%) and 14.3% (95% CI 0%, 36%) (males) and 20.0% (95% CI 0%, 41%) and 8.6% (95% CI 0%, 31%) (females), respectively.

While there was a slight improvement in the proportion of moderately stunted females (from 28.6% at baseline to 20.0 % at the end of the study), there was no improvement of HAZ scores for males on the course of iron supplementation.

Table 14: Changes in HAZ-scores and Hb status of the studied children before and after iron supplementation by sex[1]

HAZ-score[3]/ Duration	Hb status[2]							
	Male				Female			
	Severe	Moderate	Normal	Total	Severe	Moderate	normal	Total
Children receiving 10mg lactate								
Baseline								
Normal	0(0)	8(11.4)	0(0)	8(11.4)	0(0)	6(8.7)	0(0)	6(8.7)
Mild	0(0)	11(15.7)	0(0)	11(15.7)	0(0)	7(10.0)	0(0)	7(10.0)
Moderate	1(1.4)	17(24.3)	0(0)	18(25.7)	0(0)	20(28.6)	0(0)	20(28.6)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Midterm								
Normal	0(0)	6(8.6)	2(2.8)	8(11.4)	0(0)	4(5.7)	2(2.8)	6(8.7)
Mild	0(0)	10(14.3)	1(1.4)	11(15.7)	1(1.4)	6(8.6)	0(0)	7(10.0)
Moderate	0(0)	15(21.4)	3(4.3)	18(25.7)	1(1.4)	14(20.0)	5(7.1)	20(28.6)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
End of term								
Normal	0(0)	1(1.4)	4(5.7)	5(7.1)	0(0)	2(2.8)	4(5.7)	6(8.7)
Mild	0(0)	6(8.7)	5(7.1)	11(15.7)	0(0)	3(4.3)	7(10.0)	10(14.3)
Moderate	0(0)	7(10.0)	14(20.0)	21(30.0)	0(0)	9(12.6)	8(11.4)	17(24.3)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Children receiving 5 mg (2.5 Fe-Na – EDTA and 2.5 Fe- lactate								
Baseline								
Normal	0(0)	4(5.6)	0(0)	4(5.6)	0(0)	5(7.0)	0(0)	5(7.0)
Mild	0 (0)	11(15.5)	0(0)	11(15.5)	0(0)	4(5.6)	0(0)	4(5.6)
Moderate	1(1.4)	25(35.2)	0(0)	26(36.6)	0(0)	21(29.6)	0(0)	21(29.6)
Severe	0 (0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Midterm								
Normal	0(0)	3(4.2)	1(1.4)	4(5.6)	0(0)	3(4.2)	2(2.8)	5(7.0)
Mild	0(0)	9(12.7)	2(2.8)	11(15.5)	0(0)	3(4.2)	1(1.4)	4(5.6)
Moderate	2(2.8)	19(26.7)	5(7.0)	26(36.6)	0(0)	17(23.9)	4(5.6)	21(29.6)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
End of term								
Normal	0(0)	4(5.6)	0(0)	4(5.6)	0(0)	4(5.6)	0(0)	4(5.6)
Mild	0(0)	5(7.0)	5(7.0)	10(14.1)	0(0)	1(1.4)	5(7.0)	6(8.4)
Moderate	0(0)	15(21.1)	12(16.9)	27(38.0)	0(0)	4(5.6)	16(22.5)	20(28.2)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)

[1] Values are presented as number (percent)
[2]Normal Hb = > 11.0 g/dl, Moderate anaemia, Hb= 7-10.9g/dl and Severe anaemia, Hb = < 7g/dl
[3]Normal height for age= SD>-1.0, Mild stunting=SD-1- - -1.9, Moderate stunting=-2-- -2.9 and Severe stunting= SD < -3.0

Similar inconsistent results on the HAZ scores were observed for the group receiving the low dose iron (5 mg). The proportion of moderately and mildly stunted males was slightly higher (62.3% and 31.2%, respectively) ($p>0.05$) than that of their female peers (58.2% and 15.6%, respectively) at baseline.

After supplementation, the proportion of moderately and mildly stunted males remained higher (68.0% and 29.8%, respectively) ($p>0.05$) than that of their female counterparts (52.5 and 22.7%, respectively). For the entire study period, there was only slight ($p>0.05$) improvement in the linear growth (HAZ scores) of both males and females receiving low and high dose of iron supplement.

This could be due to the fact that, linear growth usually takes place very slowly and takes a long time for any increase in height to be noticed (King and Burgess, 1993). According to (NBS, 2011) prevalence of moderate stunting at national level was 45.6% and 38.5% for male and female children under the age of five years, respectively. Prevalence levels for moderate stunting observed in this study were higher than those reported at national level and higher than values (44.4%) reported at regional level.

Prevalence of moderate stunting among males in this study ranged between 62.3% and 68.0%, while for females it ranged between 58.2.% and 52.5%. Prevalence of mild stunting among males in this study ranged between 31.2% and 29.8% while for females it ranged between 15.6% and 22.7%. Prevalence of stunting did not change significantly ($p> 0.05$) on the course of iron supplementation (for both low and high doses of iron supplementation), because the rate of linear growth is usually very slow (King and Burgess, 1993).

4.7.3 Weight –for- Height Z-score (WHZ)

Children whose weight-for-height is below -2 SD from the median of the reference population are considered wasted or thin, a condition reflecting acute malnutrition (NBS, 2011). Table 15 summarizes the weight-for height z-scores of the studied children before and after iron supplementation. The study revealed that, for children receiving 10 mg iron lactate, the proportion of moderately wasted children was 98.5% (95% CI 96%, 100%) at baseline, in which the proportion of moderately wasted males was 52.8% (95% CI 37%, 69%) and 45.7% (95% CI 23%, 69%) for females.

At the end of the study, the proportion of moderately wasted males decreased to 18.6% (95% CI 0%, 39%) whereas for females the proportion decreased to 20.0% (95% CI 0%, 40%). There were no mildly wasted children among male children at baseline and at midterm. The percentage of mildly wasted female children was 1.4% (95% CI 0%, 24%) at baseline and at midterm. There were no females who were mildly wasted at the end of the study. According to the mothers, consumption of the supplement increased children's appetite and eventually increased dietary intake. Increase in dietary intake may have slightly improved ($p>0.05$) the weight for height status of the studied children.

Table 15: Changes in WHZ – scores and Hb status of the studied children before and after supplementation by sex[1]

WHZ-scores[3]/ Duration	Hb status[2] Male Severe	Moderate	Normal	Total	Female Severe	Moderate	Normal	Total
Children receiving 10 mg Fe lactate								
Baseline								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0 (0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	1(1.4)	36(51.4)	0(0)	37(52.8)	0(0)	32(45.7)	0(0)	32(45.7)
Severe	0 (0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Midterm								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	1(1.4)
Moderate	0(0)	31(44.3)	6(8.6)	37(52.8)	2(2.8)	24(34.38)	6(8.6)	32(45.7)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
End of term								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	1(1.4)
Mild	0(0)	1(1.4)	1(1.4)	2(2.8)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	13(18.6)	22(31.4)	35(50.0)	0(0)	14(20.0)	18(25.7)	32(45.7)
Severe	0 (0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Children receiving 5 mg (2.5 Fe-Na – EDTA and 2.5 Fe- lactate								
Baseline								
Normal	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)
Mild	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	1(1.4)	38(53.5)	0(0)	39(54.9)	0(0)	28(39.4)	0(0)	28(39.4)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mid term								
Normal	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)
Mild	0(0)	1.(1.4)	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	0(0)	29(40.8)	10(14.1)	39(54.9)	0(0)	21(29.6)	7(9.8)	28(39.4)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
End of term								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	3(4.2)	1(1.4)	4(5.6)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	21(29.6)	16(22.5)	37(52.1)	0(0)	9(12.7)	21(29.6)	30(42.2)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)

[1] Values are presented as number (percent)
[2]Normal Hb => 11.0 g/dl, Moderate anaemia, Hb= 7-10.9g/dl and Severe anaemia, Hb =< 7g/dl
[3]Normal weight for height= >-1.0SD, Mild wasting= -1SD-- -1.9, Moderate wasting=-2SD-- -2.9 and Severe wasting= < -3.0 SD.

For children receiving 5 mg iron Na-EDTA, the proportion of moderately wasted children was 94.3% (95% CI 5%, 99%) at baseline. The proportion of moderately wasted males at baseline was 53.5% (95% CI 38%, 69%) while for females was 39.4% (95% CI 21%, 57%). At midterm evaluation, the proportion of moderately wasted male children was 40.8% (95% CI 23%, 58%) and females was 29.6% (95% CI 10%, 49%). At the end of the study, the proportion of moderately wasted males decreased from 40.8 to 29.6 % while the proportion of moderately wasted females decreased from 39.4 to 12.7%. This decrease could have been brought about by an increase in dietary intake due to increased appetite caused by the supplement. The proportion of children who were mildly wasted at baseline was 1.4% (95% CI 0%, 24%) for males and 1.4% (95% CI 0%, 24%) for females.

During midterm evaluation, the proportion remained the same for both males and females. The proportion of mildly wasted males was 4.2% (95% CI 0%, 27%) while for females was 0.0% at the end of supplementation period. It was reported by the mothers during the study that, consumption of 5 mg iron Na-EDTA supplements increased children's appetite and eventually increased dietary intake. The slightly improvement observed in weight for height status of the children correlate with results of studies by Almeida *et al.* (2004), Friel *et al.* (2003) who found no significant influence of iron supplementation on the prevalence of wasting.

4.8 Variation in Anthropometric Measurements with the Hb Status

Children involved in the intervention were in the age range 6-36 months. Table 16 summarizes the variation of WAZ-score and Hb status with age of the children after iron supplementation.

Table 16: Variation of WAZ-scores and Hb status with age of the children[1]

WAZ z-scores[3]/ Age (Mo)	Hb status[2] Male Severe	Moderate	Normal	Total	Female Severe	Moderate	Normal	Total
Children receiving 10 mg Fe lactate (n=70)								
6 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	0(0)	1(1.4)	1(1.4)	0(0)	1(1.4)	1(1.4)	2(2.8)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7 - 12 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	6(8.6)	11(15.7)	17(24.3)	0(0)	3(4.3)	9(12.8)	12(17.1)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
13 - 36mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	1(1.4)	2(2.8)
Mild	0(0)	1(1.4)	2(2.8)	3(4.3)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	0(0)	7(10.0)	9(12.8)	16(22.8)	0(0)	8(11.4)	8(11.4)	16(22.8)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Children receiving 5mg (2.5 mg Fe Na -EDTA and 2.5 mg Fe lactate) (n=71)								
6 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	0(0)	1(1.4)	1(1.4)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7 - 12 mo								
Normal	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	0(0)	1(1.4)
Moderate	0(0)	9(12.7)	7(9.8)	16(22.5)	0(0)	2(2.8)	7(9.8)	9(12.7)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
13 - 36 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	0(0)	1(1.4)
Mild	0(0)	7(9.8)	1(1.4)	8(11.3)	0(0)	1(1.4)	2(2.8)	3(4.2)
Moderate	0(0)	6(8.4)	9(12.7)	15(21.1)	0(0)	4(5.6)	11(15.5)	15(21.1)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)

[1] Values are presented as number (percent).

[2]Normal weight for age= >-1.0SD, Mild underweight=-1SD--1.9SD, Moderate underweight=-2SD- -2.9SD and Severe underweight= < -3.0SD.

[3]Normal Hb > 11.0 g/dl, Moderate anaemia, Hb 7-10.9g/dl and severe anaemia, Hb < 7g/dl.

Results showed that, for those children who were supplemented with 10 mg of iron lactate there were no children with severe anaemia among all age groups. For children in age group 6 months, prevalence of moderate underweight with normal Hb was 1.4% (95% CI 0%, 24%) for both males and females. There were no moderately underweight males with normal Hb but for females the prevalence was 1.4% (95% CI 0%, 24%). For children aged 7-12 months, prevalence of moderately underweight children with moderate anaemia was 8.6% (95% CI 0%, 31%) for males and 4.3% (95% CI 0%, 27%) for females.

The proportion of moderately underweight children with normal Hb was 15.7% (95% CI 0%, 37%) (males) and 12.8% (95% CI 0%, 35%) (females). For age group 13-36 months results showed that, prevalence of normal weight for age with moderate anaemia among females was 1.4% (95% CI 0%, 24%) while for males the prevalence was zero. Prevalence of normal weight for age with normal Hb was 1.4% (95% CI 0%, 24%) (females) and zero for males. For both males and females the percentage of mildly underweight children with moderate anaemia was 1.4% (95% CI 0%, 24%). The proportion of males who were mildly underweight with normal Hb was 1.4% (95% CI 0%, 24%) while for females the proportion was zero.

For moderately underweight children, the proportion of males with moderate anaemia was 10.0% (95% CI 0%, 32%) while for females 11.4% (95% CI 0%, 33%). Children with normal Hb but moderately underweight were 12.8% (95% CI 0%, 35%) (males) and 11.4% (95% CI 0%, 33%) (females). For children supplemented with 5 mg iron Na-EDTA, there were no children with severe anaemia among all age groups at the end of the study period. For children aged 6 months, the proportion of males who were moderately underweight with moderate anaemia were 1.4% (95% CI 0%, 24%) while for females the proportion was zero. The proportion of children who were moderately underweight with normal Hb was 1.4% (95% CI 0%, 24%) for females while for males was zero. For age group 7-12 months, the proportion of children with normal weight for age with moderate anaemia was 1.4% (95% CI 0%, 24%) (males) while for females the proportion was zero.

Prevalence of mildly underweight children with moderate anaemia was 1.4% (95% CI 0%, 24%) (females) whereas for males the prevalence was zero. The proportion of moderately underweight children with moderate anaemia were 12.7% (95% CI 0%, 34%) (for males) and 2.8% (95% CI 0%, 25%) (for females). Those with normal Hb but moderately underweight were 9.8% (95% CI 0%, 32%) for both male and female subjects. For the age group 13-36 months, the proportion of children with normal weight for age with moderate anaemia was 1.4% (95% CI 0%, 24%) (females) whereas for males the proportion was zero. Percent of children in this age group who were mildly underweight with moderate anaemia were 9.8% (95% CI 0%, 32%) (males) and 1.4% (95% CI 0%, 24%) (females). More females were mildly underweight 2.8% (95% CI 0%, 25%) with normal Hb than their male counterparts (1.4%; 95% CI 0%, 24%).

Prevalence of moderately underweight children with moderate anaemia after 60 days of iron supplementation was reduced from 12.7% (95% CI 0%, 34%) to 8.4% (95% CI 0%, 31%) for males and from 15.5% (95% CI 0%, 37%) to 5.6% (95% CI 0%, 28%) for females. Reduction in the proportion of children who were moderately underweight was concurrent with increase in Hb status resulting from Fe-supplementation. Iron supplementation therefore improved the nutrition status of the children though insignificantly ($p>0.05$). This was due to increase in appetite as many mothers/care providers reported an increase in appetite and thus increased food consumption after starting the use of the iron supplements. These findings correlates with the studies by Dijkhuizen *et al.* (2001) who found that, iron supplementation after six months was not able to improve growth performance of the studied children. Also studies by Friel *et al.* (2003) and Almeida *et al.* (2004) found no significant changes on nutrition status are observed after iron supplementation to anaemic children.

Results showed that, for those children supplemented with 10 mg of iron lactate there were no children with severe anaemia among all age groups at the end of the study period (Table 17). For the age of 6 months, prevalence of moderate stunting with normal Hb level was 1.4% (95% CI 0%, 24%), which was similar for both males and females. There were no cases of moderate stunting with moderate anaemia among males whereas for female the prevalence of moderate stunting was 1.4% (95% CI 0%, 24%). For children aged 7-12 months, prevalence of moderate stunting with moderate anaemia was reduced to 5.7% (95% CI 0%, 28%) from 15.7% (95% CI 0%, 37%) (for males) and to 2.8% (95% CI 0%, 25%) from 11.4% (95% CI 0%, 33%) (for females).

The proportion of moderately stunted children with normal Hb concentrations at the end of the study was 11.4% (95% CI 0%, 33%) (males) and 4.3% (95% CI 0%, 27%) (females). For age group 13-36 months, results showed that, prevalence of normal height for age with normal Hb concentrations was 4.3% (95% CI 0%, 27%) (for males) while for females was 1.4% (95% CI 0%, 24%).

Table 17: Variation of HAZ-scores and Hb status with age of the children[1]

HAZ-scores[3]/ Age	Hb status[2]							
	Males				Females			
	Severe	Moderate	Normal	Total	Severe	Moderate	Normal	Total
Children receiving 10mg (N=70)								
6 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	0(0)	1(1.4)	1(1.4)	0(0)	1(1.4)	1(1.4)	2(2.8)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7 - 12 mo								
Normal	0(0)	0(0)	1(1.4)	1(1.4)	0(0)	0(0)	3(4.3)	3(4.3)
Mild	0(0)	2(2.8)	2(2.8)	4(5.7)	0(0)	1(1.4)	3(4.3)	4(5.7)
Moderate	0(0)	4(5.7)	8(11.4)	12(17.1)	0(0)	2(2.8)	3(4.3)	5(7.1)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
13 - 36 mo								
Normal	0(0)	1(1.4)	3(4.3)	4(5.7)	0(0)	2(2.8)	1(1.4)	3(4.3)
Mild	0(0)	4(5.7)	3(4.3)	7(10.0)	0(0)	2(2.8)	4(5.7)	6(8.6)
Moderate	0(0)	3(4.3)	5(7.1)	8(11.4)	0(0)	6(8.6)	4(5.7)	10(14.3)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Children receiving 5mg (2.5 mg Fe Na -EDTA and 2.5 mg Fe lactate) (N=71)								
6 mo								
Normal	0(0)	1(1.4)	0(0)	1(1.4)	0(0)	0(0)	1(1.4)	1(1.4)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7 - 12 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	0(0)	1(1.4)
Mild	0(0)	1(1.4)	3(4.2)	4(5.6)	0(0)	0(0)	1(1.4)	1(1.4)
Moderate	0(0)	9(12.6)	4(5.6)	13(18.3)	0(0)	2(2.8)	6(8.4)	8(11.3)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
13 - 36mo								
Normal	0(0)	4(5.6)	0(0)	4(5.6)	0(0)	3(4.2)	0(0)	3(4.2)
Mild	0(0)	4(5.6)	2(2.8)	6(8.4)	0(0)	1(1.4)	4(5.6)	5(7.0)
Moderate	0(0)	5(7.0)	8(11.3)	13(18.3)	0(0)	2(2.8)	9(12.6)	11(15.5)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)

[1]Values are presented as number (percent)

[2]Normal Hb > 11.0 g/dl, Moderate anaemia, Hb 7-10.9g/dl and severe anaemia, Hb < 7g/dl

[3]Normal weight for height= >-1.0SD, Mild wasting=-1SD- -1.9, Moderate wasting=-2SD- -2.9SD and severe wasting= < -3.0SD

The proportion of mild stunting with moderate anaemia was reduced to 5.7% (95% CI 0%, 28%) from 11.4% (95% CI 0%, 33%) for males whereas for females the proportion was reduced to 2.8% (95% CI 0%, 25%) from 7.1% (95% CI 0%, 30%) at the end of the study period. Mild stunting with normal Hb was 4.3% (95% CI 0%, 27%) (males) and for females the prevalence was 5.7% (95% CI 0%, 28%). Prevalence of moderately stunted children with moderate anaemia, in males was 4.3% (95% CI 0%, 27%), while for females the prevalence was 8.6% (95% CI 0%, 31%). Prevalence of normal blood hemoglobin concentrations with moderate stunting was 7.1% (95% CI 0%, 30%) for males and 5.7% (95% CI 0%, 28%) for females.

For children supplemented with 5 mg iron Na-EDTA, there were no children with severe anaemia for all age groups at the end of the study period. For children aged 6 months, 1.4% (95% CI 0%, 24%) of males and females had normal height for age with normal Hb concentrations. For age group 7-12 months, prevalence of mild stunting with moderate anaemia was 1.4% (95% CI 0%, 24%) for both males and females. Prevalence of mild stunting with normal Hb concentration was 4.2% (95% CI 0%, 27%) (males) and 1.4% (95% CI 0%, 24%) (females). Prevalence of moderate stunting children with moderate Hb concentrations was 12.6% (95% CI 0%, 34%) for males whereas for females the prevalence was 2.8% (95% CI 0%, 25%). Those with normal Hb concentrations with moderate stunting were 5.6% (95% CI 0%, 28%) for males and 8.4% (95% CI 0%, 31%) for females.

For children in age group 13-36 months, prevalence of mild stunting with moderate anaemia was 5.6% (95% CI 0%, 28%) for males while for females the prevalence was 1.4 % (95% CI 0%, 24%). Proportion of normal height for age with moderate Hb concentrations was 5.6% (95% CI 0%, 28%) for males and 4.2% (95% CI 0%, 27%) for females. Prevalence of moderate stunting with moderate anaemia was 7.0% (95% CI 0%, 30%) for males and 2.8% (95% CI 0%, 25%) for females after 60 days of supplementation. Results showed that, 11.3% (95% CI 0%, 33%) of males and 12.6% (95% CI 0%, 34%) of females had normal Hb concentrations with moderate stunting. The consumption of 5 mg iron Na-EDTA for 60 days improved the Hb concentrations significantly ($p<0.05$) but the height for age was improved only slightly ($p>0.05$). This could be due to the fact that, linear growth usually takes place very slowly and takes a long time for any increase in height to be noticed (King and Burgess, 1993).

Variation of the WHZ and Hb concentration with the age of the children is summarized in Table 18. For children aged 6 months, prevalence of moderate wasting with normal Hb level was 1.4% (95% CI 0%, 24%) which was similar for both males and females. There were no moderately wasted males with moderate anaemia whereas for females the prevalence of moderate wasting was 1.4% (95% CI 0%, 24%). For children aged 7-12 months, moderate wasting with moderate anaemia was reduced to 8.6% (95% CI 0%, 30%) from 22.3% (95% CI 0%, 46) (for males) and to 4.3% (95% CI 0%, 27%) from 17.1% (95% CI 0%, 38%) for females.

The proportion of moderately wasted children with normal Hb concentrations at the end of the study was 15.7% (95% CI 0%, 37%) (males) and 12.8% (95% CI 0%, 35%) (females). For age group 13-36 months, results showed that, prevalence of normal weight for height with normal Hb concentrations was 1.4% (95% CI 0%, 24%) for females while for males the proportion was zero.

Table 18: Variation of WHZ-scores and Hb status with age of the children[1]

WHZ-score[3]/Age	Hb status[2]							
	Male				Female			
	Severe	Moderate	Normal	Total	Severe	Moderate	Normal	Total
Children receiving 10 mg Fe lactate (n=70)								
6 months								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	0(0)	1(1.4)	1(1.4)	0(0)	1(1.4)	1(1.4)	2(2.8)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7 - 12 months								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	8.6(6)	11(15.7)	17(23.9)	0(0)	3(4.3)	9(12.8)	12(16.9)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
13-36 months								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(1.4)	1(1.4)
Mild	0(0)	1(1.4)	1(1.4)	2(2.8)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	7(10.0)	10(14.3)	17(23.9)	0(0)	10(14.3)	8(11.4)	18(25.3)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Children receiving 5 mg (2.5 mg Fe Na -EDTA and 2.5 mg Fe lactate) (n=71)								
6 months								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	0(0)	1(1.4)	1.4(1)	0(0)	0(0)	1(1.4)	1(1.4)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
7 - 12 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	1(1.4)	1(1.4)	2(2.8)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	9(12.7)	6(8.4)	15(21.1)	0(0)	3(4.2)	7(9.8)	10(14.1)
Severe	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
13-36 mo								
Normal	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)
Mild	0(0)	2(2.8)	0(0)	2(2.8)	0(0)	0(0)	0(0)	0(0)
Moderate	0(0)	11(15.5)	10(14.1)	21(29.6)	0(0)	6(8.4)	13(18.3)	19(26.8)
Severe	0(0)	0(0)	0(0)	0(n0)	0(0)	0(0)	0(0)	0(0)

[1]Values are presented as number (percent)
[2]Normal Hb > 11.0 g/dl, Moderate anaemia, Hb 7-10.9g/dl and Severe anaemia, Hb < 7g/dl
[3]Normal weight for height =>-1.0SD, Mild wasting=-1SD- -1.9, Moderate wasting=-2SD- -2.9SD and Severe wasting= < -3.0SD

The proportion of mildly wasted females with moderate anaemia was reduced to zero from 1.4% (95% CI 0%, 24%) whereas for males the proportion remained the same at 1.4% (95% CI 0%, 24%) at the end of the study period. Prevalence of mild wasting with normal Hb concentrations was 1.4% (95% CI 0%, 24%) (males) while for females the prevalence was zero. The prevalence of moderately wasted children with moderate anaemia, in males was 10.0% (95% CI 0%, 32%), while for females the prevalence was 14.3% (95% CI 0%, 36%). Prevalence of normal haemoglobin concentrations with moderate wasting was 14.3% (95% CI 0%, 36%) for males and 11.4% (95% CI 0%, 33%) for females.

For children supplemented with 5 mg iron Na-EDTA, there were no children with severe anaemia for all age groups at the end of the study period. For children aged 6 months there were 1.4% (95% CI 0%, 24%) males and females with moderate wasting and normal Hb concentrations. For age group 7-12 months, prevalence of mild wasting with moderate anaemia was 1.4% (95% CI 0%, 24%) (for males) while for females the prevalence was zero. Prevalence of moderate wasting with moderate Hb concentrations was 1.4% (95% CI 0%, 24%) (for males) whereas for females the prevalence was zero. Males with normal Hb concentration with moderate wasting were 8.4% (95% CI 0%, 31%) while females were 9.8% (95% CI 0%, 32%).

For children in age group 13-36 months, prevalence of mild wasting with moderate anaemia was 2.8% (95% CI 0%, 25%) (for males) while for females the prevalence was zero. Prevalence of moderate wasting with moderate anaemia was 15.5% (95% CI 0%, 37%) (males) and 8.4% (95% CI 0%, 31%) (females) after 60 days of supplementation. Results showed that, 18.3% (95% CI 0%, 39%) of females and 14.1% (95% CI 0%, 36%) of males had normal Hb concentrations with moderate wasting. The consumption of 5 mg iron Na-EDTA for 60 days improved the Hb concentrations significantly ($p<0.05$). The nutrition status of the children was also improved slightly (p>0.05) due to iron supplementation. Results of this study were similar to those from studies by Almeida *et al.* (2004) and Friel *et al.* (2003) who observed that, supplementation of children with iron did not result in significant improvement in their weight for height.

4.8.1 Association between anthropometric characteristics and Hb concentration

From this study, sex, age of the child, underweight, wasting and stunting had no significant (p>0.05) influence on Hb concentration of the children studied (Table 19). Underweight, wasting and stunting had no relationship with haemoglobin concentration. The study correlates with a study by Dijkhuizen *et al.* (2001) who reported that, iron supplementation after six months was not able to improve growth performance of the studied children. Also, a study by Friel *et al.* (2003) and Almeida *et al.* (2004) found no significant changes on nutrition status after iron supplementation.

Table 19: Regression analysis when the dependent variable is Hb concentration

Variable	Coefficient			Standard error			t-value			Significance		
	WAZ	WHZ	HAZ	WAZ	WHZ	HAZ	WAZ	WHZ	HAZ	WAZ	WHZ	HAZ
Constant	10.626	10.608	10.664	0.401	0.400	0.400	26.500	26.503	26.628	0.000	0.000	0.000
Sex	0.401	0.412	0.410	0.210	0.211	0.209	1.907	1.955	1.958	0.059	0.053	0.052
Age	-0.007	-0.009	-0.004	0.012	0.012	0.012	-0.556	-0.738	-0.315	0.579	0.462	0.753
Underweight	0.045	-	-	0.093	-	-	0.482	-	-	0.631	-	-
Wasting	-	0.046	-	-	0.092	-	-	-0.502	-	-	0.616	-
Stunting	-	-	0.094	-	-	0.077	-	-	1.219	-	-	0.225

Furthermore, Ziegler *et al.* (2009) showed that, although there is a high prevalence of iron deficiency among underfive children, correcting these deficiencies is not sufficient to allow optimal growth to children. There must be additional underlying factors in the diet, or circumstances to be corrected to these children that effectively impair growth. Sex was not a risk factor, however; it is a risk factor after the initiation of the puberty stage. Females have increased need for iron due to menstruation compared to the male adolescents (Stang and Story, 2005). In this study, no significant difference was observed in the weights, heights and Hb concentration between males and females after supplementation with high dose (10 mg Fe-lactate) or low dose of iron (5 mg Na-Fe-EDTA).

Age of the child was found not to have significant association with haemoglobin concentration. Results of this study were slightly different from findings by Almeida *et al.* (2004) who observed that, Hb and age of preschool children were significantly correlated and suggested that children are able to consume a more varied diet with increasing age, thus improving their iron status.

4.9 Dietary Pattern of the Studied Children

4.9.1 Food types

Common foods that were consumed by the children in Turiani included animal products, legumes and pulses, nuts and oilseeds, cereals, roots and tubers, fruits and vegetables. Animal products consumed included beef, chicken, duck, lamb meat, goat meat, game meats, fish, milk, sardines and eggs. Legumes and pulses consumed were beans, green peas, pigeon peas, pumpkin seeds, soybeans, cowpeas and lentils. Also, the children in Turiani consumed common cereals such as maize, sorghum, finger millet, rice, wheat and bulrush millet. Roots and stem tubers that were commonly eaten were cassava, sweet potatoes, irish potatoes and yams.

The common fat/ oilseeds eaten were peanuts and oyster nuts. Furthermore, they consumed common fruits such as tomatoes, cucumbers, avocadoes, ripe bananas, watermelons, monkey mangoes, mangoes, tangerines, oranges, jackfruits and pineapples. Common vegetables eaten were carrots, brinjal, Chinese spinach, amaranthus, egg plant, spider plant, corchorus, moringa leaves, sesbania, jute mallow, cabbage, milk weed, spiderflower, african night-shade, sweetpotato leaves, cowpea leaves, pumpkin leaves, ladies fingers and cassava leaves.

4.9.2 Frequency of consumption of various foods

Data in Appendix 7 a, b, c, d and e revealed that, sardines and fish were the animal products that were most frequently consumed by the studied children. On average, sardines were consumed five (5) times per month whereas fish were consumed four (4) times per month. Milk was consumed on average of three (3) times per month. Other types of animal products such as beef, goat, chicken, duck and lamb meats were consumed once or less times per month. This suggested that, there was generally low intake of animal foods by the children involved in the study. This observation correlate the report by TNP (2008) which reported that, consumption of animal foods, which are rich in essential micronutrients, such as vitamin A, iron and calcium, is low among children in central and southern zone of Tanzania.

Finger millet and maize were the most common cereals consumed by the subjects. On average, maize stiff porridge was consumed about 21 times in a month. Finger millet was consumed on average of four times per month. Beans and cow-peas were the most common legumes and pulses consumed by the children. On average, beans were consumed six times per month while cowpeas were consumed three times per month.

On the other hand, fruits and vegetables were the least consumed with the exception of tomatoes. Tomatoes were consumed frequently by the studied children at an average of 13 times per month. This could likely be due to the fact that, tomatoes are used as condiment to season the relishes used with the staple foods. Common type of beverage taken by the children was black tea. Each child was consuming tea an average of nine times per month. This indicated that, most of the children in Turiani used black tea during breakfast. It was also used as a common drink after meals. According to Wardlaw (2003), tea contains polyphenols (tannins) which interfere with iron absorption. According to WHO (2001), foods which contain polyphenols should be taken two hours after a meal to allow absorption of iron from the food.

CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

Since intake of both 10 mg iron lactate and 5 mg containing 2.5 mg Na-EDTA and 2.5 mg iron lactate resulted in significant increase in haemoglobin concentrations of the anaemic children, it is concluded that, the low dose iron-with EDTA is just as effective in controlling iron deficiency as the high dose of 10 mg iron lactate. The low dose iron Na-EDTA may therefore be administered in public intervention programs to address IDA. This in turn would reduce the risks for malaria complications that have been reported when high dose of iron was used to control IDA in malaria endemic areas.

5.2 Recommendations

It is recommended that

i. Community should be educated on the importance of feeding their children well balanced diets that contain all essential nutrients needed for growth and development. Since fruits and vegetables are good sources of micronutrients, families should be encouraged to feed their children fruits and vegetables in generous amount.

ii. Since iron fortified foods for children are sold at high prices, which many families may not afford, it is recommended that, the government and other stakeholders should promote fortification of locally produced foods for childrens at household level to prevent micronutrients deficiencies including IDA. The priority should be given to those micronutrients of public health significance such as iron and vitamin A.

iii. Malaria control program should be reinforced to address the problem of iron deficiency anaemia.

iv. Control of worms (helminthes) is essential to control blood loss that leads to IDA.

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APPENDICES

Appendix 1: Explanation given to mothers/caregivers concerning the study

1. Purpose of the study: This research is going to determine the efficacy of providing 5 mg of iron per day (2.5 mg as ferric EDTA) in improving haemoglobin concentration (g/dl) of children aged 6-36 months in malaria endemic area. This study is done in collaboration with Sokoine University of Agriculture department of food science and Technology.

2. Study procedure: If you agree your child to participate in this study, please respond to my questionnaire and provide the required information. I will ask your permission to take your child's body measurements including weight and the blood will be drawn to measure the hemoglobin level at the beginning and the presence/absence of malaria parasites (Plasimodium falciparum). Then follow up will be done at the midith of the study to measure the increase in haemoglobin concetration. After two month (at the end of the study) weight, length and haemoglobin concetration will be measured to detect changes in haeomoglobin concentrations (g/dl).

3. Risk and discomfort: Hardly, no any risk is expected but the child will fill pain during finger pricking to take blood capillary sample. If the child is happen to be allergic to Fe -Na -EDTA medical attention will be provided and can withdraw from the study.

4. Benefits: It is expected that there will be direct benefit to a child where by iron status will be improved and enhance motor development.

5. Compensation: You will not be given any allowances for your child participating in the study.

6. Confidentiality: confidentiality will be maintained.

Appendix 2: Statement of consent

I have read the above information or it has been read to me and understands. I have had the opportunity to discuss this research study with researcher, and I have had my questions answered by her in a language I understand. I let my child to take part in this study of my own free will.

Agreement to take part in the study:

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Name of caregiver of child	**Signature/thumb print**	**Date**
------------------------------	------------------------------	------------------------------
Name of researcher	**Signature**	**Date**
------------------------------	------------------------------	------------------------------

Appendix 3: Questionnaire used during screening

SECTION A: RAPPORT

1. Child name ....
2. Sex .... M/F
3. Age ....years
4. Morther's name ....
5. Farther's name........................
6. Village ....
7. Ten cell leader....................
8. Village Health Worker ....
9. Is the child sick today?.......Y/N
10. Last time the child had malaria ....
11. Was the child treated........Y/N
12. Last time the child had de-wormed ....
13. Is edema present?...............Y/N

14. Siblings under the age of five (5) in the same house hold:

Name	Age (years)	Sex
....	....	....
....	....	....
....	....	....

SECTION B: MEASUREMENTS

1. Weight (kg)
2. Height/ Length (cm)
3. Hb (g/dl)

Appendix 4: Questionnaire for baseline study

SECTION A: RAPPORT

1. Child name .... 2. Sex .... M/F 3. Date ....

4. Age ....years 5. Morther's name ....

6. Farther's name........................ 7. Village ....

8. Ten cell leader .... 9. Village Health Worker ....

10. Child ID No. .... 11. Supplement GROUP................................

SECTION B: MEASUREMENTS

1. Hb (g/dl)..

2. Malaria status ....

Appendix 5: Questionnaire for 4 (midterm) and 8 weeks (end of term) of the study

SECTION A: RAPPORT

1. Child name .... 2. Sex .... M/F 3. Date ....

4. Age ....years 5. Morther's name ....

6. Farther's name........................ 7. Village ....

8. Ten cell leader .... 9. Village Health Worker ....

10. Child ID No. .... 11. Supplement GROUP................................

SECTION B: MEASUREMENTS

1. Weight (kg)...

2. Height/ Length (cm)..............................

3. Hb (g/dl)...

4. Malaria status ....

Appendix 6: Questionnaire for dietary intake information

SECTION A: RAPPORT

1. Child name .... 2. Sex ....M/F 3. Date ....

4. Age ....years 5. Morther's name ....

6. Farther's name....................... 7. Village ....

8. Ten cell leader.................... 9. Village Health Worker ....

SECTION B: DIETARY INTAKE INFORMATION FOR PAST 24 HOURS

Choose type of food eaten by the child for the past 24 hours

Animal products	Sweet potatoes	Cabbage
Beef	Irish potatoes	Jute mallow
Chicken	Yam	Sesbania
Lamb meat	Bread nut	Moringa leaves
Goat meat	Taro	Corchorus
Game meat	Mixed maize with beans	Spider plant
Fish	Wheat	Egg plant
Duck meat	Cooked banana	Amaranthus
Milk	Maize stiff porridge	Chinese spinach
Sardines	Millet stiff porridge	Brinjal
Egg	Sorghum stiff porridge	Carrot
Legumes and pulses	Cassava stiff porridge	**Fruits**
Beans	Wheat bread	Pineapple
Green peas	Pancake	Jack-fruit
Pegion pea	Ban	Orange
Pumpkin seeds	Rice buns	Tangarine
Soy beans	**Beverages**	Mango
Cow peas	Tea	Monkey mango
Lentils	Water	Guava
Hyacinth bean	**Vegetables**	Water-melon
Nuts and oil seeds	Cassava leaves	Banana
Groundnuts	Ladies finger	Avocado
Cereals, roots and tuber foods	Pumpkin leaves	Cucumber
Maize porridge	Cow pea leaves	Tomato
Finger millet porridge	Sweet potato leaves	
Rice	African night shade	
Cassava	Milk weed	

Appendix 7: (a) Frequency of consumption of animal products per week

Food item	1st week			2nd week			3rd week			4th week			Overall for four weeks
	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	
Animal products													
Beef	110	3.0	1.0	58	1.5	0.5	63	1.9	0.6	47	1.3	0.4	1.9
Goat meat	19	0.5	0.2	57	1.5	0.5	11	0.3	0.1	21	0.6	0.2	1.0
Lamb meat	9	0.2	0.1	9	0.2	0.1	4	0.1	0.0	7	0.2	0.1	0.3
Chicken meat	73	2.0	0.7	134	3.5	1.3	28	0.8	0.3	104	2.9	1.0	3.3
Duck meat	34	0.9	0.3	53	1.4	0.5	9	0.3	0.1	52	1.4	0.5	1.4
Game meat	7	0.2	0.1	2	0.1	0.0	20	0.6	0.2	8	0.2	0.1	0.4
Milk	77	2.1	0.7	68	1.8	0.6	101	3.0	1.0	65	1.8	0.6	2.9
Fish	163	4.5	1.5	130	3.4	1.2	97	2.9	0.9	73	2.0	0.7	4.3
Sardines	183	5.0	1.7	158	4.1	1.5	157	4.7	1.5	102	2.8	1.0	5.7
Egg	3	0.1	0.0	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	0.0

Frequency[1]: Average consumption per individual per week

(b) Frequency of consumption of cereals, roots and tubers per week

Food item	1st week			2nd week			3rd week			4th week			Overall for four weeks
	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	
Cereals, roots and stem tubers													
Cassava	25	0.7	0.2	62	1.6	0.6	57	1.7	0.5	49	1.4	0.5	1.8
Sweet-potatoes	28	0.8	0.3	59	1.5	0.6	53	1.6	0.5	50	1.4	0.5	1.9
Irish potatoes	32	0.9	0.3	26	0.7	0.2	22	0.7	0.2	9	0.3	0.1	0.8
Yams	5	0.1	0.0	6	0.2	0.1	1	0.0	0.0	0	0.0	0.0	0.1
Bread nut	4	0.1	0.0	3	0.1	0.0	0	0.0	0.0	2	0.1	0.0	0.0
Taro	1	0.0	0.0	19	0.5	0.2	18	0.5	0.2	16	0.4	0.2	0.6
Green bananas	33	0.9	0.3	40	1.0	0.4	31	0.9	0.3	58	1.6	0.5	1.5
Maize	549	15.0	5.2	501	13.1	4.7	623	18.7	5.9	538	15.0	5.1	20.9
Sorghum	17	0.5	0.2	1	0.0	0.0	1	0.0	0.0	3	0.1	0.0	0.2
Pearl millet	2	0.1	0.0	1	0.0	0.0	0	0.0	0.0	2	0.1	0.0	0.0
Cassava	10	0.3	0.1	12	0.3	0.1	11	0.3	0.1	3	0.1	0.0	0.3
Finger millet	125	3.4	1.2	111	2.9	1.0	94	2.8	0.9	98	2.7	0.9	4.0

Frequency[1]: Average consumption per individual per week

(c) Frequency of consumption of legumes and pulses per week

	1st week			2nd week			3rd week			4th week			Overall for four weeks
	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	
Legumes and pulses													
Beans	177	4.8	1.7	138	3.6	1.3	201	6.0	1.9	145	4.0	1.4	6.3
Lentils	4	0.1	0.0	9	0.2	0.1	0	0.0	0.0	3	0.1	0.0	0.1
Cow peas	5	0.1	0.0	14	0.4	0.1	11	0.3	0.1	17	0.5	0.2	0.4
Pigeon pea	144	3.9	1.4	110	2.9	1.0	141	4.2	1.3	125	3.5	1.2	4.9
Green peas	10	0.3	0.1	8	0.2	0.1	2	0.1	0.0	4	0.1	0.0	0.2
Pumpkin seeds	11	0.3	0.1	13	0.3	0.1	2	0.1	0.0	3	0.1	0.0	0.2
Soy beans	0	0.0	0.0	1	0.0	0.0	0	0.0	0.0	3	0.1	0.0	0.0

Frequency[1]: Average consumption per individual per week

(d) Frequency of consumption of vegetables per week

Food item	1st week			2nd week			3rd week			4th week			Overall for four weeks
	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	
Vegetables													
Cassava leaves	24	0.7	0.2	7	0.2	0.1	16	0.5	0.2	12	0.3	0.1	0.6
Ladies finger	4	0.1	0.0	10	0.3	0.1	10	0.3	0.1	16	0.4	0.2	0.4
Pumpkin leaves	38	1.0	0.4	48	1.3	0.5	39	1.2	0.4	82	2.3	0.8	2.1
Cowpea leaves	84	2.3	0.8	141	3.7	1.3	26	0.8	0.2	86	2.4	0.8	3.1
Potato leaves	109	3.0	1.0	87	2.3	0.8	109	3.3	1.0	84	2.3	0.8	3.6
African night-shade	19	0.5	0.2	7	0.2	0.1	8	0.2	0.1	9	0.3	0.1	0.5
Milk weed	21	0.6	0.2	18	0.5	0.2	10	0.3	0.1	14	0.4	0.1	0.6
Chinese spinach	20	0.5	0.2	28	0.7	0.3	37	1.1	0.3	36	1.0	0.3	1.1
Cabbage	10	0.3	0.1	13	0.3	0.1	19	0.6	0.2	19	0.5	0.2	0.6
Jute mallow	1	0.0	0.0	1	0.0	0.0	6	0.2	0.1	2	0.1	0.0	0.1
Sesbania	16	0.4	0.2	22	0.6	0.2	27	0.8	0.3	40	1.1	0.4	1.1
Moringa leaves	2	0.1	0.0	3	0.1	0.0	4	0.1	0.0	1	0.0	0.0	0.0
Corchorus	22	0.6	0.2	15	0.4	0.1	27	0.8	0.3	23	0.6	0.2	0.8
Spider plant	2	0.1	0.0	3	0.1	0.0	1	0.0	0.0	2	0.1	0.0	0.0
Egg plant	22	0.6	0.2	24	0.6	0.2	35	1.0	0.3	36	1.0	0.3	1.0
Carrot	11	0.3	0.1	4	0.1	0.0	0	0.0	0.0	1	0.0	0.0	0.1
Amaranthus	101	2.8	1.0	168	4.4	1.6	99	3.0	0.9	139	3.9	1.3	4.8
Brinjal	73	2.0	0.7	89	2.3	0.8	11	0.3	0.1	53	1.5	0.5	2.1

Frequency[1]: Average consumption per individual per week

(e) Frequency of consumption of fruits and beverages per week

Food item	1st week			2nd week			3rd week			4th week			Overall for four weeks
	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	n	%	Frequency[1]	
Fruits													
Pineapples	22	0.6	0.2	37	1.0	0.3	9	0.3	0.1	37	1.0	0.3	0.9
Avocardos	17	0.5	0.2	11	0.3	0.1	2	0.1	0.0	16	0.4	0.2	0.5
Jack-fruits	3	0.1	0.0	4	0.1	0.0	3	0.1	0.0	1	0.0	0.0	0.0
Oranges	88	2.4	0.8	105	2.7	1.0	88	2.6	0.8	62	1.7	0.6	3.2
Tangarines	28	0.8	0.3	34	0.9	0.3	18	0.5	0.2	24	0.7	0.2	1.0
Mangoes	19	0.5	0.2	22	0.6	0.2	1	0.0	0.0	5	0.1	0.0	0.4
Monkey mangoes	7	0.2	0.1	28	0.7	0.3	4	0.1	0.0	15	0.4	0.1	0.5
Water melons	1	0.0	0.0	5	0.1	0.0	0	0.0	0.0	1	0.0	0.0	0.0
Ripe bananas	24	0.7	0.2	44	1.2	0.4	28	0.8	0.3	39	1.1	0.4	1.3
Cucumbers	0	0.0	0.0	1	0.0	0.0	1	0.0	0.0	0	0.0	0.0	0.0
Guava	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	0.0
Tomatoes	243	6.6	2.3	393	10.3	3.7	331	9.9	3.1	416	11.6	3.9	13.0
Beverages													
Black tea	141	3.9	1.3	354	9.3	3.3	193	5.8	1.8	330	9.2	3.1	9.5

Frequency[1]: Average consumption per individual per week