Childhood undernutrition and the potential of amaranth for reducing anaemia in southern Ethiopia

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Thesis for the degree of Philosophiae Doctor (PhD)
Hawassa University, Ethiopia and
University of Bergen, Norway
2022

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Date of defense: 10.05.2022

Year: 2022

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Print: Skipnes Kommunikasjon / University of Bergen

Dedication

This thesis dedicated to my dear husband Damte Data; daughters (Dibhora, Shalom and Biruktawit) and sons (Dawit and Yohannis). This work is also dedicated to my parents, Zebdewos and Emebet.

Scientific environment

This PhD is a joint PhD through the collaboration of the School of Public Health, College of Medicine and Health Sciences, Hawassa University, Hawassa, Ethiopia, and the Centre for International Health (CIH), Department of Global Health and Primary Care, Faculty of Medicine, University of Bergen (UiB), Bergen, Norway. The PhD was funded by the Norwegian Programme for Capacity Development in Higher Education and Research for Development (NORHED) through the South Ethiopia Network of Universities in Public Health (SENUPH) project. The NORHED-SENUPH project aimed to improve women's participation in postgraduate training, and I was given this opportunity. The training component of this PhD was carried out in both Universities and the research was conducted in Ethiopia. The principal supervisor was Professor Ingunn Marie S. Engebretsen at CIH, together with Professor Bernt Lindtjørn, also at CIH, and Associate Professor Eskindir Loha at the School of Public Health, Hawassa University Ethiopia as co-supervisors.

Funding: NORHED-SENUPH project, ETH-13/0025
Registration: Pan African Clinical Trials Registry number: PACTR201705002283263.
Ethical clearance: The Institutional Review Board of Hawassa University (IRB/098/08) and the Regional Ethical Committee West of Norway (no. 2016/2034).

Acknowledgements

"It is the Lord!" John 21:7

My foremost gratitude goes to God Almighty for His mercy, graces, daily guidance and everlasting providence.

This thesis was successfully accomplished by receiving a great deal of support and assistance from many people and organizations.

I would like to express my deepest appreciation and gratitude to my main supervisor, Professor Ingunn M. S. Engebretsen. I have learned so many things under your mentorship. I am impressed by your dedication, ability to mentor, and way of analysing things. I am very grateful for your support, patience, encouragement, and understanding that provided me with the energy and motivation to successfully complete this work. Your insightful feedback pushed me to sharpen my thinking and bring my work to a higher level. Your critical review and encouragement were taken as a lesson for my career. Thank you very much for all the invaluable support!

I am deeply indebted to my co-supervisor, Professor Bernt Lindtjorn. Your excellent guidance and analytical skills helped me in the course of the research, as well as repeated seminar discussions even during the corona pandemic through ZOOM meetings. I have greatly benefited from your dynamic support and input to each discussion and communication step; starting from the inception of the research and especially while we were working on rewriting paper one. I appreciate your kind and meaningful input to encourage Ethiopian women to become highly qualified researchers. Thank you very much!

I owe the deepest gratitude to my co-supervisor, Dr. Eskindir Loha for your invaluable support, encouragement and friendly guidance in the application of statistical tools. You were humble and eager to solve problems during our stay. You have been helpful in giving me adequate information, especially on R software. Thank you very much!

I am grateful to my beloved husband, Damte Data, for his continued and unfailing love, support, sharing of burdens and encouragement throughout this study. I love you so much! I also want to thank my kids, Debora, Dawit, Youhans, Shalom, and Biruktawit for their patience and understanding during my work. I love you, my little angels. God bless you all.

My sincere thanks go to the Norwegian Programme for Capacity Development in Higher Education and Research for Development (NORHED)/South Ethiopia Network of Universities in Public Health (SENUPH) for funding this study. I would like to sincerely thank the staff at the Centre for International Health, University of Bergen for their kindness and guidance on administrative facilitation and support.

I am thankful to Hawassa University College of Medicine and Health science, SNNPR regional laboratory staff, Tulla sub-city administrators, Cheffe Cotijabessa health centre, and health post workers for their administrative support. I also would like to thank all data collectors, child caregivers, drivers, bread distributors, bread makers, and supervisors.

My deepest gratitude goes to my mother, Emebet Zeleke, and my father, Zebdewos Orsango, for nursing me with affection and love, for their dedicated partnership in the success of my life, and for the continual encouragement and input they provided for this work. Emaye you are my model. You give me all the love and show me the strength to win life's challenges. (እማዬ አንቺን ለመግለጽ ቃላት ያጥረኛል ለኔ በአለም ላይ ምርጡዋ እናት ነሽ፡፡ ጥንካሬን፣ተስፋ አለመቁረጥን፣ ብርታትን፣ሰውን መውደድን ከሕጻንነቴ ጀምሮ አስተምረሽኛል:: ወድሻለው !!). Dad, when I was a child, you told me that your wish was to see me as a doctor and working in a teaching hospital. This strengthened me to try all opportunities to continue my education. May God bless the rest of your life with peace, health, and a long life.

I extend my gratitude to my beloved sisters and brothers, for your love and encouragement. My special thanks go to Kassaye, Abebayehu, Mimi (Debiritu Data) and Abinet for your incredible support during data collection. I thank my brother Wondoson and his wife Birukti for your hospitality, love, and encouragement; particularly when I came to Addis Ababa frequently for the laboratory analysis process. My brother Ephrem and his wife Lili provided me with the latest computer, which helped me to write this thesis. God bless you all!

It gives me great pleasure to thank my brothers and sisters in Christ of Hawassa Misrak Atibia Hiywet Birhan Church; Pastor Solomon and his wife Evangelist Fikireyesus, Wednesday prayer group, Agape Betlebet prayer group, Peniel Quire, Amistu Beteseb prayer group, and Zebdewos family prayer group for their continued prayers of support and encouragement during my study time. God bless you all!

I would like to express my sincere gratitude to my fellow PhD students for sharing their knowledge and skills, mainly Dr Tarekegn, Mrs Sewhareg, Mrs Samirawit and Mr Mihiretu, for their positive response and encouragement while discussing subjects related to some of the articles in this thesis. God bless you all!

My special thanks go to my Bergen Christian friends, especially Dr Yaliso Yaya and his family, and Sr Tsion for their prayers, hospitality, and kindness while I was in Bergen, especially during my first visit to Bergen, when I was sick their support was unforgettable.

Finally, I want to mention how grateful I am to the study participants that are at the core of this thesis.

Summary

Child malnutrition remains a major public health concern in Ethiopia. Anaemia and stunting are the two most prevalent nutritional problems among children and often co-exist. Iron-deficiency anaemia (IDA) is a common type of nutritional anaemia. These problems are mainly due to inadequate intake of essential micro and macronutrients from the diet, since children mainly consume monotonous cereal-based food. Thus, the burden of malnutrition in Ethiopia demands a deeper exploration of underutilized crops which are rich in nutrients and have the potential to reduce food and nutrition insecurity. The amaranth plant grows widely in Ethiopia and can improve dietary intake, but its nutritional values have so far not been recognized by consumers in the communities and health professionals.

Objective: The overall objective of this thesis was to investigate the efficacy of consuming bread containing processed amaranth on the treatment of anaemia and IDA among children 2-5 years of age in southern Ethiopia. Also, it sought to establish the prevalence of iron deficiency anaemia and associated risk factors, focusing on iron-rich food consumption among children aged 2-5 years in southern Ethiopia, and to assess the prevalence of co-morbid anaemia and stunting (CAS), and factors associated with CAS among children aged 2-5 years, in southern Ethiopia.

Methods

The nutritional situations of 340 randomly selected children aged 2-5 years were assessed to establish baseline data. Mothers were interviewed using a structured questionnaire to obtain child and household information. Anthropometric measurements and blood samples for haemoglobin were taken from 331 children. From children with anaemia 107/331, serum ferritin and C-reactive protein (CRP) were analysed to asses IDA. A cluster randomized controlled trial was then conducted, including 100/107 anaemic children (N=100), to determine the efficacy of bread containing processed amaranth compared to maize bread, followed for 6 months. Children in the amaranth arm (N=50)

received 150 g bread daily containing 70% amaranth and 30% chickpea. Amaranth grain processing included soaking, germinating, and fermenting, to decrease phytate concentration. Children in the maize arm (N=50) received 150 g bread daily, containing processed maize, roasted and fermented to give a similar colour and structure to amaranth bread. Dietary practice and morbidity patterns were monitored on a monthly basis while haemoglobin, ferritin, and CRP were measured at baseline and at the end of the interventions.

Result

Baseline results indicated that 107/331 **(**32.3 %) children were anaemic, and of those children, 27/107 (25%) had IDA, and 23/107 (20%) of children with anaemia also had elevated CRP, a sign of inflammation. Furthermore, from the baseline, 125/331 (37.8%) of the children were stunted and 59/331 (17.8%) had comorbid anaemia and stunting. Only 50/331 (15%) of children consumed iron-rich foods in the preceding 24 hours. Children from mothers with increased educational level adjusted odds ratio (AOR 1.1; 95%CI: 1.0-1.2) and households with increased dietary diversity (AOR: 1.4; 95%CI: 1.2-1.6) consumed more iron-rich foods. IDA decreased as the height for age z-score increased (AOR: 0.7; 95% CI: 0.5-0.9). CAS increased with child age (AOR: 1.0; 95%CI: 1.0-1.1) and no iron supplementation during the last pregnancy (AOR: 2.9; 95%CI: 1.3-6.2). CAS decreased in children from food secure households (AOR: 0.3; 95%CI: 0.1-0.9).

The last follow-up measurement of the trial showed that anaemia prevalence was significantly lower in the amaranth group (32%) compared to the maize group (56%) adjusted risk ratios (aRR: 0.3; 95%CI: 0.1-0.7). The haemoglobin concentration estimate was significantly higher in the amaranth group compared with the maize group adjusted beta coefficient (aβ: 8.9g/L; 95%CI: 3.5-14.3). The risk of iron deficiency anaemia was lower in the amaranth group (aRR: 0.4; 95%CI: 0.2-0.8) in the intention to treat analysis, but this was not significant in the complete case analysis. There was no difference between the groups with regard to iron deficiency (aRR: 0.8; 95%CI: 0.5-1.1).

Conclusion

Anaemia, IDA, and stunting are public health concerns among children in the study area. Household food insecurity and iron supplementation of mothers during pregnancy were factors associated with these problems in the area. Iron-rich food consumption by children was very low and the coverage of food fortification and micronutrient supplementation for children was almost nil in the study area. Consumption of bread containing processed amaranth had favourable effects on haemoglobin concentration in anaemic children. Processed amaranth grain products may therefore have a potential to improve nutritional status in children and could be considered a cost-effective strategy. Further research and awareness-raising are needed.

List of original papers

This thesis is a synthesis of the following three original research papers, which will be referred to by their respective roman numerals I–III.

Paper I: Orsango AZ, Habtu W, Lejisa T, Loha E, Lindtjørn B, Engebretsen IMS. 2021. Iron deficiency anaemia among children aged 2-5 years in southern Ethiopia: a community based cross-sectional study. PeerJ 9:e11649 doi 10.7717/peerj.11649

Paper II: Orsango AZ, Loha E, Lindtjørn B, Engebretsen IMS. Co-morbid anaemia and stunting among children 2-5 years old in southern Ethiopia: a community-based cross-sectional study. BMJ Paediatrics Open 2021;5:e001039.doi:10.1136/ bmjpo-2021-001039

Paper III: Orsango AZ, Loha E, Lindtjørn B, Engebretsen IMS. Efficacy of processed amaranth-containing bread compared to maize bread on haemoglobin, anaemia and iron deficiency anaemia prevalence among two-to-five year-old anaemic children in Southern Ethiopia: A cluster randomized controlled trial. PLOS ONE. 2020;15(9):e0239192.

Abbreviations

AOR:	Adjusted Odds Ratio
aRR:	Adjusted Relative Risk
CAS:	Co-morbid Anaemia and Stunting
CI:	Confidence Interval
CRP:	C-Reactive Protein
EDHS:	Ethiopia Demographic and Health Survey
FAO:	Food and Agriculture Organization
FFQ:	Food Frequency Questionnaire
GEE:	Generalized Estimating Equations
HAZ:	Height for age z-score
HFIAS:	Household Food Insecurity Access Score
ICC:	Intra Class Correlation Coefficients
IDA:	Iron- Deficiency Anaemia
IFA	Iron Folic Acid
IQR:	Inter Quartile Range
LASSO:	Least Absolute Shrinkage and Selection Operator
RDA:	Recommended Dietary allowance
SD:	Standard Deviation
UNICEF:	United Nations Children's Fund
WAZ:	Weight for Age Z-score
WHO:	World Health Organization
WHZ:	Weight for Height Z-score

Operational definitions

Variables	Definition
Amaranth arm	Anaemic children in geographical clusters who consumed 150g bread containing 70 mass % amaranth grains and 30 mass % mashed chickpea daily for 6 month.
Anaemia	A child was considered anaemic if the Haemoglobin concentration was less than 11 g/dl at sea level.
Child dietary diversity	The foods were counted in nine food groups (items). Scores ranged from low (≤3 food items) and medium (4–5 food items), to high (≥6 food items).
Comorbid anaemia and stunting	A child was both stunted and anaemic.
C-reactive protein	A plasma protein. The level rises in the blood with inflammation from certain conditions and is considered high when CRP level >5mg/L
Haemoglobin	Haemoglobin is a protein in red blood cells that transports oxygen and carbon dioxide and gives blood its red colour. The molecule consists of a heme fraction and a globin fraction.
Household food diversity	Categorized into 12 food types. According to the answers they were scored from low (≤3food items) and medium (4-5 food items), to high (≥6 food items).

Household food insecurity	Determined using the nine-component Household Food Insecurity Access Scale (HFIAS) for Measurement of Food Access. Based on component scores (1=rarely, 2=sometimes, 3=often), household food insecurity was classified as secure, mild, moderate, or severe.
Iron deficiency	Ferritin <12 if healthy and ferritin <30 if inflammation was detected based on C-reactive protein level.
Iron deficiency anaemia	Anaemia concurrent with iron deficiency
Maize arm	Anaemic children in geographical clusters who consumed 150g maize bread containing 100% maize daily for 6 months.
Stunting	A child with a height-for-age Z-score less than 2SD of the WHO 2006 median Child Growth Standards.

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1. Introduction

1.1. The Global picture of anaemia

Anaemia is a condition in which the number of red blood cells or the haemoglobin concentration within them is lower than normal. The optimal haemoglobin concentrations needed to meet physiologic needs vary by age, sex, and elevation of residents, consequently impairing the capacity of optimal oxygen transportation in the blood around the body (1). Anaemia is an indicator of both poor nutrition and poor health (2).

One-fourth of the global population is affected by anaemia, and about 43% of those affected are children. One-thirds of the world's anaemic children are living in Africa (3-5). The Ethiopian Demographic and Household Survey (EDHS) revealed a 57% prevalence of anaemia among children less than five years of age(6). A worsening situation of anaemia prevalence among children less than five years of age has been reported in the last two recent EDHS reports, showing an increase from 44% in 2011 to 57% in 2016 (6, 7).

Anaemia is strongly associated with poor growth and development, limited psychomotor development, and poor long-term performance in cognitive, social, and emotional functioning in children. It also increases susceptibility to diseases which ultimately cause severe illness and death (8-10). In addition, anaemia can lead to a vicious circle in which work capacity is reduced, which in turn can lead to economic loss to an individual as well as to society (11).

1.2. Causes of anaemia

The causes of anaemia are multiple and vary across geographic areas. The prevalence and distribution of anaemia is affected by a broad range of factors, including biological, socioeconomic and contextual/ecological determinants, with many acting simultaneously

(12). On top of that, multiple causes of anaemia can coexist in an individual or in a population and contribute to the severity of anaemia (12-14).

1.2.1. Nutrient deficiency

The majority of anaemia cases in low-income countries, including Ethiopia, are caused by insufficient dietary intake of essential nutrients (15-18). Nutritional anaemia results when the intake of certain nutrients is insufficient to meet the demands for the synthesis of haemoglobin and erythrocytes (15-18). Deficiencies in micronutrients (folate, vitamin A, B12, B6, C, D, E, and riboflavin), and macronutrients (proteins) can all cause anaemia. Iron deficiency anaemia is the leading cause of childhood anaemia (6, 17, 19, 20).

1.2.2. Disease (infection and inflammation)

Diseases or infections can impair nutrient absorption and metabolism or increase nutrient losses. Hookworm is an intestinal parasite associated with blood loss. Schistosomiasis leads to blood loss, but may also contribute to anaemia through splenic sequestration of erythrocytes, and increased haemolysis (17). Malaria is a blood parasite that disturbs iron metabolism related to both increased haemolysis and decreased production of red blood cells (21). Furthermore, chronic diseases such as cancer, tuberculosis, Human Immuno deficiency Virus (HIV), chronic kidney disease, and chronic heart failure can all lead to anaemia (22, 23).

1.2.3. Age, physiological state, and genetic factors

Children less than 5 years of age have a higher nutrient demand, particularly for iron, in order to meet the need for haemoglobin and erythropoiesis that is required for expansion of blood volume, muscle and tissue development. Low-birth-weight and premature infants are at greater risk because they have smaller iron stores to begin with at birth (24).

Further, genetic haemoglobin disorders that affect haemoglobin synthesis and red blood cell production and survival, such as sickle cell trait or thalassemia, are one of the top three causes of anaemia globally (12, 25).

1.2.4. Socio-economic and environmental determinants of anaemia

Socio-economic status is tightly linked to anaemia, and affects the prevalence of anaemia through several pathways. It is associated with poor living and working conditions, including poor water quality, sanitation and hygiene, and inadequate infrastructure, which can lead to increased risk of infectious diseases. Another pathway is through food insecurity and poor dietary quality (including limited access to fortified foods and animal-source foods) (17). Further, poverty is directly linked to poorer health systems and health system access.

The education level of mothers may influence decision-making and compliance with recommended health practices, as well as caretaking practices (26). Cultural, traditional, or religious reasons can affect iron intake or bioavailability (27, 28), such as through denial of nutrient rich foods to children. Induced and natural emergencies also increased the risk of anaemia (29).

1.3. Iron deficiency and iron deficiency anaemia

Based on the level of severity, iron deficiency is defined as an abnormal iron biochemistry status, with or without the presence of anaemia. Iron is a part of many enzymes and is used in many cell functions. When our bodies do not have enough iron, many parts of the human body are affected, and especially haemoglobin production will be reduced (30).

Iron deficiency has three different stages: The first stage is iron depletion characterized by a progressive reduction of storage iron in the liver. This early stage can be reflected by a fall in serum ferritin concentration. The second stage is iron-deficient erythropoiesis, which is referred to as iron-deficiency without anaemia. In the second stage, haemoglobin may or may not decline. The third stage is iron deficiency anaemia, which can be characterized by exhaustion of iron. The main feature of the third stage is declining levels of haemoglobin (30, 31).

Iron deficiency anaemia can occur at all stages of life, but children aged under 5 years bear the largest burden of IDA globally (32). Iron deficiency in young children is mainly due to the high demand for iron at this age, frequent infections and low bioavailable iron in the complementary food and monotonous diet (33, 34). Other reasons include depletion of the iron stores, which could be low at birth and further depleted through 6 months of breastfeeding (12).

Iron deficiency anaemia results from a variety of causes. The main cause of iron deficiency in Ethiopia is inadequate iron intake and infections. Inadequate iron intake may be due to low levels of iron in the diet and low bioavailability of iron in the diet (7). Increased blood loss and damage due to hookworm, schistosomiasis and malaria are other cause of iron deficiency (35, 36).

There is a paucity of data concerning IDA at a national level for most countries. The commonly used method to determine IDA is an estimate, assuming that approximately 50% of anaemia cases are caused by IDA, which is not always the case since anaemia has multiple causal factors (5). The reported magnitude of IDA in Ethiopia has not been consistent. The Ethiopia National Micronutrient Survey (ENMS) undertaken in 2014 estimated a national prevalence of 8.6% of iron deficiency anaemia among children 6-59 months old (37). Another recent study conducted in southwest Ethiopia reported 37% IDA among school children (19).

1.4. Co-existence of stunting and anaemia

Undernutrition comprises impaired growth performance, resulting in being too short for ones age as in stunting, or too thin as in wasting, or both. Being underweight can result from impaired growth performance or thinness or both. In addition, undernutrition also comprises micronutrient deficiencies. Various forms of undernutrition conditions co-exist, and they may share risk-factors, or one condition may be a risk factor for another type of undernutrition (38-40).

Globally, 151 million (22%) children less than 5 years old are stunted. Out of these, developing countries account for 91% of stunted children, and more than one in three of the stunted children live in Sub-Saharan Africa (41). In Ethiopia, about 38 percent of children under five years of age are stunted, which is classified as a severe public nutritional problems (6, 41).

Stunting is one of the major nutritional causes of morbidity and mortality among children under-five years, and is estimated to cause one million child deaths yearly (42-44). Stunting reflects a failure to receive adequate nutrition over a long period of time. It may not be detected or addressed adequately and in time in early life, and is a risk factor for adverse functional consequences: poor cognition and educational performance, low adult wages, and lost productivity. Both anaemia and stunting share common risk factors and often coexist (45, 46).

1.5. Strategies to address anaemia

WHO broadly categorizes two strategies to address anaemia: nutrition-specific and nutrition-sensitive (12).

1.5.1. Nutrition-specific strategies

Nutrition-specific strategies focus on addressing the immediate causes of anaemia, principally poor dietary intake of haematopoietic nutrients like iron or vitamin A, as well as infant feeding practices, access to fortified foods, and supplementation (12).

1.5.1.1. Dietary diversification

Dietary diversification is a strategy that aims to enhance the availability, access, and utilization of locally available and acceptable foods with high content and bioavailability of micronutrients throughout the year. It involves changes in food production practices, food selection patterns, and traditional household methods for preparing and processing indigenous foods (12).

The key actions that are recommended by WHO to promote dietary diversification include increasing the production and consumption of iron-rich foods (red meat, poultry, and fish) and vitamin A-rich foods (green leafy vegetables, orange-fleshed fruits and vegetables, dairy products, eggs, liver and fish oils), adding fruits and vegetables that are rich in citric or ascorbic acid (e.g. citrus fruits), promoting cultural food processing (germination, fermentation and soaking), avoiding known inhibitors with a meal (tea and coffee) and consuming dairy products (milk, cheese and other foods made from milk) as a between meal snack, not at meal times (12).

Dietary diversification is considered a sustainable means to address child malnutrition by the Ethiopian government (47, 48) and is promoted by non-government organizations. Iron-rich food consumption by children is quite low in Ethiopia. The 2016 EDHS reported that only 22% of children consumed iron-rich foods during the 24 hours before the interview, and 14% of children had a minimum dietary diversity in which they had been given foods from the appropriate number of food groups (6).

1.5.1.2. Infant feeding

Infants should be exclusively breastfed for the first 6 months of life (49). After 6 months of age, infants should receive nutritionally adequate and safe complementary foods, while continuing to breastfeed for up to 2 years or beyond. Complementary food should be safe and provide an increasingly greater proportion of a child's daily nutrient needs as the child grows older. Complementary food should contain the greater proportion of iron content required per day (50, 51).

1.5.1.3. Food fortification

Food fortification is the process of deliberately increasing the content of essential micronutrients in food in order to improve the nutritional quality of the food supply and provide a public health benefit with minimal risk to health(52). Fortification has the potential to improve the nutritional status of a large portion of the population, and is effective, simple, and inexpensive once established (53).

Iron fortification at the national or regional level is recommended only if there is laboratory evidence of iron-deficiency anaemia prevalence >5% (12). Ferrous sulfate and ferrous fumarate are recommended for fortification of wheat flour (54).

1.5.1.4. Iron and folic acid supplementation

Nutritional supplements are any dietary supplement that is intended to provide nutrients that may otherwise not be consumed in sufficient quantities. In areas where anaemia prevalence is higher than 40% in pre-school children and pregnant women, daily iron supplementation is recommended. The recommended public health intervention is 30 mg of elemental iron for children 2-5 years of age for three months per year. Also recommended for pregnant women is 30–60 mg of elemental iron with 0.4 mg of folic acid (12, 55).

Although, the WHO recommends iron supplementation as a public health intervention for children aged 6 months and above, and pregnant women living in areas where anaemia is highly prevalent (55, 56), the most recent EDHS reported that only 9% of children aged 6-59 months had received iron supplements in the 7 days preceding the survey. This data may reflect treatment of anaemia, since iron supplementation for child anaemia prevention is not active in Ethiopia. Furthermore, the report indicated more than half of the women with a child born in the last 5 years (58%) did not take any iron folic acid tablets during their most recent pregnancy (6).

1.5.1.5. Social and behaviour-change communication strategies

A social and behaviour-change communication strategy should accompany nutrition-specific interventions aimed at preventing anaemia. These strategies can promote optimal infant and young child feeding practices and behaviours, including during pregnancy and lactation. It can create awareness and correct use of iron supplements and/or home fortificants, along with other practices such as hand washing with soap, prompt attention to fever in malaria settings, and measures to manage diarrhoea, particularly among younger children (12).

1.5.2. Nutrition-sensitive strategies

Nutrition-sensitive solutions to control anaemia address the underlying and basic causes of anaemia that require input from a wide range of sectors, including disease control, water, sanitation and hygiene, and reproductive health. They focus on intersectoral strategies that address root causes such as poverty, lack of education, and gender norms (12).

1.5.2.1. Malaria control

Malaria is the primary cause of anaemia globally, particularly among pregnant women, infants, and children (57, 58). Malaria control in endemic areas can reduce anaemia (59). Malaria prevention measures should include vector control with insecticide-treated nets or indoor residual spraying (IRS), chemoprevention, and diagnostic testing and treatment (60).

In Ethiopia, malaria incidence and deaths are decreasing (61), but the practice of malaria control mechanisms by households is still poor (62).

1.5.2.2. Helminth control

Micronutrient utilization can be impaired by parasitic infections. Hookworm and schistosomiasis are associated with blood (and iron) loss and iron-deficiency anaemia in pregnant women and in preschool and school-age children (63, 64).

In Ethiopia, almost half of preschool children were infected with at least one intestinal parasite(65). The high burden of poor sanitation and low coverage of safe water in rural areas could increase the risk (6). WHO recommends periodic deworming with anti-helminthic medicines for people living in areas where the baseline prevalence of any soil-transmitted infection is 20% or higher among children (66, 67). But the 2016 EDHS report indicated quite low coverage of deworming: only 13% of respondents were given deworming medication 6 months before the survey (6).

1.5.2.3. Reproductive health practices

Preventing and treating maternal anaemia decreases the risk of anaemia in children. Women’s anaemia prevention focuses on their adolescent and adult years, including delaying the age of first pregnancy, ensuring optimal access to and coverage of quality prenatal care when a woman does become pregnant, ensuring that evidence-based delivery and postnatal care practices are employed, and promoting optimal birth spacing (12).

1.6. Anaemia prevention strategies currently being implemented in Ethiopia

The national nutrition programme focuses on integrated nutrition-specific and nutrition-sensitive interventions (68-70). The success of anaemia control in Ethiopia has varied considerably across the geographic regions, and the prevalence of anaemia has not declined as per the target set of 24% in 2020 (6). The prevalence of anaemia systematically decreased from 2008 to 2015, but started to increase from 2016. If present trends are maintained, it is unlikely the desired anaemia control goals (58, 71) will be achieved.

Table 1 Anaemia prevention strategies currently being implemented in Ethiopia.

Target group	Intervention
Pregnant women	Promote iron folic acid (IFA) supplementation, sleeping under a bed net, and taking intermittent preventive treatment (IPTp) for malaria and deworming pills
Infants, young children, and mothers	Promote delayed cord clamping, sleeping under a bed net, exclusive breastfeeding, and birth spacing.
Young children	Continued breastfeeding and adequate complementary feeding (including micronutrients), preventing and treating malaria, and taking deworming pills
Adolescence	Promote IFA supplements and deworming pills, family planning, and delaying the age at first birth.

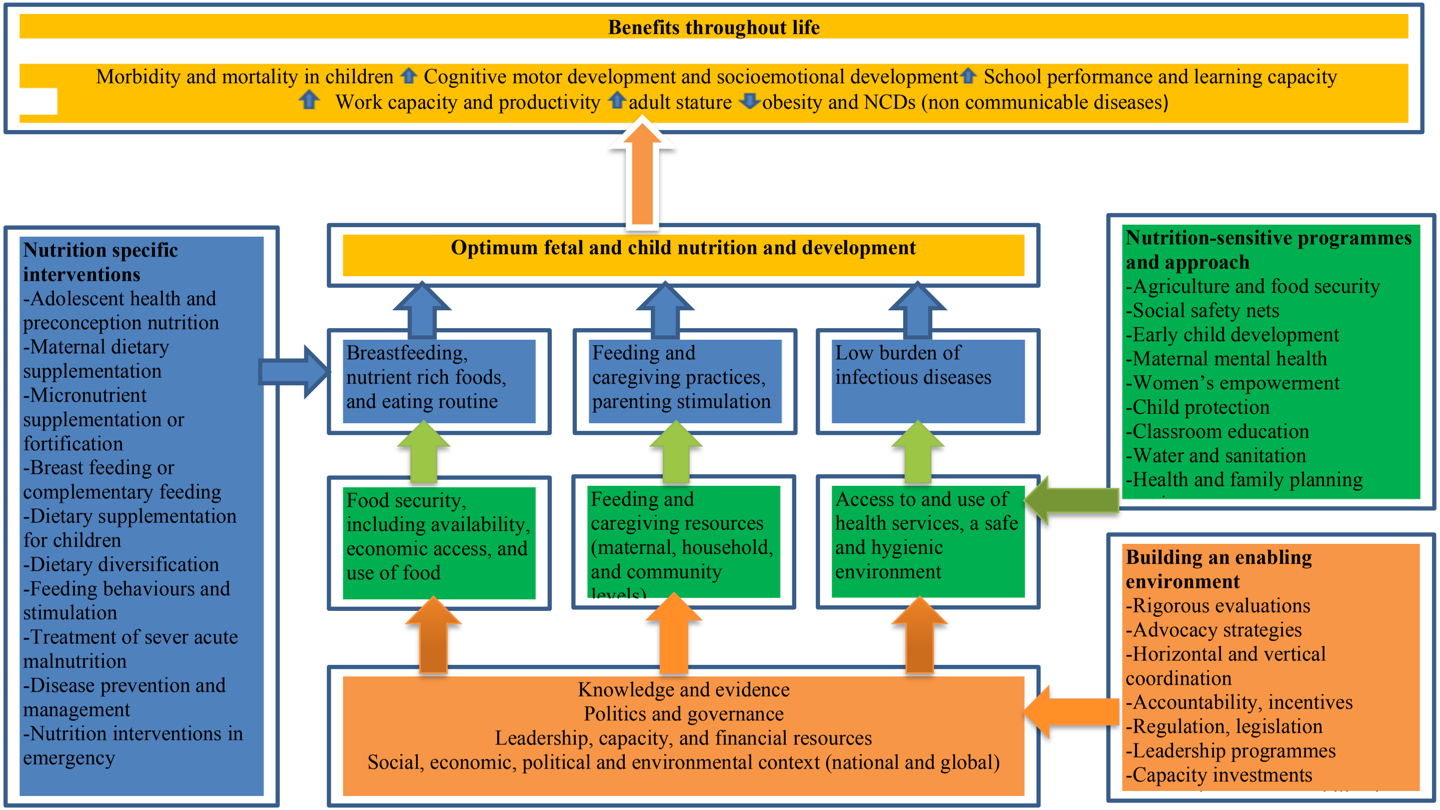

Figure 1 This framework summarizes the socioeconomic and cultural problems which require the action of different sectors to achieve optimum fetal and child nutrition and development (Adopted from Executive Summary of the Lancet Maternal and Child Nutrition Series) (72).

1.7. Amaranth

1.7.1. Origin and history of amaranth production worldwide

Amaranth originated in America and Europe and has been cultivated for more than 8000 years as a staple food in some countries. It is an annual plant with upright growth habits cultivated for both its seeds, which are used as a grain, and its leaves, which are used as vegetables (73, 74).

1.7.2. Agronomy of amaranth

Amaranth has an outstanding agronomic performance owing to its resistance to drought, hot climate, and pests. It can grow in all weather conditions, especially in the places where maize can grow (75, 76). Harvesting of leaves normally starts just 4 weeks after planting, while grains can be harvested in 60-90 days. It grows very rapidly, especially under conditions of high temperatures, bright light, and dry soil, thereby tolerating dry conditions. Grain amaranth requires low water, fertilizer, and energy relative to traditional cereals (e.g. corn, wheat, rice) (77) and can give a higher yield than maize (75). Grain amaranth has a long shelf life in storage once well dried and is not easily infested by pests (76, 78).

1.7.3. Production and utilization of amaranth in Africa and Ethiopia

Amaranth production and utilization have been gaining momentum recently in Africa, including Ethiopia, because of its nutritional value and functional properties. In Kenya, grain amaranth was officially registered as a food crop in 1991 by the Kenya Ministry of agriculture. The poverty eradication commission of Kenya is promoting amaranth as a cash crop in order to fight poverty (79-81). Recently, in Uganda, amaranth production was encouraged because of its high productivity and income generation potential (82).

In Ethiopia, amaranth is cultivated as a cash crop in the western part of the country, in the Benchi Maji area. Three varieties of amaranths differentiated by colours (white, red and black) are growing in Ethiopia. It is cultivated three times per year as single or inter-cropping with maize or sorghum (83). In the study area, amaranth grows like a weed and only the leaf parts are consumed by the communities during drought times. Recently, non-governmental organizations have been initiating and promoting the production of amaranth by some volunteer farmers.

1.7.4. Nutritional value of amaranth

Amaranth grain is appreciated for its high nutritional properties, including its high mineral content. Studies indicate that amaranth contains protein and micronutrients at unusually higher levels than staple cereals (84, 85). Amaranth grain is a good source of iron, calcium and zinc and most other micro and macronutrients. The nutrient content of amaranth seed in 100g of edible amaranth contains: crude protein 11g to 19g containing two essential amino acids – lysine and methionine – that are not frequently found in other grains, lipids 7.5%, carbohydrates 66.0% and fibre 6.9% (86). Iron around 7.6-18.0 mg/100g, zinc 3.2-3.7 mg/100g, vitamin C 4.2-4.5mg/100g (87). Unsaturated acids range from 76.2% to 77.6%, and saturated fatty acids from 22.4% to 22.8% (88).

1.7.5. Medicinal value of amaranth

In addition to high nutrient content, amaranth has been known for its medicinal value. Traditionally amaranth has been used as medicine for broken bones, internal bleeding, diarrhoea, and excessive menstruation. It has also been used in the treatment of snake bites and to treat ulcerated mouths, vaginal discharge, nose bleeding, and wounds. In India, amaranth is used as food and in traditional medicine due to its antioxidant effect (89, 90). A study evaluated the bioactivity of amaranth by determining antioxidant, anti-inflammatory, and cytotoxicity activities, and found that there is high antioxidant activity, high anti-inflammatory activity and no liver toxicity. The same study suggested that amaranth can be used in bioactive formulations against inflammatory processes and in free radical production (91). Amaranth seeds might be rich in several phytonutrients that act as powerful dietary antioxidants (92, 93). The amaranth plant is valued for the chemical composition of seeds that do not contain gluten, which is a medically accepted treatment for celiac disease (94).

1.7.6. The suitability of amaranth grain for supplementation

Amaranth can be used as seeds or flour to make products such as cookies, cakes, pancakes, bread muffins, crackers, pasta and other baked products (74). A study done in Nigeria to assess the protein quality of amaranth species revealed that amaranth is a good source of high-quality protein, with adequate amounts of all 9 essential proteins (86). A study done in Ethiopia found amaranth had potential to create nutrient-dense complementary foods. Combining amaranth and chickpea in a 70/30 ratio resulted in foods with high iron, protein and calcium levels (95).

A study from Kenya showed that amaranth grain also has low-Fe MNP (Ferro-Magnetic Nano Particles) relative to maize-based porridge. This has the potential to improve iron uptake and status in pre-school children and can decrease iron deficiency by 27%. Another study assessed the effect of consuming a cooked recipe consisting of sun-dried amaranth and cowpea leaves on the levels of beta carotene (BC), retinol, and haemoglobin in preschool children in Kenya and found that sun-dried amaranth leaves can improve serum beta carotene, retinol, and haemoglobin levels (96). Germinated amaranth grain viscosity was decreased, and was shown to be acceptable to consumers, with no adverse effects observed (97).

1.7.7. The potential of amaranth for food security

The amaranth plant has great potential to contribute to the food security of farming households, which have been hit hard by bad climatic conditions in recent years. Its production can also help to provide an all year round income for smallholder farmers. Amaranth produces a large amount of biomass in a short period of time, and therefore has the potential to contribute to a substantial increase in food production. A grain yield of up to 3000-5000 kg/ha has been reported (76, 98). The study conducted in southwestern

Nigeria indicated that under dry conditions, 1 to 6.7 tones/ha of seed and up to 70 tones/ha of green material were harvestable. This suggests that the amaranth plant is a promising future crop with the potential to contribute to a substantial increase in food production due to the large amount of biomass produced in a short period of time (99).

Picture 1 Amaranth plants growing in the area in different stages. Pictures were taken by the investigator**.**

The other peculiar feature of amaranth is the high content of nutrients in both seeds and leaves. The high-quality protein and micronutrient content, which is often lacking in the diets of resource-poor populations, makes it a good commodity for fighting malnutrition and nutritional deficiencies among the poor (76, 100).

1.7.8. Phytate content of amaranth grain

Plant-based foods have been reported to contain high levels of phytate and phenolic compounds, which impact on iron bioavailability (101). Amaranth grain contains quality amino acids, high iron and other micro-nutrients, but also has a high concentration of phytic acid that can reduce the bioavailability of iron and other essential nutrients. A study from Kenya (84) indicated that consumption of amaranth-containing porridge by school children had not yielded a positive impact on the iron status of children due to the high phytate level in the amaranth grain.

1.7.9. Homemade processing to reduce phytate levels in amaranth grain

Traditional processing technologies such as soaking and germination of grains and fermentation of flour have been widely practiced in the Ethiopian community. Soaking and germination of grain and fermentation are carried out at low cost without the use of any sophisticated or expensive equipment. Germination reduces the phytate level, thus improving the nutritional properties of the food (102). Furthermore, it is suggested that the use of low-cost processing techniques like soaking, germination, and fermentation increase nutrient bioavailability in plant-based foods (103). Studies have indicated that homemade processing can significantly reduce the phytate level of amaranth grain. Research conducted in Ethiopia and Kenya showed that homemade processing such as soaking and germination of amaranth grain could significantly reduce the phytate level and increase the bioavailability of iron and other macro and micronutrients (95, 104).

1.8. The rationale for this thesis

Child malnutrition remains a big challenge in Ethiopia. The recent period has seen a significant rise in anaemia prevalence among children under five years of age. Stunting also remains a severe public nutritional problem in Ethiopia. Both anaemia and stunting are associated with harmful effects on physical development, cognition and educational performance, increased susceptibility to infection and chronic disease, and reduced work capacity (105-107).

The majority of household diets in Ethiopia have low dietary diversity. The diets of children are dominated by starchy cereals, which are low in micronutrients. Animal source foods are rich in iron and other micronutrients (108, 109) but their intake has been declining in the study area. Thus, there is a need to explore cost-effective strategies for improving the deteriorating nutritional status of children in Ethiopia (48).

Amaranth grain would, if adopted for consumption, have the potential to enrich local diets and increase nutrient intake. However, consumption of amaranth grain is still low in Ethiopia and it is even considered to be a wild plant in southern Ethiopia. One of the adverse characteristics of amaranth grain is that it contains a high phytate level, which can inhibit the absorption of micro and macronutrients from the gut. This drawback can be decreased by processing the grain, including soaking, germinating, and fermenting, which can increase the absorption of nutrients from the gut. This was also assessed and confirmed by a previous study by the same researcher. This thesis therefore aimed to assess the effect of consuming processed amaranth grain on anaemia, iron deficiency anaemia and haemoglobin levels of children. As well as the prevalence and contributing factors associated with iron-deficiency anaemia and co-morbid anaemia and stunting in the study area.

2. Objectives of the study

2.1. Overall objectives

The overall objective of this study was to assess the impact of processed amaranth grain on the treatment of anaemia and iron deficiency anaemia among children 2-5 years of age in semi-urban southern Ethiopia.

2.2. Specific objectives

1. To establish the prevalence of iron deficiency anaemia and associated risk factors, focusing on iron-rich food consumption among children aged 2-5 years (paper I.)
2. To assess the prevalence of co-morbid anaemia and stunting and associated factors among children 2-5 years old (paper II).
3. To evaluate the efficacy of processed amaranth grain compared to the commonly consumed maize on haemoglobin concentration and anaemia prevalence among children aged 2-5 (paper III).

3. Methods and Materials

3.1. Study area

Ethiopia, one of the oldest civilizations in the world, covers an area of 1.1 million square kilometres. Based on topography and climate, Ethiopia has three broad agro-ecological zones – the hot zone (less than 1,500 metres above sea level), areas of average (mid) climatic conditions (1,500-2,300 metres above sea level) and the cool highlands (above 2,300 metres above sea level). The general annual rainfall distribution is seasonal and varies across geographic locations in the country (110). The total population of the country was estimated at over 117 million in 2021. Of that, 46% is below 15 years of age and 80% of the population lives in rural communities.

Hawassa city is located in the southern part of the country, 270 kilometres from Addis Ababa, the capital of Ethiopia. Hawassa city is the capital city of the Southern Nation Nationalities and Peoples Region (SNNPR) and the new region of Sidama, which became the tenth new region of Ethiopia. The altitude of the study area ranges from 1708 to1920 meters above sea level, and the annual average rainfall is 961mm. The city of Hawassa administration is divided into 8 sub-cities and 20 urban and 12 rural kebeles (lowest administrative unit in Ethiopia) (111). According to the 2016 national census, the total population of the city was 351,469, and it has an estimated annual population growth rate of 4%.

Chaffe Cotijabessa Kebele was the study site. It is one of the rural kebeles of Hawassa city, which can be characterized as a semi urban area. Within this project, a census was conducted in 2017 prior to sampling and data collection. The total population of the study kebele was 23,010 and the total number of households was 3,900. The total number of children less than five years old was 2,180 in those households, and 1,689 were 2-5 years old.

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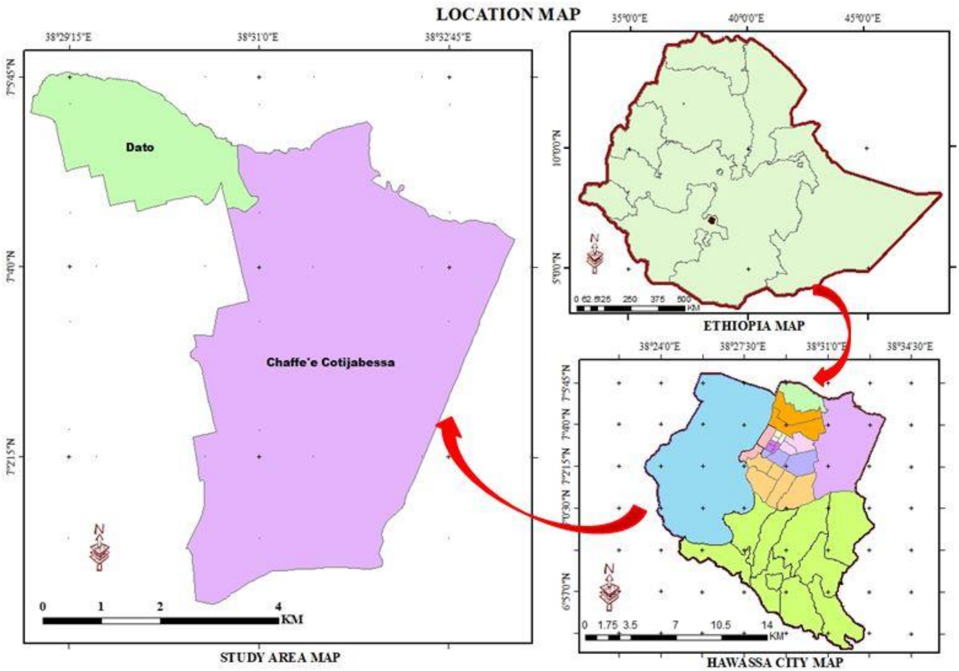

Figure 2 Geographic location of the study area – Ethiopia, Hawassa, and Chaffe Cotijabessa (Source: Hawassa city administration).

The people in the communities around the study area are low-income earners, deriving their livelihood from subsistence agriculture and daily labour. Most people have diets predominantly consisting of staple foods such as cereals and roots. The main crops cultivated in the study area include maize, haricot beans, *enset*, and Irish potatoes. Amaranth grows as a wild crop in the study area and its leaves are used as food during drought time. The grain part of amaranth is not utilized by the community members. Agricultural production has been declining in the study area due to the conversion of farmland into urban settlement. As a result, access to animal products has become a challenge for a large number of poor households. Milk, which is rich in proteins and micronutrients, is produced in some of the study communities, but largely sold to neighbouring city communities to generate income to cover household expenses. The combination of declining agricultural production and limited income generation from other alternatives has led to food insecurity for many households, which is the main underlying cause of child undernutrition in the study area.

3.1. Study design

This thesis included a community-based cross-sectional study (paper I and II) and a cluster randomized controlled trial (paper III), where the participants in the trial were drawn from the cross-sectional study.

3.2. Sample size

Cheffe Cotijabessa kebele was purposively selected, mainly for the trial component of the PhD project. The candidate had prior knowledge from a study on amaranth grain-based food preparation and the availability of amaranth grain and its uses by the population in the area. As mentioned, the census identified 1,689 children 2-5 years old prior to the cross-sectional study. The houses were given a number tag to collect socio-demographic data during the census. The sample size was calculated by assuming a 33% anaemia prevalence and 95% confidence level, +/- 5%. Thus, the total sample size was 340 children in the cross-sectional study to identify children with anaemia (paper I and II). The sample size of the trial was calculated using a mean difference of 0.7 in haemoglobin level (112), 1 standard deviation, power of 90%, and an alpha level of 0.05 with two-sided testing and an attrition rate of 12%. The total sample size of the trial was 100, with 50 children in each arm (paper III).

3.3. The study participants

Caregiver-child pairs (n=340) were selected from 1,689 children using a simple random sampling technique, in which the mother was the targeted respondent. The children included in the cross sectional studies were two-to-five year-olds who lived in the area.

Children were excluded with conditions that precluded them from participating in measurements and venipuncture. Children included in the trial were two-to-five year-olds with haemoglobin levels from 70 to <110 g/L, who lived in the area, and planned to live in the area for the next year. Children were excluded from the trial if they had chronic illnesses, such as HIV/AIDS, tuberculosis, or cancer, were taking iron supplements, had received a blood transfusion in the last six months, had repeated malaria at least three times in the last three months, or were unwilling to participate in the study.

We conducted the cross-sectional study from 15 February to 30 March 2017 (paper I and II) and identified 100 eligible children with anaemia. Then, the intervention (feeding of bread) started for children with anaemia in both allocation arms on 14 April 2017 and continued for the next six months. The identified children were sparsely distributed in the study area. We therefore grouped them into eight clusters by their geographic locations, to ease the distribution of bread and for close follow-up. The eight clusters were randomly allocated into four clusters in the maize arm and four clusters in the amaranth arm, and each arm had 50 children. The random allocation was done by an external colleague without knowledge of the cluster. We were aware of a 'violation' of the classical cluster design description implying that clusters are identified first and participants within thereafter. Yet we still chose to label it as a cluster randomized trial, as in practice, the clusters were the unit of randomization.

3.4. Trial allocation arms

The children in the experimental and control groups were labelled as 'amaranth group' and 'maize group', respectively. In both groups, 150 g bread was provided to the children under the researcher team's supervision on a daily basis for a period of six months. Bread containing 70% amaranth grain and 30% mashed chickpeas was given to the amaranth group. The maize group was supplied with the same amount of bread, but containing 100% maize. All participants in the amaranth and maize groups were treated with albendazole 400 mg single dose, irrespective of their last dosage, prior to the start of the feeding intervention and at the end of the feeding intervention (paper III).

3.5. Recipe preparation

Home level processing was applied to the amaranth grain to reduce phytate levels. Amaranth grain was soaked in water by adding 5 ml of lemon juice per 100 ml of water for 24 hours and germinated for 72 hours. After sun drying, it was roasted and milled with a local electrical mill, and then fermented bread was prepared (95). At the same time, maize grain was also roasted and fermented in order to make the recipe similar in colour and structure to the amaranth bread. The recipe was prepared based on the recommended dietary allowance (RDA); according to RDA, 150 g of bread (70% amaranth and 30% chickpea) contained 22.3 mg of iron, which can fulfil 50% of RDA assuming 15-20% iron absorption. The acceptability was found to be satisfactory in the community in a previous study (95) (paper III).

Picture 2 Soaked amaranth grain

Picture 3 Germinated amaranth grain

Picture 4 Amaranth grain flour

Picture 5 Amaranth combined bread

(Pictures were taken by the investigator)

3.6 Distribution and masking

The study participants, bread distributers, and data collectors were uninformed about arm allocation and the content of the bread. The bread distributers were different for the amaranth and maize arms, and there were eight bread-givers, one for each cluster. Eight boxes, one for each cluster, were packed and labelled with the respective caregiver's name. Each bread-giver fed the child at their home every day under their direct supervision. In case of refusals or absence, unopened bread was returned and registered by the coordinator each day (paper III).

Table 2 Data collected and activities at each phase of the trial

Data collected	**Baseline**	**Follow up visit every month for 6 consecutive visits**	**Endline**
Deworming	X		X
Food frequency	X	X	X
Illness history	X	X	X
Weight	X	X	X
Height	X	X	X
Haemoglobin	X		X
Ferritin*	X		X
CRP*	X		X
Feeding bread	Feeding every morning for 6 consequent months		

*Tested on the condition that haemoglobin was < 11 g/dl

Table 3 Data collected, inclusion and exclusion criteria and number of participants for each study design.

	Numbers	**Inclusion**	**Exclusion**	**Data collected**
Census	23,010	All households	No	Age, sex and household size
Cross-sectional study	340	Children aged 2 to 5 years and their mothers or caregivers, all of whom lived in the study area and provided informed consent.	Children with conditions that precluded them from participating in measurements and venepuncture	Mother's age, parental occupation, and education; electricity access; ownership of domestic animals and land; and iron supplementation during gestation. Household: wealth index, size, food security, monthly income; type of latrine; and water-source. Child immunization status; history of hospital admission, weight, height, child-illness (diarrhoea, cough), dietary intake, iron-rich food consumption, current breastfeeding, meal frequency, supplementation (vitamin A and iron), fortified food consumption and haemoglobin. Ferritin and CRP were tested in those children with haemoglobin < 11 g/dl
Trial	100	Children included in the study were two-to-five year-olds with haemoglobin levels from 70 to <110 g/L, who lived in the area, and planned to live in the area for the next year	Children with chronic illnesses, such as HIV/AIDS, tuberculosis, or cancer. Children who were taking iron supplements, received a blood transfusion in the last six months, had repeated malaria at least three times in the last three months, or were unwilling to participate in the study	Child age, sex, diarrhoea, cough, hospital admission, dietary diversity, height, weight, mother age, mother's and father's occupation, electricity access, household food security, household income, iron-rich food consumption, haemoglobin, ferritin, CRP

3.6. Research team training

The research team consisted of twenty members. Two supervisor's research assistants were recruited and trained on the purpose, objectives, data collection and methods of monitoring the interventions. Two people, one for maize flour and one for amaranth flour preparation, were trained how to prepare flour using home processing (soaking, germinating, drying, roasting, and milling). Two bread makers were trained in mixing ratios, duration of cooking and packing of the bread. Two bread distributers with two drivers were trained in how to check packages while collecting the bread from bread makers. Eight bread-givers were trained to serve and feed the children with the bread at the children's homes on a daily basis. Two medical laboratory technologists were trained in how to collect blood samples, haemoglobin analysis, and how to prepare and transport blood serum to the regional laboratory centre. Two laboratory experts at the national level analysed serum ferritin and CRP. Six trained data collectors were used to collect socio-economic and nutritional data using a structured questionnaire. The principal researcher guided the whole process of the research. Orientation on ethical issues was given to all research teams. A pre-test was done among 17 mother-child pairs in the nearest village outside of the study area before the actual study, also supervised by the main PhD supervisor. Then we revised the tools based on the problems that we faced during pre-testing of data collection tools. The data collection was checked daily for accuracy, consistency, and completeness by the supervisors. Anthropometry and haemoglobin instruments were calibrated as per standards.

3.7. Data collection tools

Prior to any data collection, informed written consent was provided. The research team visited the homes of the included children to collect the required data, including a structured questionnaire with a dietary recall, blood sample collections, and anthropometry. Interviews were conducted with the households. Blood collections and anthropometric measurements were performed at nearby health centres, health posts or temporary centres.

3.8.1. Structured questionnaire

The pre-tested, structured questionnaire was used to collect data. The tools were translated into local languages (Amharic and Sidamiffa) (Appendix II). It was used to collect data on socio-demographic and economic characteristics, dietary practices, morbidity patterns and nutritional status (paper I-III).

3.8.2. Dietary intake

A 7-day structured food-frequency questionnaire (FFQ) and a 24-hour dietary diversity questionnaire were used to collect information about children's diets (paper I-III). The foods were counted in nine food groups (items): cereals, roots and tubers; vitamin A-rich fruits and vegetables; other fruit; other vegetables; legumes and nuts; meat, poultry and fish; fats and oils; dairy; and eggs in which the food-frequency items were created based on Food and Agriculture Organization (FAO) guidelines. The scores ranged from low (≤3 food items), medium (4–5 food items), to high (≥6 food items) (113). In addition, dietary diversity and iron-rich food consumption frequency (red meat, organ meat, and fish) were assessed every month to control for confounding (paper III).

The household food diversity scale was categorized into 12 food groups: cereals, white tubers and roots, vegetables, fruits, meat, eggs, fish and other seafood, legumes, nuts and seeds, milk and milk products, oils and fats, sweet spices, condiments, and beverages, according to the Food and Nutrition Technical III project, and the answers were scored the same as for the food-frequency items (113).

The prevalence of household food insecurity was determined using the nine-component Household Food Insecurity Access Scale (HFIAS) (114). Based on component scores (1=rarely, 2=sometimes, 3=often), household food insecurity was classified as secure, mild, moderate, or severe (115).

3.8.3. Blood sample collection

Blood samples were collected at the start of the study from 340 children in order to determine haemoglobin levels and identify children with anaemia. Serum ferritin and CRP levels were then measured in 107 of the children with anaemia. After the 6-month intervention, 82 children were still in the trial, and haemoglobin, serum ferritin, and CRP were assessed (paper III). Standard methods for blood collection were adopted during the collection of blood samples. Using vein puncture, 3–5 ml of blood was collected in test tubes with a lithium heparin plasma separator test tube to measure haemoglobin, serum ferritin, and CRP (116). Haemoglobin was measured immediately onsite using HemoCue analysers 301(Sweden). Only samples with haemoglobin levels <11g/dl were tested further for serum ferritin and C-reactive protein (CRP) concentration. CRP was measured on Cobas 6000 (c501 module) Roche (Germany) and ferritin was measured on Cobas 6000 (e601 module) Roche (Germany).

3.8.4. Determination of haemoglobin levels

In line with WHO standards, haemoglobin concentrations were corrected for altitude (117), and anaemia was classified as normal (11.0 g/dl), mild (10.0–10.9 g/dl), moderate (7.0–9.9 g/dl), and severe (7.0 g/dl) (118).

3.8.5. Determination of serum ferritin level

For ferritin determination, the blood samples in the test tubes were immediately wrapped in aluminium foil, continuously shielded from light, and stored for 30 minutes at 4 °C until centrifugation. After centrifuging, the serum was kept in screw-capped test tubes labelled with the participant's identity. The serum was then stored at 20 °C until it was transported with a portable cold chain courier to the Ethiopian Public Health Institute in Addis Ababa for ferritin and CRP analysis.

Ferritin was adjusted for inflammation using a higher ferritin-cut-off adjustment approach. The inflammatory state of each individual was classified as "healthy" if CRP<5 mg/L and the low cut-off ferritin was <12 µg/L, and "infection" if CRP >5 mg/L and the low cut-off ferritin was <30 µg/L. Iron deficiency is defined as a ferritin concentration <12 µg/L if healthy and <30 µg/L if concurrent with infection (30, 119). Iron deficiency anaemia was defined as anaemia with concurrent iron deficiency (119, 120). Ferritin and CRP were assessed only for those children with anaemia (paper I and III).

Different ferritin correction methods recommended by WHO were used (121). The different methods were: The exclusion approach excluded individuals with elevated CRP concentrations >5 mg/L, and calculated the prevalence of low ferritin in the remaining individuals, considering the ferritin concentration cut-off <12 µg/L. The higher ferritin cut-off adjustment approach uses a higher ferritin concentration cut-off <30 µg/L. For this thesis, we used a cut-off for the subset of individuals with elevated CRP (121). Internal correction factor: The correction factor (CF) was calculated as the ratio of the geometric mean of the reference group (non-elevated CRP) to that of the respective inflammation group (elevated CRP). We found a CF value equal to 0.85 (122, 123). Then, to get the adjusted ferritin, the subgroup with CRP>5mg had their ferritin multiplied by 0.85 (30, 119). Subsequently, low ferritin was defined as <12 µg/L for both groups of elevated CRP or not elevated CRP. Finally, we used the higher ferritin cut-off adjustment approach because we had only one biomarker (CRP) (121). Iron deficiency anaemia was defined as children having a combination of a haemoglobin level of less than 11g/dl and an adjusted low ferritin concentration (<12 µg/L) (119, 120).

3.8.6. Anthropometry

Anthropometric measurements, namely weight and height, were taken to assess the child's nutritional status. Height was taken using a Seca213 height board (Seca 213, Seca GmbH, Hamburg, Germany) with a sliding headpiece, when the child was standing straight. It was ensured that the head, buttocks and heels were touching the board. Measurements were taken to the nearest 0.1 cm. Weight was taken using a calibrated Seca 874 electronic flat scale (Seca 874, Seca GmbH, Hamburg, Germany) with the child barefoot and wearing light clothing. The scale was calibrated to zero before weighing a child and recalibrated after every measurement with a known weight. Measurements were taken to 0.1 kg accuracy. Monitoring of weight and height was done monthly during the trial.

3.8. Statistical analysis

Data were double-entered and checked in EpiData v. 3.1 (Odense, Denmark), before being transferred to IBM SPSS v. 20 (Chicago, IL, U.S.A.), STATA 15 (paper I-III), R Core Team v. 4.1.0 (2021) (paper II), and Emergency Nutrition Assessment for SMART software 2011 (Toronto, Canada), for analysis.

Anthropometric data (weight, height) was converted to z-scores and percentiles. The results were categorized according to WHO Child Growth Standards (124) as follows. For WHZ, a z-score <-2 indicated wasting, ≥-2 to ≤2 indicated normal, and >2 indicated overweight. For HAZ, a z-score <-2 indicated stunting and ≥-2 indicated normal. WAZ z-scores <-2 indicated underweight, while ≥-2 indicated normal. Moderate and severe undernutrition were defined as z-scores <-2 and <-3, respectively (125).

Descriptive statistics, including frequency counts, percentages, and chi-square tests were used to present categorical data. Means, medians, confidence intervals (CI), and interquartile ranges (IQR) were used to present continuous variables and summarized using tables and figures. The conceptual hierarchical modelling approach published by

Victoria *et.al* was used to determine factors associated with iron deficiency anaemia and co-morbid anaemia and stunting (126) (Figure 3).

Bi-variable analysis was done for the outcome variables, taking all factors into consideration according to the framework (Figure 3). Variables that had p-values <0.3 were included in the multivariable regression analysis of factors associated with iron deficiency anaemia, using the three-level hierarchical regression analyses (level one underlying factors, level two intermediate factors, and level three immediate factors).

The two analytical papers (I-II) assessed iron-deficiency anaemia as explained above as a dependent variable (paper I). For paper II, the dependent variable consisted of four nominal categories: ‘no anaemia or stunting’, ‘stunting only’, ‘anaemia only’ and ‘stunting and anaemia together’ (CAS). The ‘no anaemia or stunting’ category was set as the reference. Potential factors associated with CAS were identified based on a conceptual framework. Then the least absolute shrinkage and selection operator (LASSO) was used to select variables for the multinomial logit model. Thus, independent variables were analysed with a multinomial logistic regression model to identify factors associated with co-morbid anaemia and stunting (CAS), and reported as crude odds ratio (COR) and adjusted odds ratios (AOR).

Before multivariate analysis was performed, we checked for baseline differences in anaemia, haemoglobin, and ferritin distributions between the study arms. The intra class correlation coefficients (ICC) were checked prior to running the multivariate analysis among the arms’ 1:1 randomly allocated eight clusters. Intention-to-treat analysis and complete case analysis were used for the prevalence of anaemia, iron deficiency anaemia, iron deficiency and haemoglobin level between arms. A bivariate and multivariate generalized linear model for binomial family regression was then run, and a relative risk (RR) with confidence intervals was reported. The generalized estimating equation (GEE) modelling was used to carry out multivariate linear regression analysis for the dependent variable haemoglobin and beta coefficient reported. All variables with a p-value less than 0.05 at a 95% confidence interval were considered to be statistically significant.

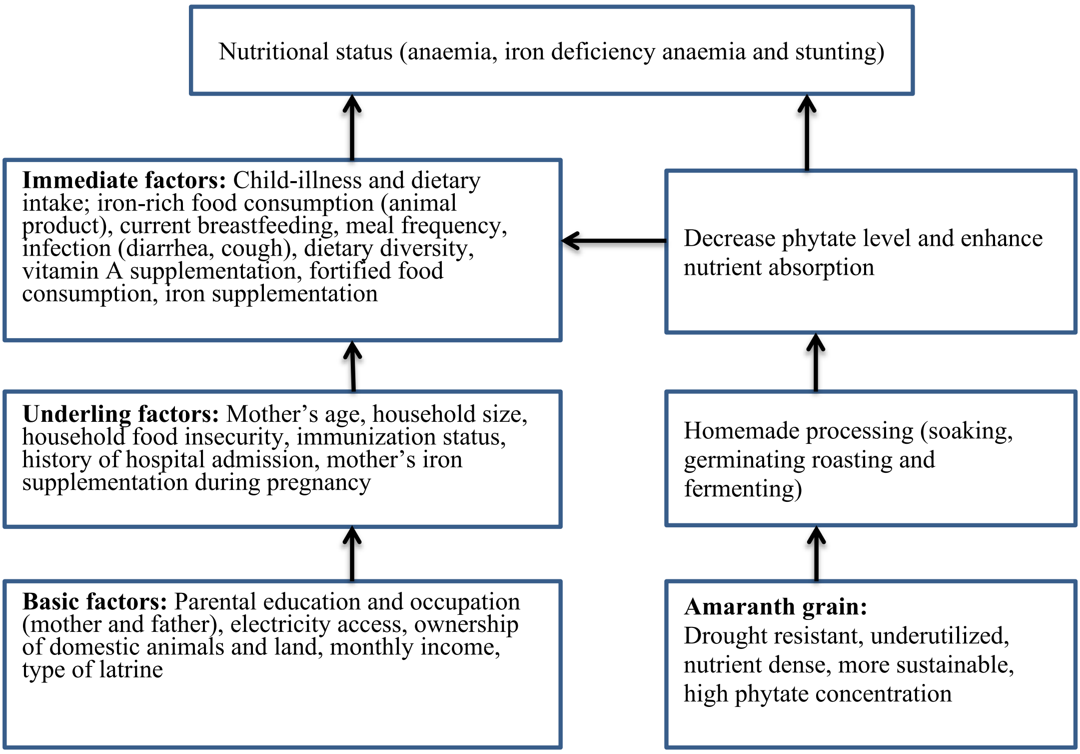

Figure 3: The connection between the factors could be summarized in a framework adapted to the UNICEF framework by Urban Johnson (1991) (38, 126-128), the relationship of amaranth to factors added.

3.9. Ethical considerations

The Institutional Review Board of Hawassa University (IRB/098/08) and the Regional Ethical Committee West of Norway (No. 2016/2034) provided ethical approval. Written permission was obtained from the Tulla Health Bureau. The Cheffe Cotijabessa health centre and health post authorities also granted official permission. Informed written consent was obtained from the mothers. The trial protocol was registered at the Pan African Clinical Trials (registry number: PACTR201705002283263). The respondents were the caregivers of the children as the children at this age were too young to respond. An informed and signed consent was sought from the mothers/caregivers before the study. The research purpose and protocols were explained in detail to the local administration, community leaders and the mothers/caregivers, and permissions were granted before the data collection. The caregivers were informed of the benefits of the study, which were expected to improve the health status of the children, and the anticipated risks of participating in this study. In the study, risks included blood collection risks at the baseline and after the intervention. The respondents were assured of confidentiality, which was maintained during and after the study, and also assured that any information from the study was to be used for the purpose of the study only.

Table 4 Summary characteristics of the PhD papers studying IDA and CAS in children 2-5 years of age in Southern Ethiopia, and an experiment to reduce anaemia in children

	Paper I	**Paper II**	**Paper III**
Paper topics	Iron deficiency anaemia	Co-morbid anaemia and stunting and associated factors	The efficacy of processed amaranth-containing bread on iron deficiency anaemia, haemoglobins and anaemia prevalence
Study design	Cross-sectional	Cross-sectional	Trial
Study participants consented	340	340	100
Study participants included in complete-case analysis	331	331	82
Data/key variables	Interview data, haemoglobin, ferritin, and CRP, weight, height	Interview data, haemoglobin, weight, height	Interview data, haemoglobin, ferritin, and CRP, weight, height
Descriptive statistical methods	Descriptive statistics and summary statistics of diet and anaemia and IDA	Descriptive statistics and summary statistics of stunting and anaemia and factors presented according to UNICEF framework (1991)	Descriptive statistics and summary statistics by arm allocation of haemoglobin and anaemia
Analytical methods Regression analysis	Backward logistic regression	Multinomial logistic regression LASSO	Intention-to treat analysis using: Generalized linear model for binomial family(anaemia) Generalized estimating equation (haemoglobin)
Outcome variable	IDA (composite variable: Hgb < 11g/dl and CRP adjusted ferritin <12 μg/L) analysed as presence/absence of IDA	CAS (composite variable including no anaemia and no stunting, anaemia no stunting, stunting, no anaemia, and stunting and anaemia combined)	Anaemia, haemoglobin and iron deficiency anaemia.

4. Results

4.1. Study population child characteristics

From the sampled 340 children, 331 participated in the survey (97.4% response rate). Of the 331 children, 172 (52%) were girls, and the mean age was 39.2 months (95% CI: 38.1–40.6) (paper I and II). Out of 107 anaemic children, 100 were eligible to be included in the trial. They were divided into eight clusters that were randomized 1:1, resulting in 50 children in each allocation group. Of those, 56% and 44% were girls in the maize and amaranth groups, respectively, and the mean age of the children was 37 months (95% CI: 34.8-39.1) with a small difference between the allocation groups.

In the trial, 82 children remained at the end of the scheduled visits, 41 children in each group. In total, 10 refused to take bread, 3 changed their village and 5 refused to attend the last measurement session (Fig. 4) (paper III).

4.2. Mother and household characteristics

A total of 331 mothers participated in the survey and their median age was 27 years (IQR 24–30). The majority of the mothers, 231 (69.8%) were housewives (working at home) and 75 (22%) had no formal education. The mean number of people living in the household was 5.0 (95% CI 4.8–5.2). Three-quarters of the households had a monthly income which was less than 1,900 Birr per month (<$1.90 per day) and 207 (62.5%) households were food insecure (paper I and II).

In the trial, of the 100 children included all the caregivers were mothers. The mothers' median age was 26 (IQR: 24-30) years. The median completed school years for mothers was 4 (IQR: 0-7) years, and 63 of the mothers were unemployed (working at home). The majority (77) of the households earned less than USD 1.90 per day, and 69 of the households were food insecure (paper III).

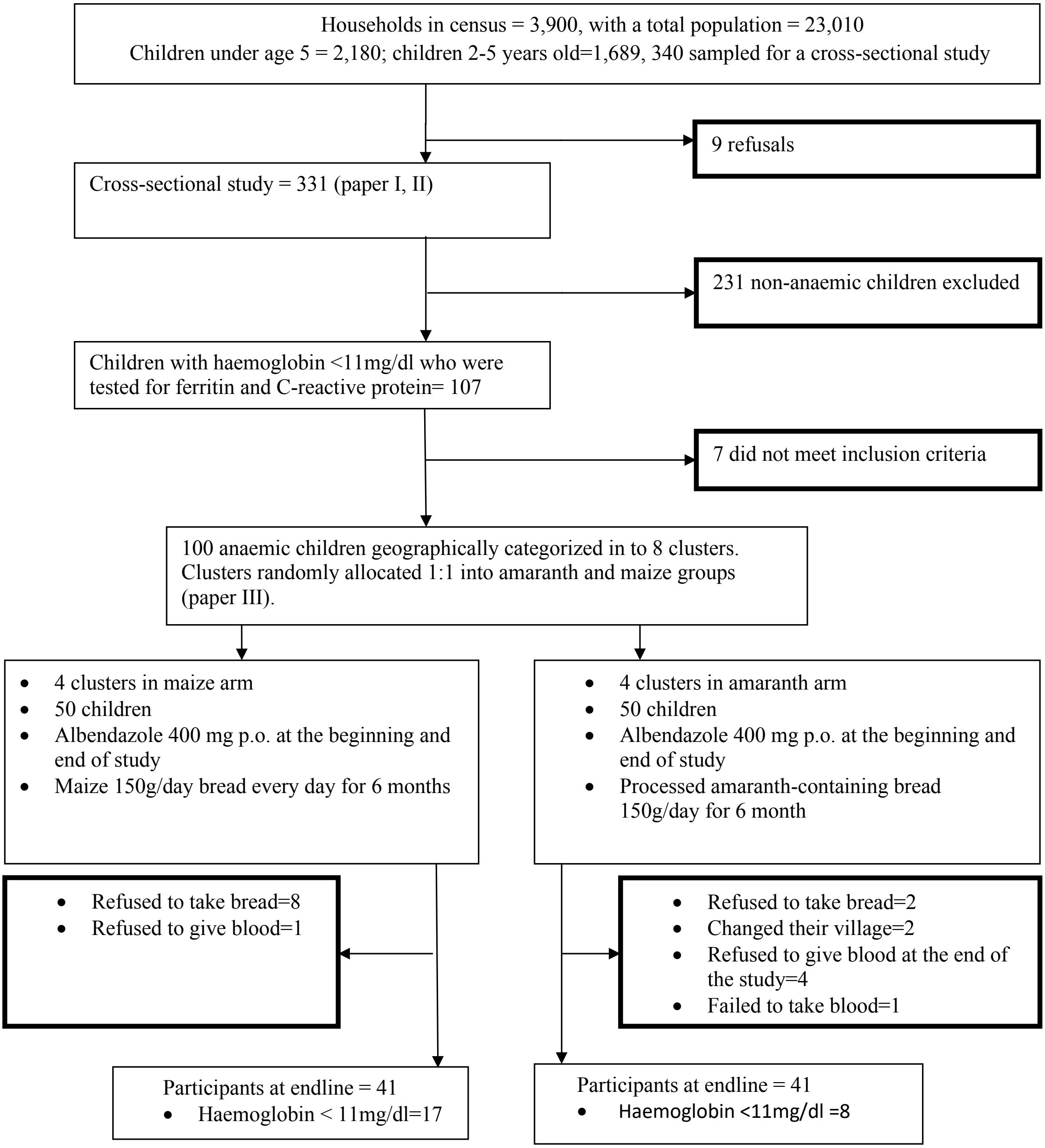

Figure 4 Project profile indicating participants in the census, cross-sectional study and the trial and number of exclusions at the various stages.

4.3. Morbidity

The children's illness history indicated that in the last two weeks preceding the survey, 100/331 (30%) children had watery diarrhoea, and 71/331 (21%) had a cough (paper I and II). Among the anaemic children participating in the trial, 38/100 (38%) had a history of watery diarrhoea and 22/100 (22%) had a history of cough in the past 15 days preceding the survey at baseline (paper III).

4.4. Dietary practice

Close to three-quarters of the children participating in the cross-sectional study, 242/331 (73%), had the lowest dietary diversity score, according to the 24-hour recall. About 15/331 (4.5%) of children consumed fortified food and only 10/331 (3%) children received iron supplementation (paper I and II). Only 50/331 (15%) of the children had consumed iron-rich food (meat, organ meat and fish). Almost all children ate starchy staples 326/331 (98.6 %). The seven-day food frequency data showed that 104/331 (33%) children consumed iron-rich food at least once within the last seven days. Furthermore, household iron-rich food intake was similar to that of the children for the last 24 hours, as only 51/331 (15%) of the households consumed iron-rich foods (Paper-II).

4.5. Children's nutritional status

Anaemia affected approximately one-third of the children in the cross-sectional study 107/331 (32.3%). Of those, only 1 (0.9%) child had severe anaemia (excluded from trial and sent to treatment), 29 (27.1%) had moderate anaemia, and 51 (47.7%) had mild anaemia (paper I-III). The CRP measurements indicated that 23/107 (21 %) of anaemic children had some sort of inflammation (paper I and III). From the total number of anaemia cases, IDA was 27 (25%) (paper I and III). The prevalence of stunting was 125/331 (38%) (paper I and II). The overall prevalence of co-morbid anaemia and stunting (CAS) among the study participants was 59/331 (17.8%) (Paper II).

In the trial population at baseline, the majority 73/100 (73%) of children scored in the lowest dietary diversity group, 53/100 (53%) were stunted, 4/100 (4%) wasted and 28/100 (28%) were underweight. Stunting was more common in the amaranth group 31/50 (62%) than in the maize group 22/50 (44%). At six-month follow-up, the majority of children also scored in the lowest dietary diversity group. The differences between the allocation groups were small and not statistically significant. The seven-day food frequency recall indicated that 59% of children had not consumed iron-rich food at least once in the last six consecutive months of follow-up. There was no significant difference between the allocation groups on iron-rich food consumption (paper-III).

4.6. Factors associated with IDA

Using a higher ferritin cut-off adjustment approach, we estimated the odds ratios for IDA. The adjusted odds ratio showed that the prevalence of IDA significantly decreased as the height for age z-score increased (AOR: 0.7; 95%CI: 0.5-0.9) (paper I). Children's iron-rich food consumption at least one time in the last seven days was associated with mother's education (AOR: 1.1; 95%CI: 1.0-1.2) and high household dietary diversity (AOR: 1.4; 95%CI: 1.2-1.6) (paper I).

4.7. Factors associated with anaemia and stunting

Children whose mothers had not taken iron supplementation during pregnancy were more likely to develop anaemia than children whose mothers had such history, AOR: 3.1; 95%CI: 1.5–6.5.

Age and meal frequency were significantly associated with stunting. Stunting increased as the age of the child increased, (AOR: 1.1; 95%CI: 1.0-1.1). When the meal frequency increased, the occurrence of stunting decreased, (AOR: 0.6; 95%CI: 0.3-0.9).

4.8. Factors associated with co-morbid anaemia and stunting (CAS)

Child age, iron supplementation during pregnancy and household food security status were associated with CAS. When children's age increased by one month, it increased the occurrence of CAS (AOR: 1.0 95%CI: 1.0-1.1). Children from a mother who did not take iron supplementation during the last pregnancy were three times more likely to develop CAS (AOR: 2.9; 95%CI: 1.3-6.2) compared with those who took iron supplementation during the last pregnancy. Further, CAS was less common in children who lived in food-secure households, compared to children who lived in food-insecure households, (AOR: 0.3; 95%CI: 0.1-0.9) (Paper-II).

4.9. Trial: amaranth effect

Also, there was no large difference between those that discontinued (n=18) and those that remained in the trial through to the last follow-up measurement. There were some differences between the allocation groups at baseline, including sex and stunting as mentioned. The six month daily bread consumption attendance indicated that there was no significant mean differences between the amaranth and maize groups, (-5.3; 95% CI:-20.1; 9.5) with p-value > 0.5 (paper III).

At the last follow-up measurement session, the overall prevalence of anaemia decreased to 44% as compared to 100% at baseline. Intention-to-treat analysis indicated that children who received amaranth containing bread had a 61% significant reduction in the risk of anaemia, compared to those who received maize bread, (aRR: 0.3; 95%CI: 0.1-0.7). The prevalence of anaemia was significantly lower (32%) in the amaranth group compared to the maize group (56%). Similarly, the complete-case analysis results indicated a significant decrease in anaemia prevalence in the amaranth group (17 %) as compared with (46%) in the maize group, (aRR: 0.1; 95%CI: 0.0-0.5), (paper III).

The last follow-up measurement indicated that haemoglobin concentration was increased in both the amaranth and maize arms, compared with the baseline results. The adjusted beta coefficient (aβ) indicated that children who received amaranth bread had a significantly higher amount of haemoglobin concentration than those receiving maize bread, (aβ 8.9 g/L; 95%CI: 3.5-14.3) (paper III, Table 9).

The overall prevalence of iron deficiency anaemia decreased from 29% at baseline to 18% at the last follow-up measurement. Intention to treat analysis showed that iron deficiency anaemia risk significantly decreased in the amaranth group, from 35% at baseline to 15% at the last follow-up, (aRR: 0.4; 95% CI: 0.2-0.8). However, this difference was not significant in the complete case analysis. In the amaranth group, iron deficiency anaemia decreased from 34% at the baseline measurement to 26% at the last follow-up measurement, which was not statistically significant, (aRR: 0.8; 0.5-1.1) (paper III).

4.10. Summary of the result

The study found that children who ate amaranth bread had significantly higher haemoglobin concentrations (aβ: 8.9 g/L; 95% CI: 3.5-14.3) and lower anaemia prevalence (aRR: 0.3; 95% CI: 0.1-0.7) than children who ate maize bread. Anaemia, stunting and CAS were highly prevalent and food-insecurity was associated with the undernutrition. Three-quarters of the households had a monthly income which less than $1.90 per day, and 207 (62%) households were food insecure. Close to three-quarters of the children, 242 (73%), had the lowest dietary diversity score. Only 15 (4%) of children consumed fortified food and only 10 (3%) of those children received iron supplementation. In the last 24 hours, only 50 (15%) children had consumed iron-rich food (meat, organ meat, and fish).

5. Discussion

5.1. Methodological discussion

5.1.1. Study design

This thesis used two types of study design: a cross-sectional study (paper I and II) and a cluster randomized controlled trial (paper III). Initially, we planned a simple randomized controlled trial, but we ultimately ended up with a cluster-randomized controlled trial. A cluster-randomized trial minimizes the likelihood of bias in the assignment of alternative interventions, but it is less effective and needs a larger sample size than a simple randomized controlled trial.

Preceding the trial, a cross-sectional survey was conducted using a simple random sampling technique, and from this, 100 eligible children with anaemia were identified. Children with anaemia were sparsely distributed in the study area. We therefore recognized that a simple randomized allocation for the trial intervention at the individual level was unmanageable. Since we planned to feed children every day at home under direct supervision, we decided to group the children in manageable geographical clusters and randomize the clusters. This made distribution of bread and close follow-up easier. At this decision stage, we might have introduced selection bias, but we anticipate this was minimized due to the simple random sampling technique used to identify the target population (paper I and II). During statistical analysis; intra class correlation (ICC) was used to determine how large the differences between the clusters were, and these were found to be small and not statistically significant (paper III).

5.1.2. Sampling

Sampling is the process of selecting a number of subjects from all the subjects in a particular group. This is a practical option when one cannot select everybody from that group (e.g. all women in sub-Saharan Africa) (129). This thesis used a simple random sampling technique for paper I and paper II within a strictly defined sampling frame.

Prior to the survey, we conducted a census of the study area to obtain a sampling frame. A simple random sampling technique was used to find the target population from the census list (paper I and II). We calculated the sample size assuming 33% anaemia prevalence, for paper I and II. However, we did not calculate a sample size specifically for paper II, but we did a post-hoc test and we found that the sample size we used for paper I (anaemia) was adequate to assess co-morbid anaemia and stunting (paper II). The sample size for the trial was calculated with the aim of being able to detect a clinically interesting mean difference in haemoglobin levels.

5.1.3. Validity

Validity is used in epidemiology to assess the degree to which the information collected accurately answers the research question. For this thesis, the focus is on internal validity (study validity), measurement validity (variable) and external validity (generalizability) (130).

Internal validity is the degree to which the observed findings lead to correct identification of inferences regarding the phenomena taking place in the study sample. A study is not valid if it cannot provide accurate information, or cannot enable well-founded inferences to be drawn from the population studied. Internal validity can be compromised by selection bias, information bias, uncontrolled confounding, and a small study sample.

5.1.4. Selection bias

Selection bias is a systematic error in a study that stems from the procedures used to select subjects and from factors that influence study participants. It happens when the association between exposure and disease differs for those who participate and those who do not participate in the study (130). In paper I and II, the study area was selected purposively to be used as a baseline for paper III, which was the experimental part of this project, but the sampling method was a simple random sampling method (paper I and II). The simple random sampling method gives an equal chance higher than zero for each of the participants within the sampling frame to be selected, thus sampling bias for the cross-sectional study was most likely small.

Initially, in paper III, we designed the sample size using the simple random sampling method. When we got to the field, after identifying the target group of children with anaemia, we recognized that the children with anaemia were sparsely distributed among the community. This could challenge the implementation of the intervention, which was to feed children every day at their home under direct observation. For this reason, we choose to group children with anaemia based on their geographical location, which could be manageable during fieldwork. We saw in our descriptive tables stratified on randomization allocation that there were only small differences in sociodemographic characteristics, but there was a difference in the distribution of stunting based on baseline result. We adjusted for this variable in the multivariate analysis. Further, there was no statistically significant intra-class correlation between groups.

In the trial, we tried to decrease the non-response rate by doing day-to-day follow-up through caregivers. Caregivers had to do close follow up every day and follow those who moved from their homes. At the end of the study, we faced an 18% non-response rate, 9% from the amaranth group and 9% from the maize group. This might have created selection bias, or loss-to-follow-up bias. The loss-to-follow up was non-differential with respect to allocation groups, and not related to any particular socio-demographic characteristics. In general, we assume selection bias was minimized in this thesis, particularly due to the simple random sampling and randomization of clusters.

5.1.5. Information bias

Information bias is a systematic error in a study that can arise because the information collected about or from study subjects is erroneous. It could be observer bias, subject bias or instrument bias (130). All three papers are prone to information bias in one way or another.

It is difficult to avoid information bias, but we can minimize it by using different techniques. In general, to minimize information bias, all data collectors in the research studies received training in the tools, including informed consent forms, measurements and questionnaires. Following training, a community pre-test was conducted, and the tools were revised prior to the start of the survey.

Recall bias is the main one that can be introduced into all three papers. In all the papers, age was one of the variables with a high chance for recall bias, and it is essential for calculating age-dependant anthropometry measures in all three papers. All children lacked a vital registry card, and the majority of the mothers were uneducated and may have struggled to recall the exact date of their childbirth. To solve this problem, we used the immunization card and the local calendar. During data cleaning, we went back to the exact homes where we were in doubt to recheck the age of the children. Further, the 24-hour dietary diversity recall, 7-day food frequency recall, history of illness recall, and other child feeding characteristics recalls were prone to recall bias in all three papers. To minimize recall bias for dietary intake in the trial part, we measured dietary diversity and iron rich food frequency every month and the mean intake was calculated and compared across the study arms.

This thesis is also prone to instrument bias. To minimize this bias, adequate training was given to measuring anthropometry for the data collectors, and the instruments were checked every morning before starting data collection using a standard weight and height. The average of two measurements was taken to reduce intra-observer bias, and close follow-up was done during the anthropometry measurements. In all papers, the haemoglobin measurement was done by two laboratory technicians, each of whom had experience from fieldwork. Ferritin and CRP levels were analysed in a standard laboratory and by qualified blood chemistry professionals. The same laboratory technicians did baseline and endline measures. We acknowledge that information bias may exist despite our attempts to reduce it.

5.1.6. Confounding

Confounding is the effect of another variable on the relationship between that of the investigated exposure on an outcome and is a type of bias. Confounding should be identified to avoid systematic error in interpretation of results. Confounding can be controlled for in two phases: in the design phase and in the analysis. In the design phase, the methods are randomization, restriction, and matching (130). In this thesis, we didn't use restriction and matching for arm allocation, but randomization of clusters was used for paper III. Since randomization alone did not ensure completely similar groups we controlled for confounding in the analysis (paper I-III).

5.1.7. Random error (chance)

A random error (chance) is the error that remains after a systematic error (bias) is eliminated. It can arise from the sampling variability of a study when an inference about the entire population is based on a sample of the population. Chance cannot be avoided without including the whole population, but it can be reduced by increasing the sample size (131). It can also be estimated by performing appropriate statistical tests using p-values and confidence intervals (130). A p-value indicates incompatibility between a particular set of data and a proposed statistical model for the data (132). If the p-value is low (e.g. <0.05), it is unlikely that the observed results are caused by chance alone. If it is high, it is more likely that the results are due to chance. Further, the confidence interval provides a range of values in which the true estimated effect is likely to lie, with a certain degree of assurance. The confidence interval is more informative than the p-value because it provides a range of magnitude of the effect and variability in the estimate due to sample size (130). In this thesis, we evaluated the role of chance using appropriate statistical models by applying both p-values (<0.05) and 95% confidence intervals.

5.1.8. External validity (generalization)

External validity depends on adequate internal validity. It refers to whether or not a research finding can be generalized to a population not in the study or outside the study area (130). The studies for this thesis were conducted in a farming community, and the area was purposely selected to make it accessible for the experimental study (paper III). Even if the area represents a semi-urban part of a town, many of the household members practice agriculture typical of many areas in southern Ethiopia. The study used a representative sample using a simple random sampling technique at a community level. Moreover, we used validated questionnaires and standard and calibrated haemoglobin, height, and weight measurements. Due to all these factors, the result of paper I and paper II could be reproducible in the same semi-urban area of Ethiopia, and in a similar setting in another low income country. As paper I and II used a cross-sectional design, the direction of the observed relationships between exposures and outcomes cannot be determined, but as discussed in the individual papers, many of the findings are supported by others.

Paper III followed standard consort requirements for an experimental study (133) (Appendix consort checklist) that used a simple random method to identify the study participants and cluster randomization to allocate anaemic children into the two study arms. The bread makers, bread distributors, caregivers, and data collectors were unaware of the maize or amaranth bread allocations. The caregivers fed their children daily under direct supervision. Flour blending was performed carefully, considering the ratio of amaranth, and the ratio and size of the bread were checked regularly by a trained supervisor. The results obtained in this thesis would most likely be reproducible in a similar setting if done in a similar manner. A multi-site and larger cluster randomized trial would increase the confidence in the results obtained.

Iron deficiency anaemia was significantly decreased in the amaranth group in the intention to treat analysis but not in the complete case analysis. This may be due to the small sample size, so caution should be taken interpreting this negative finding. This thesis did not assess other nutrient-related anaemia, such as folic acid deficiency, vitamin A deficiency, and copper deficiency, and did not consider genetically related haemoglobin disorders; however they are not common in the study area. Further, we didn't measure AGP (a-1-acid glycoprotein) concentration to adjust ferritin. In addition, we did not conduct malaria tests or stool examinations (paper I-III), all of which would have contributed to improving the study.

5.2. Discussion of the main finding

The central theme of this thesis was to assess the efficacy of bread containing processed amaranth grain on the anaemia and IDA status of children. Amaranth containing bread consumption increased haemoglobin concentration and decreased anaemia prevalence in the targeted children when compared with the maize bread. Child malnutrition in the form of anaemia and stunting is a public concern in the study area, and was found to be associated with the common predisposing factors including household food insecurity, child age, and maternal iron supplementation during pregnancy. Poverty, inadequate intake of nutrients and the monotonous feeding habits in the community were the root causes of the nutritional problems for children in the study area.

To our knowledge, there have only been a few other studies that have examined the effect of processed amaranth grain on anaemia status, to which our results can be compared. However, one study conducted on amaranth leaf in Ghana showed a similar result to this study (112). In contrast, a study conducted in Kenya using unprocessed amaranth grain demonstrated that amaranth did not decrease anaemia prevalence significantly (134). This may be due to the high phytate levels in raw amaranth. The same study suggested that reducing the phytate level of amaranth grain may contribute to increasing the haemoglobin level. Other studies have shown that homemade processing of amaranth grain, such as soaking, germinating, and fermenting, can result in a lower phytate level (95, 135). Therefore, we believe that the results obtained in our study are related to the homemade processing that we applied to amaranth grain, which decreased the phytate level and enhanced the absorption of iron and other micro-and macro-nutrients.

Iron deficiency anaemia was significantly decreased in the amaranth group in the last follow-up measurement in the intention to treat analysis. But the change was not significant in the complete case analysis. In addition to the epidemiological limitations, this finding may also be due to the presence of inflammation among the studied children at baseline, confirmed by high CRP levels in both arms when compared with the last follow up. This is supported by other studies reporting that CRP and ferritin have a positive correlation (136). This means some of the ferritin levels observed as normal at baseline measurement may have been related to the presence of inflammation. We adjusted ferritin with CRP but this may not have controlled for elevated ferritin (123). In the future, a larger sample size and the inclusion of additional iron indicator tests may provide further details on the iron status of children (123).

Anaemia was noticeably decreased in the maize arm, as well. This may be attributable to a reduction of helminth burden due to the albendazole given to children in both allocation groups. This can be supported by research conducted on the effect of deworming on anaemia prevalence (137, 138). Furthermore, the daily supervision by a bread provider at the household level may have created awareness of sanitation and child-feeding practices, which in turn may have had a positive impact by decreasing the incidence of inflammation and improved dietary diversity. The fermented maize bread itself may also have increased the intake of iron, as all of the children may have benefitted from the actual feeding itself.

Results presented in paper I indicated that one-fourth of the cases with anaemia were due to IDA. The 24-hour and 7-day food frequency recalls revealed that only 15% and 30% of children consumed iron-rich food, respectively. These findings are comparable with 20% of iron-rich food consumption by children less than five years of age within the last 24 hours preceding the survey reported in Ethiopia (139). In this study, improved height-for-age was inversely associated with IDA, and similar findings were reported from Pakistan (140). This could be because iron deficiency may lead to delayed growth and development in children. A study from Qatar showed that treatment with iron for iron-deficiency anaemia improved the growth of children under 2 years of age (141).

Findings presented in paper II demonstrated an 18% prevalence of CAS among the studied children. This finding was lower than in a study done in Ethiopia among younger children, 6 to 23 months of age, that reported a 24% prevalence of concurrent anaemia and stunting (46) and higher than a study done in southern Ethiopia among school children, 10% (142). Children living in food-insecure households had higher rates of CAS than those in food-secure households. Higher anaemia prevalence (16) and higher stunting prevalence (143, 144) in food-insecure households have been reported in Ethiopia. Food-insecure households have limited access to diversified food and have lower meal frequency than food-secure households. The study done among children 6 to 23 months of age in Ethiopia found that low meal frequency significantly increased the co-occurrence of stunting and anaemia (46). This was supported by other studies undertaken in Ethiopia which reported inadequate energy intake and low-quality protein consumption as the main causes of stunting (108, 145).

We also found that children born to mothers who took iron supplements during their last pregnancy were less likely to develop CAS than those who had mothers who did not get supplements during pregnancy. We have not found other studies that have investigated the relationship between iron supplementation during pregnancy and CAS in children aged 2-5 years. However, some studies report low iron intake of mothers and having a mother with symptoms of anaemia as predictors of anaemia in children less than 5 years of age (45, 146).

The described relationship between stunting and anaemia, and IDA and HAZ (where higher HAZ was associated with reduced risk of IDA) were supported by a study done in Ethiopia which found that children who were stunted were 2.5 times more likely to have anaemia compared with children who were not stunted (147). Several other studies have also reported that the presence of anaemia increase the occurrence of stunting and *vice versa* (45, 146).

This study also identified low dietary diversity, low meal frequency, and low consumption of animal source food by children in the study area (paper I-III). Food fortification had not been implemented and further, more essential micronutrient supplementation had not been applied in the study area. WHO recommends iron-fortified food implementation when the iron-deficiency anaemia prevalence is >5% and iron daily supplementation for pre-school children if the anaemia prevalence is more than 40% in children aged 0-24 months. In settings where the prevalence of anaemia in preschool (age 24–59 months) children is ≥20%, WHO recommends intermittent use of iron supplements as a public health intervention to improve iron status and reduce the risk of anaemia among children(12). None of these recommendations were implemented in the study area, even though the prevalence of anaemia and IDA exceed the recommended WHO cut-off.

6. Conclusion

The trial results revealed that the consumption of bread containing processed amaranth increased mean haemoglobin levels and decreased anaemia prevalence, and improved ferritin levels among the study participants. Processed amaranth grain bread therefore has the potential to minimize the prevalence of anaemia in children. CAS and IDA are public health concerns in the study area. We found that one-third of the children aged 2-5 years in this study of households in southern Ethiopia were anaemic. Close to one-fifth of the children were iron-deficient and had signs of inflammation. Stunting was a severe public health problem in the study area with 37.8% prevalence, and the prevalence of comorbid anaemia and stunting was 18%.

The common factors associated with co-morbid anaemia and stunting were household food insecurity and lack of iron supplementation by mothers during pregnancy. Coverage of food fortification and micronutrient supplementation for children was almost negligible in the study area. The high prevalence of anaemia and stunting is a chronic manifestation of child nutritional problems in the study area. It should receive the attention of the public and decision-makers.

7. Recommendations

There is a need to strengthen the implementation of iron supplementation during pregnancy and focus on food supplementation and fortification strategies as short-term implementation strategies. Long term strategies need to include improving household food security, food diversification, nutritional education, promoting women's education and reducing the high burden of inflammation in the study area.

Furthermore, emphasis should be given to promoting and investigating the production and consumption of amaranth plants, which are widely grown in the study area but underutilized by the communities. Promoting amaranth production could be a cost-effective strategy that could improve the food security of households in the study area.

8. Future research

Future research should consider the assessment of IDA by including AGP and malaria status to see any variation in the prevalence. Further, studies on potential causes of other types of anaemia that could be prevalent in the area, such as folic acid deficiency, vitamin A deficiency, and copper deficiency, as well as genetically related haemoglobin disorders, malaria and gastric tract diseases and infections should also be considered. The energy content of processed amaranth containing bread and its effect on the weight and height of children also should be studied. Thus, we recommend multi-site larger cluster randomized and more comprehensive trials on this potentially beneficial food item.

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Appendices

Paper I

Iron deficiency anemia among children aged 2–5 years in southern Ethiopia: a community-based cross-sectional study

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ABSTRACT

Background: Iron-deficiency anemia (IDA) is a common type of nutritional anemia in low-income countries, including Ethiopia. However, there is limited data on iron deficiency anemia prevalence and associated factors in Ethiopia, particularly for children aged 2 to 5 years.
Objectives: To establish the prevalence of iron deficiency anemia and associated risk factors, focusing on iron-rich food consumption among children aged 2 to 5 years in southern Ethiopia.
Methods: A community-based cross-sectional study was conducted in southern Ethiopia in 2017, involving 331 randomly selected children aged 2 to 5 years old. A structured questionnaire was used to collect information about the children and the households. Venous blood was collected from each child in a test tube to measure hemoglobin, ferritin, and C-reactive protein (CRP). Hemoglobin levels were determined using Hemocue®301 and adjusted for altitude. Anemia was defined as hemoglobin levels <11 g/dl. Ferritin was adjusted for inflammation based on CRP concentration and low ferritin concentration defined as adjusted ferritin concentration <12 μg/L. IDA was considered when a child had both hemoglobin level <11g/dl and low ferritin concentration. Bi-variable and multivariable logistic regression models were performed to identify factors associated with IDA and iron-rich food consumption.
Results: The prevalence of iron deficiency anemia was 25%, and the total anemia prevalence was 32%. Only 15% of children consumed iron-rich foods in the preceding 24 h, and 30% of children consumed iron-rich foods at least once in the preceding week. IDA decreased as the height for age z-score increased (Adjusted Odds Ratio 0.7; 95% CI [0.5–0.9]). Mothers with increased educational level (AOR 1.1; 1.0–1.2) and households with increased dietary diversity (AOR 1.4; 1.2–1.6) consumed more iron-rich foods.
Conclusions: Iron deficiency anaemia was a moderate public health problem in southern Ethiopia, and the iron-rich food consumption was low. Interventions should focus on food supplementation and fortification, food diversification and nutritional education, and promoting women's education.

Submitted 16 December 2020
Accepted 29 May 2021
Published 28 June 2021

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Academic editor
Charles Okpala

Additional Information and Declarations can be found on page 15

DOI 10.7717/peerj.11649

How to cite this article Orsango AZ, Habtu W, Lejisa T, Loha E, Lindtjørn B, Engebretsen IMS. 2021. Iron deficiency anemia among children aged 2–5 years in southern Ethiopia: a community-based cross-sectional study. **PeerJ 9:e11649 DOI 10.7717/peerj.11649**

Subjects Epidemiology, Hematology, Nutrition, Pediatrics, Public Health
Keywords Iron deficiency anemia, Iron-rich food consumption, Serum ferritin, Dietary diversity, C-reactive protein

BACKGROUND

One-fourth of the global population is affected by anemia, and the highest prevalence is in preschool age children (*World Health Organization, 2015*). In Ethiopia, 56% of the children under the age of 5 years were anemic (*Central Statistical Agency, 2016*). It has long been assumed that up to half of the cases of anemia are due to iron deficiency (*World Health Organization, 2005*). But the recent review found that iron deficiency accounts for 25 percent of anemia in young children and 37 percent of anemia in women of reproductive age. Moreover, significant variations exist between countries, which may render generalized assumptions misleading (*Green et al., 2017*).

Children are particularly vulnerable to iron-deficiency anemia because of their increased iron requirements during periods of rapid growth, especially in the first five years of life. In children, iron deficiency can affect cognitive and motor development and increase susceptibility to infections (*Batra & Sood, 2005*).

In developing countries diets are dominated by starchy cereals that are low in lipids, proteins, vitamins, and minerals including iron (*Harika et al., 2017*). Poor families, which constitute the majority of the population in Ethiopia as well as in other low-income countries, often cannot afford animal food products that have high amounts of easily absorbable iron (*Gebreegziabiher, Etana & Niggusie, 2014*; *Kejo et al., 2018*). In Ethiopia, iron-rich food consumption by children is less than 21% (*Tiruneh et al., 2020*; *Herrador et al., 2015*) and more than 70% of the children scored low on dietary diversity assessments (*Temesgen et al., 2018*; *Worku et al., 2020*).

The current public health policy is supporting efforts to reduce child malnutrition in general and child anemia in particular, by promoting dietary diversity, deworming, iron supplementation during pregnancy, and malaria prevention (*Federal Democratic Republic of Ethiopia, 2018*). Despite these efforts, the prevalence of anemia among children less than five years old increased from 44% in 2011 to 57% in 2016 (*Central Statistical Agency, 2016*).

Diagnosing iron deficiency anemia is challenging in the presence of acute or chronic inflammations (*Grant et al., 2012*). In Ethiopia studies of IDA are often restricted by a shortage of laboratory facilities, and because of challenges in interpretation of biomarkers for IDA due to high burden of infections (*Namaste et al., 2017b*). We assumed there would be a high prevalence of IDA among children in the study area due to the high burden of anemia, limited access to iron supplementation and fortified foods, and lack of iron in the foods (*Central Statistical Agency, 2016*). In Ethiopia, literature on IDA is not only limited but also inconsistent. Different studies reported varying prevalence rates. The Ethiopia National Micronutrient Survey in 2014 estimated 8.6% of IDA prevalence at the national level among children 6–59 months old (*Ministry of Health, 2016*), and a study from southwest Ethiopia reported 37% of IDA among school children (*Desalegn, Mossie & Gedefaw, 2014*). Hence, determining the prevalence of iron deficiency anemia is important to plan appropriate interventions and thereby to minimize future health risks (*Garcia-Casal et al., 2018*). This study aimed to establish the prevalence of iron- deficiency anemia and associated risk factors, focusing on the assessment of iron-rich food consumption among children aged 2 to 5 years in southern Ethiopia.

METHODS

Ethical consideration

The institution's ethical board of Hawassa University (IRB/098/08) and the Regional Ethical Committee West Norway (No. 2016/2034) provided ethical approvals. Hawassa University College of Medicine and Health Sciences approved to undertake field work (No.1471/09). Local administrative and health authorities of Hawassa City Administration Hawella Tula sub city Health Office (No. 3214/09) also granted official permission. Informed written consent was obtained from mothers for all study participants.

Study area

This cross sectional study is a part of a larger study that aimed to evaluate the efficacy of home processed amaranth grain containing bread in the treatment of anemia among 2–5 year-old children in southern Ethiopia (*Orsango et al., 2020*). Thus, the Cheffe Cote Jebessa Kebele was purposively selected because amaranth grain grows as a wild crop in the village. The plant amaranth was used to prepare an alternative plant-based iron-rich food that is easily accessible and acceptable for children (*Orsango et al., 2020*; *Zebdewos et al., 2015*). The study area is located in a semi-urban part of Hawassa city in southern Nations Nationalities Peoples Regional state, 273 km south of Addis Ababa, the capital city of Ethiopia. The study was conducted from February 15, to March 30, 2017. According to a census we conducted before the start of our study, the population of the village was 23,010 people with 3,900 households, and 1,689 children aged 2–5 years old. Since it is a semi-urban area, some of the household members are farmers and some of them are engaged in non-agricultural activities in Hawassa city. The farmers cultivate maize, haricot beans, *ensete*, and Irish potatoes.

Sampling procedure and sample size

The sample size of 340 children was calculated using OpenEpi software version 3.03 (*Sullivan, Dean & Soe, 2009*) considering the 33% national average prevalence of anemia among children aged 3–5 years old (*Central Statistical Authority, 2012*), 95% confidence level, and 5% margin of error. From the census list, 340 children were randomly selected using SPSS random generation technique (Fig. S1).

Study participants

This cross-sectional study was done before the trial (*Orsango et al., 2020*), and we randomly selected 340 children aged 2 to 5 years and their mothers or caregivers, all of whom lived in the study area and who provided informed consent. We excluded children with conditions that precluded them from participating in measurements and venipuncture.

Study variables

The primary outcome variable of this study was iron deficiency anemia, and secondary outcome variable was iron-rich food consumption. Independent variables of the study were socio-demography factors, economic status, dietary intake, and nutritional conditions of children.

Blood collection and laboratory methods

Professional laboratory technicians have collected 3–5 ml of blood specimens from the vein by using a lithium heparin plasma separator test tube that used to measure hemoglobin, serum ferritin, and CRP (*Wei et al., 2010*). Hemoglobin was determined immediately on-site using HemoCue analyzers 301(Sweden). All samples with hemoglobin levels <11g/dl were tested further for serum ferritin and CRP concentration. For determination of serum ferritin level and CRP, the blood samples in the test tubes were immediately wrapped in aluminum foil, continuously shielded from light, and stored for 30 min at 4 °C until centrifugation. After centrifuging, serum was kept in plastic screw-capped tubes with the participant's identity and stored at −20 °C. Then the serum was transported in a cold box chain for analysis of ferritin and CRP to the Ethiopian Public Health Institute at Addis Ababa.

C-reactive protein (CRP) was measured on Cobas 6000 (c501 module) Roche (Germany) by applying enhanced immunoturbidimetric assay principle. Ferritin was measured on Cobas 6000 (e601 module) Roche (Germany) by applying electrochemiluminescence immunoassay (ECLIA) specifically the sandwich principle. Ferritin results were determined by 2-point calibration and a master curve provided via the reagent barcode.

Biomarkers and definitions used to assess ferritin level

IDA was evaluated based on hemoglobin, serum ferritin, and CRP concentration. Hemoglobin concentrations were corrected for altitudes by subtracting 0.8g/dl per 500m altitude to get the estimated sea level value (*Sullivan et al., 2008*; *World Health Organization, 2011a*). Anemia was defined as hemoglobin concentration adjusted for altitude less than 11g/dl and classified as normal (≥11.0 g/dl), mild (10.0–10.9 g/dl), moderate (7.0 –9.9 g/dl), and severe (<7.0 g/dl) (*Sullivan et al., 2008*).

Ferritin concentration was adjusted for inflammation using correction methods recommended by WHO in 2020 (*World Health Organization, 2020*). First, the prevalence of low ferritin (<12 μg/L) was calculated on the basis of unadjusted ferritin estimates. Subsequently, the following three adjustment approaches were applied to account for inflammation under this study:

- Exclusion approach: The exclusion approach excluded individuals with elevated CRP concentration >5 mg/L, and calculated the prevalence of low ferritin in the remaining individuals considering ferritin concentration cut-off <12 μg/L.

- Higher ferritin cut-off: The higher ferritin cut-off adjustment approach uses a higher ferritin concentration cut-off <30 μg/L. There were two different approaches when the high ferritin cut-off was used. The cut-off applied for the entire sample as well as the subset of individuals with elevated CRP as defined by a CRP concentration >5 mg/L. For this study, we used cut-off for the subset of individuals with elevated CRP (*World Health Organization, 2020*).

- Internal correction factor: The correction factor (CF) was calculated as the ratio of geometric means of the reference group (non-elevated CRP) to that of the respective inflammation groups (elevated CRP). We found a CF value equal to 0.85 (*Thurnham et al., 2010*; *Namaste et al., 2017b*; *Namaste et al., 2017a*). Then, to get the adjusted ferritin the subgroup with CRP>5mg was multiplied by 0.85 (*Gibson, 2005*; *World Health Organization, 2011b*). Subsequently, low ferritin was defined <12 µg/L for both groups of elevated CRP or not elevated CRP.

Finally, we used the higher ferritin cut-off adjustment approach because we got the higher prevalence of iron deficiency anaemia by this approach. Furthermore, we measured only CRP not have the data on AGP concentration and malaria, this could prevent us to rely completely on correction factor and the regression correction approach (*World Health Organization, 2020*). Iron deficiency anemia was defined as children having a combination of hemoglobin level of less than 11g/dl and adjusted low ferritin concentration (<12 µg/L) (*World Health Organization, 2014*; *World Health Organization, 2011b*).

Anthropometric measurements

The weight and height of children were measured to assess nutritional status. Height was taken using a Seca213 height board (Seca 213; Seca GmbH, Hamburg, Germany) with a sliding headpiece while the child stood straight. Weight was taken using a calibrated Seca 874 electronic flat scale (Seca 874; Seca GmbH, Hamburg, Germany) with the child barefoot and wearing light clothing. Anthropometric measurements were calculated according to the Emergency Nutrition Assessment for SMART software 2011 (Toronto, Canada), developed using WHO child growth standards (*Juergen, Golden & Seaman, 2015*). Weight and height measurements were converted to height-for-age (HAZ), weight-for-age (WAZ), and weight-for-height (WHZ) z-scores, based on WHO reference standards. For WHZ, a z-score <-2 indicated wasting, ≥–2 to <2 indicated normal, and >2 indicated overweight. For HAZ, a z-scores<-2 indicated stunting, and >-2 indicated normal. WAZ z-scores<-2 indicated underweight and >-2 indicated normal. Moderate and severe undernutrition were defined as z-scores <-2 and <-3, respectively (*WHO Multicentre Growth Reference Study Group, 2006*).

Dietary assessment

A 24-h dietary recall questionnaire and 7 days structured food-frequency questionnaires adapted to the local context from Food and Agriculture Organization (FAO) guidelines were used to collect information about dietary practices (*Food & Agricultural Organization, 2010*). The 24-h and 7 days child food frequency recall were computed in to nine food groups: cereals, roots and tubers; vitamin A-rich fruits and vegetables; other fruit; other vegetables; legumes and nuts; meat, poultry and fish; fats and oils; dairy; and eggs. Dietary diversity score calculated by summing the number of food groups based on FAO guidelines, with scores ranging from low (≤3 food items), medium (4–5 food items), and high (≥6 food items).

The 24 h household dietary diversity was computed into twelve food groups: cereals, white tubers and roots, vegetables, fruits, meat, eggs, fish and other seafood, legumes, nuts and seeds, milk and milk products, oils and fats, sweets spices, condiments, and beverages (*Food & Agricultural Organization, 2010*). The Dietary Diversity score was based on the computation of the different food groups consumed and the volume (quantity) of food and nutrient intake were not measured.

The consumption of iron rich food was defined as if a child consumed at least one iron-rich food item among the following food groups: organ meat, flesh meat, or fish at any time in the last 24 h or at least once in the last 7 days preceding the interview.

The household food insecurity status was determined using the 9-component Household Food Insecurity Access Scale validated in Ethiopia (*Gebreyesus et al., 2015*). The HFIAS is composed of nine items, which are asked with a recall period of 1 month. For each item, there was a follow-up of the frequency of the occurrence question. Responses scored 'never' received a score of 0, 'rarely' scored 1, 'sometimes' scored 2, and 'often' scored 3, so that when summed, the lowest possible score was 0 and the highest 27. Household food security was categorized into four levels (food secure, mildly food insecure, moderately food insecure, and severely food insecure) based on the guidelines (*Coates, Anne & Paula, 2007*).

Furthermore, data on child morbidity (diarrhoea and cough for the last 15 days), child immunization, history of hospital admission, iron-fortified food consumption of children, and iron supplementation for the mother and child were collected.

Socioeconomic status

The principal component analysis was used to assess the household wealth index. The presence or absence of each household item was coded as '0' for No and '1' for Yes. Variables included electricity access, as well as ownership of a radio, telephone, television, refrigerator, electric stove, bicycle, motorcycle, car, or computer. Ownership of land, domestic animals, household monthly income, and the number of people in the household, parent education, and occupation were excluded from the model. The first component explained most of the variance in the observed set of variables, and the final model explained 32% of the variance. We then created a wealth index using tertiles that were distributed into three groups: poor, medium, and richest (*Beaumont, 2012*).

Statistical analysis

Data were double-entered and checked using EpiData v.3.1 (EpiData.dk, Odense, Denmark) and transferred to IBM SPSS v.20 (SPSS Inc., Chicago, IL, USA) for analysis. Descriptive statistics were used to summarize categorical variables. Means with 95% confidence intervals (CI), medians, and interquartile range were used to present continuous variables. The bi-variable analysis was done to check the association between each independent variable with the IDA and iron-rich food consumption. All variables with *P*-value < 0.3 were included in the multivariable logistic regression analysis to retain some potential confounding variables. Variables with a *P*-value less than 0.05 were considered statistically significant. Factors associated with consumption of iron-rich food at least once in the last 7 days were determined. Multicollinearity among independent variables was checked and the variables with tolerance test <0.2 and variance inflation factor (VIF) >5.0 were excluded from the models.

The conceptual modelling approach published by Victora et.al was adopted to develop for this specific study which used to determine factors associated with iron deficiency anemia (*Victora et al., 1997*). In the multivariable analysis factors associated with IDA were determined using three-level regression analyses (level one underlying factors, level two intermediate factors, and level three immediate factors). Figure two described variables in the conceptual modelling developed for this study (Fig. S2).

Data quality and control

To assure the quality of the data generated, standard operating procedures were followed during blood sample collection and all laboratory procedures. Structured questionnaires were translated into the local Sidamo and Amharic languages. A 2-day intensive training was given for data collectors and supervisors regarding study objective, questionnaire, interview techniques, anthropometric measurements, blood sample collection, and ethical issues during data collection. A pre-test was done among 17 mother-child-pairs in the nearest village outside of the study area before the actual study. The data collection was checked daily for accuracy, consistency, and completeness by the supervisor. Anthropometry and hemoglobin instruments were calibrated as per standards.

RESULTS

Child characteristics

From the sampled 340 children, 331 participated in the survey (97.4% response rate). Of the 331 children, 172 (52%) were girls, and the mean age was 39.2 (95% CI [38.1–40.6]) months. From the total of 331 children, only 10 (3%) children had received iron supplementation, and two of them were diagnosed with anemia and one was diagnosed with iron deficiency anemia. The children's illness history indicated that in the last 2 weeks preceding the survey, 100 (30%) children had watery diarrhea, and 71 (21%) had a cough. The nutritional status of children indicated that 125 (37%) children were stunted, four (1.3%) were wasted, and 72 (21%) were underweight (Table 1).

Socio-demographic characteristics of mothers and households

A total of 331 mothers participated in the survey and their mean age was 27.7 years (95% CI [27.1–28.3]). The majority of mothers 231 (70%) were women working at home, 75 (22%) had no formal education, and 207 (62%) households were food insecure (Table 2).

Table 1 Demography, morbidity, and nutritional status of children aged 24–59 months in Cheffe Cote Jebessa kebele, southern Ethiopia, 2017.

Variables	Characteristics	Frequency $N = 331$	Percent
Child sex			
	Male	159	48.0
	Female	172	52.0
Child age in months			
	24–36	145	43.8
	37–59	186	56.2
Mother response immunization completed			
	No	25	7.6
	Yes	306	92.4
Immunization completed confirmed in child health card			
	No	28	8.5
	Yes	207	62.5
	No card	96	29.0
Cough lasting 15 days			
	No	260	78.5
	Yes	71	21.5
Diarrhoea lasting 15 days			
	No	231	69.8
	Yes	100	30.2
History of hospital admission			
	No	289	87.3
	Yes	41	12.4
	Missing	1	0.3
Iron-rich food within last 24 h			
	No	281	84.9
	Yes	50	15.1
Iron supplementation			
	No	321	97.0
	Yes	10	3.0
Iron-fortified food			
	No	316	95.5
	Yes	15	4.5
Child dietary diversity score			
	Low	242	73.1
	Medium	65	19.6
	High	24	7.3
Current breast feeding			
	No	229	69.2

Table 1 (continued)

Variables	Characteristics	Frequency $N = 331$	Percent
	Yes	101	30.5
	Missing	1	0.3
Height-for-age score			
	Stunting	125	37.8
	Normal	206	62.2
Weight-for-height score			
	Wasting	4	1.2
	Normal	304	91.8
	Overweight	9	2.7
	Missing	14	4.2
Weight for age score			
	Underweight	72	21.8
	Normal	259	78.2

Note:
Missing value observed under the variable weight for height score was not the actual missing but the result flagged with WHO reference using ENASMART software.

Iron rich food consumption

The 24-h child dietary recall indicated that the mean dietary diversity was 2.8 (95% CI [2.6–3.0]) and the median was 3 (IQR 2–4, range 0–8). Most of the children 242 (73%) scored low on dietary diversity (Table 1), only 50 (15%) children had consumed iron-rich food (meat, organ meat, and fish). The diet of the children contained mostly starchy staples 326 (98.6 %). The 7 days food frequency data showed that 104 (33%) children consumed iron-rich food at least once within the last 7 days (Table 3). Furthermore, household iron-rich food intake was similar to child iron intake of the last 24 h, it showed that only 51 (15%) of the household consumed iron-rich food (Fig. 1).

Magnitude of anemia and inflammation

The mean hemoglobin concentration of the children was 11.6 g/dl (95% CI [11.5–11.7]), and the median was 11.8 g/dl (IQR: [10.9–12.5]). The prevalence of anemia was 107/331 (32.3%). Of the total number of anemic children, only 1 (0.9%) child had severe anemia, 35 (32.7 %) had moderate, and 71 (66.4 %) had mild anemia. The CRP measurements indicated that 23/107 (21 %) of anemic children had some sort of inflammation (Table 4).

Iron deficiency anemia (IDA)

The estimated prevalence rates of IDA varied when using different adjustment methods. It was low in the unadjusted ferritin for inflammation group (18.7%) and, its prevalence ranged from 22% with the exclusion approach, 23% with correction factor approach and 25% with adjustment methods (Table 5).

Table 2 Demographic and socio-economic characteristics of mothers and households, in Cheffe Cote Jebessa kebele, southern Ethiopia, 2017.

Variables	Characteristics	Frequency N = 331	Percent
Mothers' age in years			
	<25	135	40.8
	25–30	133	40.2
	>30	63	19.0
Mother's education			
	No formal education	75	22.7
	1–8 school years	180	54.4
	>8 school years	76	23.0
Father's education			
	No formal education	47	14.3
	1–8 school years	168	51.2
	>8 school years	113	34.5
Mother's occupation			
	Housewife	231	69.8
	Employed	100	30.2
Number of people in household			
	<5 people	222	67.1
	>5 people	109	32.9
Income per month			
	Low	196	59.2
	Medium	117	35.3
	High	18	5.4
Land ownership			
	No	188	56.8
	Yes	143	43.2
Livestock ownership			
	No	188	56.8
	Yes	143	43.2
Wealth index			
	Poor	126	38.1
	Medium	82	24.8
	Rich	123	37.2
Household food insecurity			
	Secure	124	37.5
	Mild	32	9.7
	Moderate	72	21.8
	Severe	103	31.1

Table 3 Twenty-four hour's dietary recall and 7 days food frequency recall.

Food group	24-h recall	Frequency (N = 331)	Percent	1 week-frequency recall	Frequency (N = 331)	Percent
Starchy stable	No	5	1.5	No intake	2	0.6
	Yes	326	98.5	Once	32	9.7
				2–4 times	108	32.6
				≥5 times	189	57.1
Dark green vegetables	No	215	65.0	No intake	101	30.5
	Yes	115	34.7	Once	49	14.8
				2–4 times	145	43.8
				≥5 times	36	10.9
Vitamin A rich fruit and vegetables	No	231	69.8	No intake	90	27.2
	Yes	100	30.2	Once	36	10.9
				2–4 times	120	36.3
				≥5 times	85	25.7
Fruit and vegetables recode	No	230	69.5	No intake	289	87.3
	Yes	101	30.5	Once	25	7.6
				2–4 times	14	4.2
				≥5 times	3	0.9
Organ meat	No	330	99.7	No intake	326	98.5
	Yes	1	0.3	Once	5	1.5
Meat and fish	No	281	84.9	No intake	227	68.6
	Yes	49	14.8	Once	62	18.7
				2–4 per week	37	11.2
				≥5 times	5	1.5
Egg	No	302	91.2	No intake	239	72.2
	Yes	29	8.8	Once	33	10
				2–4 times	54	16.3
				≥5 times	5	1.5
Legume and nuts	No	216	65.3	No intake	149	45
	Yes	115	34.7	Once	46	13.9
				2–4 times	99	29.9
				≥5 times	36	10.9
Milk and milk products	No	236	71.3	No intake	149	45
	Yes	95	28.7	Once	35	10.6
				2–4 times	94	28.4
				≥5 times	53	16

Factors associated with iron deficiency anemia (IDA)

Using higher ferritin cut-off adjustment approach we estimated the Odds Ratios for IDA. The adjusted odds ratio showed that the prevalence of IDA significantly decreased as the height for age z-score increased (AOR 0.7; 0.5–0.9) (Table 6).

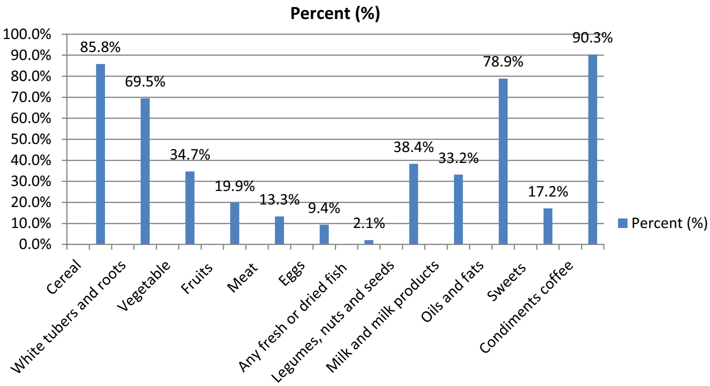

Figure 1 Percentage of household dietary consumption in the last 24 h.
Full-size DOI: 10.7717/peerj.11649/fig-1

Table 4 Magnitude of anemia and inflammation among children aged 2–5 years in Cheffe Cote Jebessa kebele, southern Ethiopia.

Variables	Characteristics	Frequency	Percent
Anemia N = 107/331	Severe	1	0.9
	Moderate	35	32.7
	Mild	71	66.4
Inflammation N = 107	No (CRP < 5mg/L)	84	78.5
	Yes (CRP > 5mg/L)	23	21.5

Table 5 Median (IQR) of ferritin level and prevalence of iron deficiency anemia given different adjustment methods.

Correction method N = 107	Median (IQR) Ferritin (µg/L)	Iron deficiency anemia n (%)	
		No	Yes
Ferritin not adjusted	25.7 (25.7–59.6)	87 (81.3)	20 (18.7)
Exclusion approach	25.2 (12.8–35.3)	65 (77.4)	19 (22.6)
Correction Factor approach	24.5 (12.6–34.6)	82 (76.6)	25 (23.4)
Higher ferritin-cut-off adjustment	25.7 (25.7–59.6)	80 (74.8)	27 (25.2)

Note:
CI, confidence interval; IQR, inter quantile range; µg/L, Micrograms per Litre.

Factor associated with iron-rich food consumption

Iron-rich food consumption at least one time in the last 7 days indicated that mother's education (AOR 1.1; 1.0-1.2) and high household dietary diversity (AOR 1.4; 1.2–1.6) were positively associated with child iron-rich food consumption (Table S3).

Table 6 **Factors associated with iron deficiency anemia among children age 2–5 years in Cheffe Cote Jebessa kebele, southern Ethiopia, 2017.**

N = 107		Iron deficiency anemia		Crude OR (95% CI)	Adjusted OR (95% CI)
		No (%)	Yes (%)		
Inherent factors					
Age (continuous)				1.02 [0.98–1.06]	0.96 [0.91-1.03]
Child sex	Male	29 (36.3)	12 (44.4)	1	1
	Female	51 (63.8)	15 (55.6)	1.07 [0.47–2.47]	0.91 [0.32–2.57]
Underlying factors					
Land ownership	No	55 (68.8)	14 (51.9)	1	1
	Yes	25 (31.3)	13 (48.1)	1.59 [0.69–3.67]	1.91 [0.69–5.30]
Intermediate factors					
Household food security	Food insecure	55 (68.8)	18 (66.7)	1	1
	Food secured	25 (31.3)	9 (33.3)	0.97 [0.41–2.33]	0.76 [0.25–2.28]
Height for age (HAZ) (continuous)				0.86 [0.73–1.00]	**0.74 [0.56–0.98]**
Weight for height (WHZ) (continuous)				0.99 [0.63–1.56]	1.22 [0.73–2.06]
Immediate factors					
Child dietary diversity	Low	61 (76.3)	18 (66.7)	1	1
	Medium and high	19 (23.8)	9 (33.3)	1.60 [0.62–4.16]	1.79 [0.57–5.58]
Meal frequency (continuous)				1.48 [0.81–2.70]	1.80 [0.89–3.62]
Green vegetable consumption per week	0 intake	19 (23.8)	7 (25.9)	1	1
	At least once	61 (76.2)	20 (74.1)	0.63 [0.24–1.64]	0.59 [0.19–1.83]

Note:
Variables with a *P*-value < 0.3 were included in multi-variable analysis. Bold format $P \leq 0.05$; CI, confidence interval; OR, odds ratio.

DISCUSSION

This study has revealed that one-fourth of the anemia cases were due to IDA, and the 24 h food frequency recall showed that only 15% of children consumed iron-rich food and 30% of children consumed iron-rich food at least once in the last week preceding the survey. These findings are comparable with 20% of iron-rich food consumption by children less than 5 years of age within the last 24 h reported in Ethiopia (*Tiruneh et al., 2020*). Iron deficiency anemia was associated with a low height for age of the children, while low iron-rich food consumption was associated with mother's education and household dietary diversity score. In our study, the prevalence of anemia was 32%,which is comparable with the prevalence studies in northern Ethiopia for children in the 6–59 months age group (*Gebreegziabiher, Etana & Niggusie, 2014*).

The 25% prevalence of iron deficiency anemia in our study was higher than the Ethiopian National Micronutrient Survey which estimated a 9% prevalence of iron deficiency anemia among pre-school children in 2016. To our knowledge, there are no earlier community based studies done in Ethiopia on iron deficiency anemia among young children, however, a study from south-western Ethiopia in school children reported a higher prevalence of 37% of iron-deficiency (*Desalegn, Mossie & Gedefaw, 2014*).
The discrepancy may be due to age differences and because the socio-economic and dietary practices could vary across the regions. Studies from Rwanda showed a similar result with

our findings in the age range 6–59 months (*Rutayisire, Nwaike & Marete, 2019*), whereas in Angola 46% of anemia cases were because of iron deficiency in children 3–36 months of age (*Fancony et al., 2020*). Furthermore, a recent review finding indicated that 25% of anemia in young children was because of iron deficiency (*Green et al., 2017*).

We used different inflammation adjustment methods under this study to estimate IDA and we found some variation of the prevalence among these different methods (*World Health Organization, 2020*; *Namaste et al., 2017b*; *Thurnham et al., 2010*). The variation of proportion of iron deficiency anemia with inflammation adjustment could be explained by the high burden of inflammation among children in the study area (*Engle-Stone et al., 2017*; *Merrill et al., 2017*). Furthermore, the study shows that inflammation is one of the causes of iron deficiency anemia (*Fancony et al., 2020*).

In this study improved height-for-age was inversely associated with IDA, and similar findings were reported in children less than 5 years of age from Pakistan (*Habib et al., 2016*). This could be because iron deficiency may lead to delayed growth and development in children. A study from Qatar showed that treatment of IDA with iron supplementation improved the growth of children under 2 years of age (*Soliman et al., 2009*).

In our study iron deficiency anemia was not associated with iron-rich food consumption, however iron-rich food consumption was associated with demographic and socio-economic characteristics of the households. Nevertheless, the lack of association with IDA must be interpreted with care as only 107 cases were involved, and the study may not have been sufficiently powered to detect all risk factors statistically. However, studies from Rwanda and Brazil have also show that socio-demographic characteristics of the household were not associated with iron deficiency anemia (*Nobre et al., 2017*; *Rutayisire, Nwaike & Marete, 2019*). In this study, mothers with better education ate more iron-rich foods, as it is supported by other studies in Ethiopia (*Tiruneh et al., 2020*; *Choi et al., 2011*).

Strength and limitation of the study

One of the strengths of this study is that it represents one of the few studies in Ethiopia that has assessed iron deficiency anemia using a representative sample of a community. The study was done in a farming community, and was a part of a larger study that aimed to evaluate the efficacy of home processed amaranth grain containing bread in the treatment of anemia among two-to-five year-old children in southern Ethiopia (*Orsango et al., 2020*). Even if the area represents a semi-urban part of a town, many of the household members practice agriculture typical of many areas in southern Ethiopia. Furthermore, iron deficiency anemia was assessed based on ferritin concentration adjusted for inflammation. A limitation of the study was that we did not assess potential causes of other types of anemia that could be prevalent in the area. Our sample size calculation was based on the EDHS 2011 report since our proposal development and approval was done before the release of 2016 EDHS report. Also children aged 3–5 years of age were used for sample calculation as EDHS were not reporting on the age group 2–5 years of age. Moreover, it was not possible to know the cause-effect relationship of the factors as it was a cross-sectional study. We used higher ferritin cut-off adjustment approach. Using a higher ferritin–concentration cut-off value for individuals with infection/inflammation

<30 μg/L was preferred to give a conclusion in our study. Because we didn't have the data on AGP concentration and malaria, results have to rely completely on the arithmetic correction factor approach and the regression correction approach.

CONCLUSION

This study found that one-third of the children were anemic, and one fourth of the anemic children had iron deficiency anemia among children 2–5years of age in a semi-urban area of southern Ethiopia. Furthermore, the consumption of iron rich food by children was low, and iron deficiency anemia was linked to low height for age measures. Based on the WHO classification, IDA was a moderate public health problem among our study participants. Thus, both short and long term interventions should be implemented to mitigate the adverse effect of iron deficiency anemia in the study area. Short-term solutions could include food supplementation and fortification strategies, and long-term intervention should focus on improving food diversification, nutritional education, and promoting women's education. Furthermore, emphases should be given to reduce high burden of inflammation in the study area. We recommend further studies on area specific iron deficiency anemia to include AGP and malaria status to see any variation in the prevalence of IDA and study on potential causes of other types of anemia that could be prevalent in the area.

ADDITIONAL INFORMATION AND DECLARATIONS

Funding

This work supported by the Norwegian Programme for Capacity Development in Higher Education and Research for Development (NORHED) and the South Ethiopia Network Universities in public health (SENUPH No. ETH-13/0025). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Grant Disclosures

The following grant information was disclosed by the authors:
NORHED.
SENUPH: ETH-13/0025.

Competing Interests

The authors declare that they have no competing interests.

Author Contributions

- Alemselam Zebdewos Orsango conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, and approved the final draft.
- Wossene Habtu analyzed the data, authored or reviewed drafts of the paper, and approved the final draft.

- Tadesse Lejisa analyzed the data, authored or reviewed drafts of the paper, and approved the final draft.
- Eskindir Loha conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, and approved the final draft.
- Bernt Lindtjørn conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, and approved the final draft.
- Ingunn Marie S. Engebretsen conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, and approved the final draft.

Human Ethics

The following information was supplied relating to ethical approvals (i.e., approving body and any reference numbers):

The institution's ethical board of Hawassa University (IRB/098/08) and the and Regional Ethical Committee West Norway (No. 2016/2034) provided ethical approvals.

Field Study Permissions

The following information was supplied relating to field study approvals (i.e., approving body and any reference numbers):

Hawassa University College of Medicine and Health Science (No. SPH/1471/09).

Data Availability

The following information was supplied regarding data availability:

The raw measurements are available in the Supplemental Files.

Supplemental Information

Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.11649#supplemental-information.

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Paper II

BMJ Paediatrics Open

Co-morbid anaemia and stunting among children 2–5 years old in southern Ethiopia: a community-based cross-sectional study

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To cite: Orsango AZ, Loha E, Lindtjørn B, *et al*. Co-morbid anaemia and stunting among children 2–5 years old in southern Ethiopia: a community-based cross-sectional study. *BMJ Paediatrics Open* 2021;**5**:e001039. doi:10.1136/bmjpo-2021-001039

► Additional supplemental material is published online only. To view, please visit the journal online (http://dx.doi.org/10.1136/bmjpo-2021-001039).

Received 26 January 2021
Accepted 29 May 2021

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ABSTRACT

Background In Ethiopia, 38% of children less than 5 years of age are stunted and 57% are anaemic. Both have a negative impact later in life on physical growth and cognitive development and often coexist. There are few studies in Ethiopia that assessed co-morbid anaemia and stunting (CAS) and context-specific factors associated with it.

Objective The objective of this study was to assess the prevalence of CAS, and factors associated with CAS among children aged 2 to 5 years, in southern Ethiopia.

Methods A community-based cross-sectional survey was conducted among 331 randomly selected children in 2017. Mothers were interviewed using a structured questionnaire to obtain child and household information. Anthropometric measurements and blood samples for haemoglobin were collected. Stunting was defined as height-for-age Z-scores (HAZ) less than −2 SDs and anaemia was defined as altitude-adjusted haemoglobin levels less than 11.0 g/dL. CAS was defined when a child was both stunted and anaemic. Crude and adjusted multinomial logistic regression analyses were used to identify factors associated with CAS.

Results Out of 331 children studied, 17.8% (95% CI 13.87% to 22.4%) had CAS. Factors found significantly linked with higher odds of CAS were increased child age (adjusted OR (AOR) 1.0 (1.0 to 1.1)) and no iron supplementation during the last pregnancy (AOR (95% CI) 2.9 (1.3 to 6.2)). One factor found significantly linked to lower odds of CAS was food secured households (AOR (95% CI) 0.3 (0.1 to 0.9)).

Conclusions Co-morbid anaemia and stunting among children in the study area is of concern; it is associated with household food security, iron supplementation during pregnancy and child age. Therefore, comprehensive interventions focusing on improving household food security and promoting iron supplementation for pregnant women are suggested.

What is known about the subject?

- One in four children 6–24 months of age had anaemia and stunting in Ethiopia based on the Ethiopian Demographic and Health Survey.
- Vitamin A supplementation, consumption of vitamin A–rich fruit and vegetables, meat and legumes, meal frequency, low household wealth, low caregivers' education level, male sex, age, history of infection and small birth size have been found associated with anaemia and stunting in children less than 2 years of age in Ethiopia.

What this study adds?

- One in six children 2–5 years of age in southern Ethiopia had co-morbid anaemia and stunting.
- Food insecurity, child age and iron supplementation during pregnancy were associated with co-morbid anaemia and stunting among children 2–5 years of age.

BACKGROUND

Globally, 22% of children less than 5 years old were stunted in 2017[1] and about 42% were anaemic.[2] According to the Ethiopian Demographic and Health Survey (EDHS) of 2016, 38% of children less than 5 years of age were stunted and 57% were anaemic.[3] When looking only at the age group of 2–5 years, the recent study in Ethiopia reported 33% of prevalence for anaemia[4] and 45% of prevalence for stunting.[5]

Both anaemia and stunting during early life can lead to serious health effects later in life such as increased susceptibility to infections and chronic diseases, impaired cognitive and physical development, and low economic productivity of individuals and the society.[6–8] Chronic anaemia has a negative effect on physical growth during all stages of growth (infancy, childhood and adolescence).[9]

Both anaemia and stunting share common risk factors such as poverty, poor sanitation, large family size, low nutrient intake and infections.[10 11] In recent years, there has been a growing interest to conceptualise and address syndemic health problems including

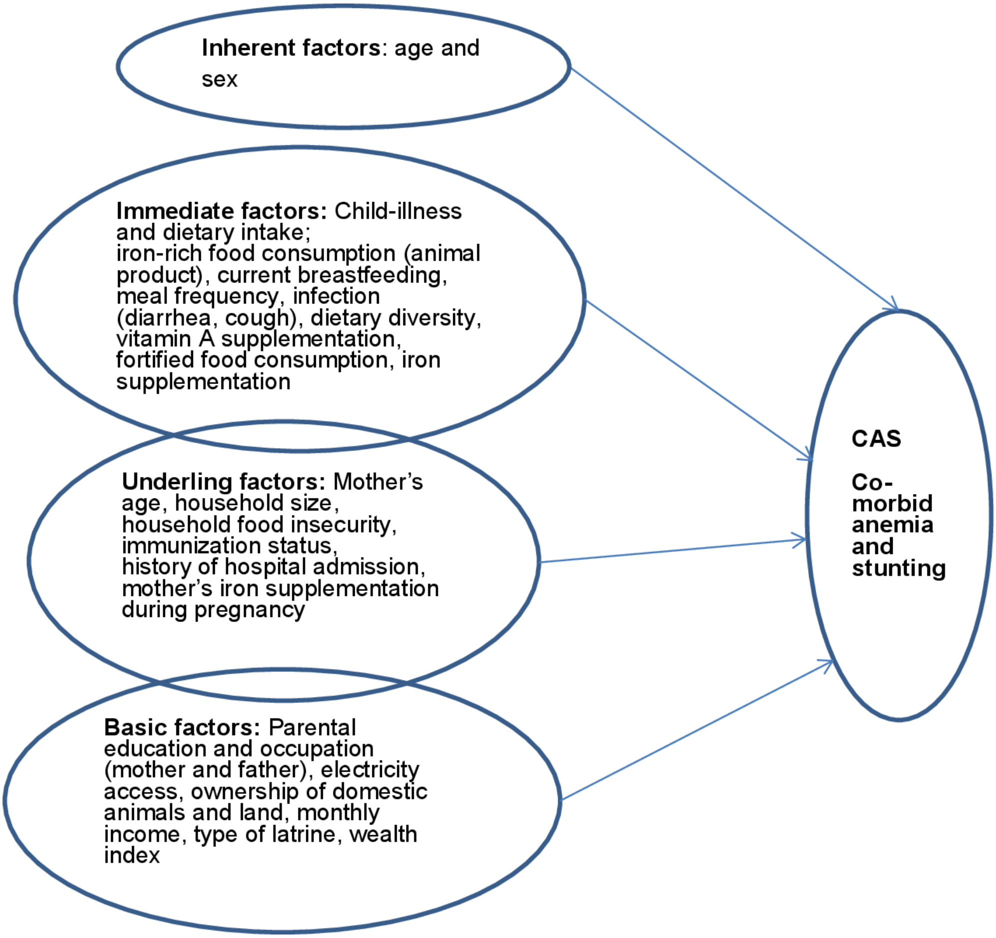

Figure 1 Conceptual framework used in the multivariable regression analysis based on the Unicef framework from 1991.[26] The model is sorting the variables as inherent, immediate, underlying and basic risk factors for co-morbid anaemia and stunting, CAS.

malnutrition.[12 13] In public health nutrition, a syndemic health problem is defined as the aggregation of two or more nutritional health problems in a population with factors that exacerbate the burden of disease.[12] A systematic review of demographic and health survey (DHS) data from 46 low-and-middle-income countries showed that the prevalence of co-morbid anaemia and stunting vary by country and socioeconomic groups.[14] The same study suggests that co-morbid anaemia and stunting should be considered under a syndemic framework, the Childhood Anaemia and Stunting (CHAS). The syndemic approach aggregates anaemia and stunting, and also acknowledges social and environmental factors that exacerbate the occurrence of stunting and anaemia. This approach can be more useful in resource-constrained settings as the prevention strategies focusing on stunting and anaemia differs.[14]

The Ethiopian government is working to reduce child malnutrition by promoting dietary diversity, deworming, iron supplementation during pregnancy, malaria prevention and nutrition education.[15 16] Despite several interventions, both anaemia and stunting remain the largest nutritional problems among children.[3 17] There are several studies in Ethiopia which have investigated the prevalence and determinant factors of either stunting or anaemia.[18–21] However, studies on co-morbid anaemia and stunting are few. The only study reporting national data to our knowledge was based on an analysis of EDHS data that reported 24% prevalence of co-morbid anaemia and stunting in children younger than 2 years.[11] Furthermore, a controversial idea was that a study from Asia reported that anaemia and stunting are independent conditions[22] opposing a systematic review from low-and-middle-income countries recommending that stunting

Table 1 Characteristics of the study children aged 2–5 years in Cheffe Cote Jebessa kebele, southern Ethiopia, 2017

Variables	Total N=331 n (%)
Child age category	
24–36 months	145 (43.8)
36–48 months	116 (35.0)
48–60 months	70 (21.1)
Child's sex	
Boy	159 (48.0)
Girl	172 (52.0)
Current breast feeding	
Yes	101 (30.5)
No	229 (69.2)
Child dietary diversity	
Lowest dietary diversity	242 (73.1)
Medium dietary diversity	65 (19.6)
Higher dietary diversity	24 (7.3)
Meal frequency per day	
<4 times per day	236 (71.3)
≥4 times per day	95 (28.7)
Immunisation completed confirmed in child health card	
No	28 (8.5)
Yes	207 (62.5)
No card	96 (29.0)
Diarrhoea in the past 15 days	
No	231 (69.8)
Yes	100 (30.2)
Cough in the past 15 days	
No	260 (78.5)
Yes	71 (21.5)
Hospital admission since birth	
No	289 (87.3)
Yes	41 (12.4)
Vitamin A supplementation	
No	118 (35.6)
Yes	213 (64.2)
Iron-rich food within the last 24 hours (animal product)	
No	281 (84.9)
Yes	50 (15.1)
Child ever received iron supplementation	
No	321 (97.0)
Yes	10 (3.0)
Child ever received fortified food	
No	316 (95.5)
Yes	15 (4.5)

Note: missing values <1% are not presented.

and anaemia should be considered as a syndemic.[14] Therefore, more research is needed on context-specific findings in terms of co-morbidity and risk factors. This study aimed to assess the prevalence of co-morbid anaemia and stunting and associated factors among children 2–5 years old in southern Ethiopia. Thus, the findings could inform planning of nutrition interventions to mitigate the problems.

METHODS AND MATERIALS

Study area and population

We conducted a cross-sectional study among children aged 2–5 years from 15 February to 30 March 2017 in Cheffe Cote Jebessa kebele (lowest administrative unit in Ethiopia), located in the semi-urban part of Hawassa City, in southern Ethiopia. Hawassa City is located in South Nation, Nationality and People Regional State, 273 km from Addis Ababa, the capital of Ethiopia. According to a census, we conducted before the start of our study, the total population of the village (Kebele) was 23 010 people within 3900 households, and 2180 children were less than 5 years of age. The main crops cultivated in the study area include maize, haricot beans, *inset* and Irish potatoes. Based on our observation during census and survey, the study area (Kebele) is characterised by poor economic status, inadequate health facilities, poor environmental hygiene and sanitation, and inadequate water supplies.[23]

Sample size and sampling procedure

The sample size was calculated using anaemia prevalence because this study was a part of a larger study that aimed to evaluate the efficacy of home-processed amaranth grain containing bread in the treatment of anaemia among 2-to-5-year-old children in southern Ethiopia.[24] Thus, the sample size of 340 was calculated assuming a 33% national average anaemia prevalence among children aged 3–5 years,[17] 95% confidence level and ±5% margin of error. A post hoc sample size calculation done for this study was 281 assuming a 24% CAS prevalence, 95% confidence level and ±5% margin of error.[11] From the list of 1689 children aged 2 to 5 years residing in the study area, 340 were selected by a simple random technique method using IBM SPSS V.20. Participants were eligible if the child was 2 to 5 years of age and residing with their mother who provided informed consent. We selected the age group 2–5 years old for our experimental study, as children would be fed with the prepared bread. We excluded children who were sick or suffering from severe medical conditions.

Study variables

The primary outcome variable of this study was co-morbid anaemia and stunting (CAS), defined when a child was both anaemic and stunted. Secondary outcomes were anyone having either stunting or anaemia. Stunting was defined as height for age (HAZ) z-scores below −2 SD

Table 2 Socioeconomic characteristics of the study household in Cheffe Cote Jebessa kebele, southern Ethiopia, 2017

Variables	Total N=331 n (%)
Mother's age	
<25	135 (40.8)
25–30	133 (40.2)
>30	63 (19.0)
Household family size	
<5	162 (48.9)
≥5	169 (51.1)
Household food security status	
Food secure	124 (37.5)
Mild insecurity	32 (9.7)
Moderate insecurity	72 (21.8)
Severe insecurity	103 (31.1)
Mother's education	
No formal education	75 (22.7)
1–8 years in school	180 (54.4)
>8 years in school	76 (23.0)
Father's education	
No formal education	47 (14.2)
1–8 school years	168 (51.2)
>8 school years	113 (34.5)
Mother's occupation	
Housewife	231 (69.8)
Employed	100 (30.2)
Father's occupation	
Unemployed	29 (8.8)
Employed	302 (91.2)
Income per month	
Monthly income ≤1900 ET Birr	247 (74.6)
Monthly income >1900 ET Birr	84 (25.4)
Wealth index	
Poor	126 (38.1)
Medium	82 (24.8)
Rich	123 (37.2)
Access to electricity	
No	71 (21.5)
Yes	260 (78.5)
Ownership of land	
No	188 (56.8)
Yes	143 (43.2)
Ownership of livestock	
No	188 (56.8)
Yes	143 (43.2)
Latrine type	
Not improved pit latrine	155 (46.8)
Improved pit latrine	175 (52.9)
Last pregnancy iron supplementation	
No	194 (58.6)
Yes	137 (41.4)

Continued

Table 2 Continued

Note: missing values <1% are not presented.

whereas anaemia was defined as altitude adjusted haemoglobin values <11 g/dL.[25] Independent variables were selected based on the conceptual framework from Unicef where the variables were grouped into inherent, immediate, underlying and basic factors.[26] The specific factors in each group are given in the illustration (figure 1).

Data collection

Data were collected from mothers on various demographic, socioeconomic, health and nutrition variables using a structured questionnaire. Seven-day food frequency (FFQ) and 24-hour dietary diversity questionnaires were used to collect information about children's diets. The questionnaires were adapted to the study area based on Food and Agriculture Organization guidelines.[27]

Household food insecurity status was measured by using the Household Food Insecurity Access Scale (HFIAS) developed by the Food and Nutrition Technical Assistant (FANTA) project and validated in Ethiopia by Ghebreyesus *et al*.[28] The HFIAS is composed of nine items, which are asked with a recall period of 1 month. For each item, there was a follow-up of the frequency of the occurrence question. If they experienced the item, they were asked whether they experienced it rarely (once or twice in the past month), sometimes (three to ten times in the past month) or often (more than ten times in the past month). Responses were scored so that 'never' received a score of 0, 'rarely' scored 1, 'sometimes' scored 2 and 'often' scored 3, so that when summed, the lowest possible score was 0 and the highest 27. The household's food security status was categorised into four levels of food security according to the categorisation scheme recommended by the HFIAS Indicator Guide.[29]

Anthropometry and haemoglobin measurements

Height was measured using a Seca 213 portable stadiometer (Seca 213; Seca GmbH, Hamburg, Germany), and the height of the child was read to the nearest 0.1 cm. Weight was taken using a calibrated digital Seca874 scale (Seca 874, Seca GmbH) electronic flat scale. Emergency Nutrition Assessment for SMART software 2011 (Toronto, Canada) was used for analysis of anthropometric data. Weight and height measurements were converted to height-for-age (HAZ), weight-for-age (WAZ) and weight-for-height (WHZ) z-scores, based on WHO reference

Table 3 Magnitude of stunting, anaemia, and co-morbid anaemia and stunting

Variable	Total N=331 n (%)
No anaemia and stunting	158 (47.7)
Anaemia, no stunting	48 (14.5)
Stunting, no anaemia	66 (19.9)
Anaemia and stunting	59 (17.8)

standards. For HAZ, z-scores <−2 indicated stunting and >−2 indicated normal height.[30]

Using vein puncture, 3–5 mL of blood was collected by using a lithium heparin plasma separator test tube to measure haemoglobin. The blood was shaken slowly six to eight times prior to measuring haemoglobin levels, which were determined immediately on-site using a HemoCue analyser 301 (Angelholm, Sweden). The HemoCue analysers were calibrated every morning before the start of data collection. Then haemoglobin was corrected for altitudes, according to WHO standards.[31] Anaemia was defined as altitude-adjusted haemoglobin values<11.0 g/dL.[25]

Socioeconomic status

Principal component analysis was used to assess household wealth index. The presence or absence of each household items such as access to electricity, ownership of a radio, telephone, television, refrigerator, electric stove, bicycle, motorcycle, car or computer were asked and their responses were coded as '0' for No and '1' for Yes. The first component explained 32% variance and sampling adequacy test was 0.77. Finally, the common factor score for each household was produced for grouping households as poor, medium and richest wealth thirtiles.

Statistical analysis

Data were double-entered and checked using EpiData V.3.1 (EpiData.dk, Odense, Denmark), independent variables were selected for multivariable model using R Core Team V.4.1.0 (2021) and analysis was done using IBM SPSS V.20. Descriptive statistics (frequency counts and percentages) were used to summarise categorical variables. Continuous variables were presented using means and 95% CIs, median with IQRs and prevalence estimates were reported along with CIs.

Before doing multinomial analysis, the dependent variable was assumed nominal and mutually exclusive. The dependent variable consists four nominal categories: 'no anaemia or stunting', 'stunting only', 'anaemia only' and 'stunting and anaemia together' (CAS). The 'no anaemia or stunting' category was set as the reference. Complete case analysis was used for missing values to minimise potential bias. Twenty-three potential factors associated with CAS were identified based on a conceptual framework (figure 1), and then 10 variables were selected according to minimum lambda using least absolute shrinkage and selection operator (LASSO) for multinomial logit model; this was implemented using glmnet package of R (online supplemental table 1). Thus, the 10 selected independent variables were analysed with multinomial logistic regression model to identify factors associated to CAS.

RESULTS

Child characteristics

From a total of 340 children selected for the study, 331 (97.4%) participated in the study and 9 (2.6%) of the mothers refused to provide the written consent as the survey involved blood sample collection from their children. Of the 331 children involved in the study, 172 (52%) were girls and the mean age was 39.3 (95% CI 38.0 to 40.5) months. Close to three-fourths of the children, 242 (73%), had the lowest dietary diversity score. About 15 (4.5%) of children consumed fortified food and only 10 (3%) children received iron supplementation (table 1).

Socioeconomic characteristics of the study households

The median age of mothers was 27 years (IQR 24–30). The majority of the mothers, 231 (69.8%), were housewives. More than half, 169 (51%), of the household size was greater than five people per household. Three-quarters of the households had a monthly income which was less than 1900 Birr per month (which is <US$1.90 per day) and 207 (62.5%) households were food insecure (table 2).

Magnitude of CAS)

More than half of the children, 173/331 (52.3%), had either anaemia or stunting. The prevalence of stunting was 125/331 (38%) (95% CI 32.5% to 43.1%), and for anaemia it was 107/331 (32%) (95% CI 27.3% to 37.4%). The overall prevalence of CAS among the study participants was 59/331 (17.8%) (95% CI 13.7% to 21.9%) (table 3).

Factors associated with CAS)

Crude ORs compared for factors were considered in the study and the outcomes are presented in online supplemental table 2. The adjusted multinomial logistic regression showed that child age, iron supplementation during pregnancy and household food security status were associated with CAS.

From the inherent factors, when child age increased by 1 month, it increased the likelihood of the occurrence of CAS (adjusted OR (AOR) 1.0 (1.0 to 1.1)). From the underlying factors, children from a mother who did not take iron supplementation during the last pregnancy were three times more likely to develop CAS (AOR 2.9 (1.3 to 6.2)) compared with those who took iron supplementation during last pregnancy. Further, CAS was less common in children who lived in food secure households (AOR 0.3 (0.1 to 0.9)) as compared with children who lived in food insecure households (table 4).

Table 4 Factors associated with CAS, anaemia and stunting among the study participants in Cheffe Cote Jebessa kebele, southern Ethiopia, 2017

	Anaemia		Stunting		CAS	
	COR (CI)	**AOR (CI)**	**COR (CI)**	**AOR (CI)**	**COR (CI)**	**AOR (CI)**
Inherent factor						
Child sex						
Boy	0.6 (0.3 to 1.1)	0.6 (0.3 to 1.2)	1.4 (0.8 to 2.5)	1.5 (0.7 to 2.9)	0.6 (0.3 to 1.2)	0.6 (0.3 to 1.3)
Child age	0.9 (0.9 to 1.0)	0.9 (0.9 to 1.0)	1.1 (1.0 to 1.1)	1.1 (1.0 to 1.1)	1.1 (1.0 to 1.1)	1.0 (1.0 to 1.1)*
Basic factor						
Mother education	0.9 (0.8 to 1.0)*	0.9 (0.8 to 1.1)	0.8 (0.8 to 0.9)*	0.9 (0.8 to 1.1)	0.8 (0.8 to 0.9)*	0.9 (0.8 to 1.1)
Father education	0.9 (0.8 to 1.0)	1.0 (0.8 to 1.1)	0.8 (0.8 to 0.9)*	0.8 (0.7 to 1.0)	0.9 (0.8 to 0.9)*	0.9 (0.8 to 1.1)
Latrine						
Not improved pit latrine	0.8 (0.4 to 1.6)	0.7 (0.3 to 1.6)	1.2 (0.7 to 2.2)	0.8 (0.4 to 1.7)	2.1 (1.1 to 4.0)	1.5 (0.6 to 3.2)
Improved pit latrine	1	1	1	1	1	1
Underlying factor						
Household family size	1.1 (0.9 to 1.3)	1.1 (0.8 to 1.3)	1.1 (0.9 to 1.3)	0.9 (0.7 to 1.2)	1.3 (1.1 to 1.5)	1.1 (0.9 to 1.4)
Meal frequency	1.2 (0.7 to 1.9)	1.3 (0.8 to 2.0)	0.7 (0.4 to 1.0)	0.6 (0.3 to 0.9)	0.7 (0.5 to 1.1)	0.8 (0.4 to 1.4)
Household food security status						
Food secure	0.7 (0.3 to 1.4)	0.5 (0.2 to 1.3)	0.6 (0.3 to 1.2)	0.7 (0.2 to 1.8)	0.4 (0.2 to 0.8)*	0.3 (0.1 to 0.9)*
Mild and moderate food insecurity	0.6 (0.2 to 1.4)	0.5 (0.2 to 1.3)	0.8 (0.4 to 1.7)	1.0 (0.4 to 2.5)	0.8 (0.4 to 1.6)	0.6 (0.2 to 1.5)
Severe food insecurity	1	1	1	1	1	1
Last pregnancy iron supplementation						
No	3.1 (1.5 to 6.5)*	3.2 (1.5 to 6.8)	1.2 (0.7 to 2.1)	1.0 (0.5 to 2.0)	3.7 (1.8 to 7.4)*	2.9 (1.3 to 6.2)*
Immediate factor						
Current breast feeding						
No	0.6 (0.3 to 1.2)	0.7 (0.3 to 1.6)	0.3 (0.5 to 6.2)	1.1 (0.4 to 2.7)	5.1 (2.1 to 12.6)*	2.0 (0.6 to 5.9)

*p<0.05.
AOR, adjusted odds ratio; CAS, co-morbid anaemia and stunting; COR, crude odds ratio.

DISCUSSION

This study described that almost one-fifth of children were affected by CAS. It was associated with child age, father's educational status, iron supplementation during pregnancy and household food security status.

The 18% prevalence of CAS was lower than the study done in Ethiopia among younger children 6 to 23 months of age that reported a 24% prevalence of concurrent anaemia and stunting.[11] This variation may be due to the different age groups and the use of national demographic health data. In the DHS data analysis from different low-income countries, the national prevalences of co-morbid stunting and anaemia among children aged 6–59 months were 25% in Ethiopia, 22% in Gabon and 25% in Congo.[14]

Stunting, with 38% prevalence, is categorised as a severe nutritional public health problem in the study area. Our finding is comparable with the study done in Ethiopia reporting 41% of stunting among children under 5 years of age.[32] Furthermore, an anaemia prevalence of 32% is categorised as a moderate public health problem of children aged 2–5 years in the study area. This finding is similar to a study from northern Ethiopia finding a prevalence of 37% in children less than 5 years of age.[19]

We found that children living in food secured household had lower odds of CAS than those in food insecure households. A higher anaemia prevalence[20] and stunting prevalence[33] in food insecure households have been reported previously in Ethiopia. Food insecure households could have limited access to diversified food and had lower meal frequency than food secure households. The study done in Ethiopia among children showed that low meal frequency significantly increases the co-occurrence of stunting and anaemia.[11] In our study, meal frequency was not associated with CAS but food insecurity may have had a relationship with meal frequency. Similarly, studies conducted in Ethiopia showed that stunting was high among children who had low meal frequency, less than four times per day, compared with those who had more frequent meals.[34 35] The explanation could be that children who get frequent meals, whatever its content would be, probably get more nutrients. This is supported by the study undertaken in Ethiopia which reported inadequate energy intake and low-quality protein consumption as the main cause of stunting.[36] Low meal frequency and low energy intake could be aggravated by household food insecurity.

In this study, we found that children born to mothers who took iron supplements during their last pregnancy were less likely to develop CAS than those who had mothers who did not get supplements during pregnancy. We have not found other studies that have investigated the relationship between iron supplementation during pregnancy and CAS in children. However, some studies report on low iron intake of mothers and having a mother with symptoms of anaemia as predictors for anaemia in children less than 5 years of age.[10 37]

In this study, increased child age had higher odds of CAS than those in the lowed age. However, we could not find similar studies supporting these findings related with CAS, but there are studies which showed similar results for stunting.[38] This may be due to growth impairment becoming more severe with time. Anaemia has shown various associations with age as multiple issues including the mother's nutritional status, breast feeding and diet, genetic issues and co-morbidity may be closely related to it.[10]

Strength and limitation

Our study is one of few studies done in Ethiopia presenting CAS and contextually relevant co-factors. The study used a representative sample using a simple random sampling technique at a community level. However, the study area was selected purposively since this study was a part of a larger study that aimed to evaluate the efficacy of home-processed amaranth grain containing bread in the treatment of anaemia among 2-to-5-year-old children in southern Ethiopia.[24] Also, our sample size calculation based on the EDHS 2011 report since our proposal development and approval was before the release of the 2016 EDHS report. Furthermore, children aged 3–5 years were used for sample size calculation as EDHS was not reporting on the age group 2–5 years. Moreover, we used validated questionnaires, and standard and calibrated haemoglobin, height and weight measurements. As this study used a cross-sectional design, caution should be exercised in evaluating cause–effect relationships.

CONCLUSION

In conclusion, our study provided a concerning high-level co-morbid anaemia and stunting among children in the study area, which was associated with household food security status, child age and mother's iron supplementation during pregnancy. The high prevalence of anaemia and stunting at the same time indicates that it is necessary to tackle both burdens simultaneously. Policy-makers should promote and strengthen the implementation of the iron supplementation during pregnancy as a short-term implementation strategy and improving household food security as a long-term implementation strategy to treat children with CAS.

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Acknowledgements The authors are grateful to Mr. Damte Data Daba, field coordinators, all data collectors, Cheffe Cote Jebessa Health Centre, all health post workers, Cheffe Cote Jebessa community leaders and all study participants.

Contributors AZO conceived the research idea, implemented the study, collected the data, performed the statistical analyses and drafted the manuscript. EL, BL and IMSE participated in developing the research idea, proposal review, data analysis and critical review of the draft manuscript. IMSE also participated in the training of the data collectors and the evaluation of the pre-test. All authors approved the final version of the manuscript.

Funding The funding body of this research was NORHED/SENUPH (Norwegian Program for Capacity Development in Higher Education and Research for Development/South Ethiopia Network of Universities in Public Health).

Disclaimer The role of the funding body was covering the administrative cost of the research but had no role in the design of the study and collection, analysis, and interpretation of the data, and writing the manuscript.

Authors declared that there is no computing interest. Authors declared that there is no computing interest.

Patient and public involvement statement Patients or the public were not involved in the design, conduct, reporting and dissemination plans of our research.

Patient consent for publication Not required.

Ethics approval The institution ethical board of Hawassa University (IRB/098/08) and the Regional Ethical Committee West of Norway (No. 2016/2034) provided ethical approval. Local administrative and health post authorities granted official permissions to undertake the study. Informed written consent was obtained from mothers for the study participants.

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement Data are available on reasonable request. Also, data can be available on reasonable request from the corresponding author AZO (zalemselam@yahoo.com; ORCID: https://orcid.org/0000-0001-8300-2412?lang=enhttps://orcid.org/0000-0001-8300-2412?lang=en).

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Paper II Supplementary Table: Multinomial logistic regression crud odds ratio and p-value

	Anaemia		Stunting		CAS	
Child sex	**COR**	**p-value**	**COR**	**p-value**	**COR**	**p-value**
Boy	0.6 (0.3-1.2)	.131	1.4 (0.8-2.5)	.215	0.6 (0.3-1.2)	.149
Girl	1		1		1	
Child age						
24-36 month	1.9 (0.5-7.1)	.300	0.1 (0.0-0.2)	.000	0.1 (0.0-0.2)	.000
36-48 month	2.2 (0.6-8.4)	.249	0.5 (0.2-1.0)	.068	0.4 (0.2-0.9)	.035
48-60 months	1		1		1	
Current Breast feeding						
No	0.6 (0.3-1.2)	.139	3.0 (1.4-6.2)	.003	5.4 (2.2-13.3)	.000
Yes	1		1		1	
Child Dietary Diversity score						
Lowest dietary diversity	0.9 (0.3-3.1)	.920	1.8 (0.5-6.8)	.348	1.3 (0.4-4.2)	.650
Medium dietary diversity	1.1 (0.3-4.3)	.832	1.9 (0.5-7.9)	.349	0.9 (0.2-3.6)	.932
Higher dietary diversity	1		1		1	
Meal frequency						
<4 times per day	0.6 (0.3-1.2)	.146	2.4 (1.1-4.9)	.019	1.8 (0.9-3.8)	.088
>/=4 times per day	1		1		1	
Vaccine completed						
No	3.5 (0.9-12.8)	.053	3.6 (1.1-11.8)	.033	4.7 (1.5-15.3)	.008
Yes	1		1		1	
Diarrhea in the past 15 days						
No	0.9 (0.5-1.9)	.842	0.9 (0.5-1.8)	.934	0.9 (0.5-1.8)	.913
Yes	1		1		1	
Cough in the past 15 days						
No	1.1 (0.5-2.4)	.846	0.9 (0.5-1.9)	.925	1.2 (0.6-2.6)	.574
Yes	1		1		1	
Hospital admission since birth						
No	1.9 (0.6-5.7)	.264	1.1 (0.5-2.5)	.844	1.8 (0.7-5.1)	.234
Yes	1		1		1	
Vitamin -A supplementation						
No	1.6 (0.8-3.1)	.171	1.2 (0.6-2.1)	.619	1.1 (0.6-2.1)	.710
Yes	1		1		1	
Household family size						
<5 peoples	0.7 (0.3-1.3)	.239	0.5 (0.3-0.9)	.039	0.4 (0.2-0.7)	.002
≥5 peoples	1		1		1	
Household food security						
Household food secure	0.7 (0.3-1.4)	.309	0.6 (0.3-1.2)	.161	0.4 (0.2-0.8)	.016

Mild and moderate food insecurity	0.6 (0.2-1.4)	.223	0.8 (0.4-1.7)	.628	0.7 (0.4-1.5)	.465
Severe food insecurity	1		1		1	
Mother education status						
No formal education	1.4 (0.5-3.6)	.458	2.0 (0.8-4.8)	.117	3.1 (1.2-7.6)	.015
1-8 school years	0.7 (0.3-1.6)	.507	1.2 (0.6-2.5)	.620	1.2 (0.5-2.8)	.574
> 8 school years	1		1		1	
Father education status						
No formal education	1.5 (0.5-4.8)	.500	1.7 (0.7-4.0)	.215	2.9 (1.1-8.2)	.038
1-8 school years	2.9 (1.4-6.3)	.005	1.4 (0.7-2.6)	.334	3.7 (1.7-8.1)	.001
> 8 school years	1		1		1	
Mother Occupation						
Housewife	1.2 (0.6-2.5)	.571	1.4 (0.7-2.7)	.253	1.3 (0.7-2.6)	.367
Employed	1		1		1	
Father occupation						
Un employed	1.3 (0.4-3.8)	.639	1.5 (0.6-3.9)	.365	0.6 (0.1-2.2)	.435
Employed	1		1		1	
Monthly income						
</=1900 ETB	0.9 (0.5-2.0)	.945	0.9 (0.5-1.7)	.735	1.7 (0.8-3.8)	.143
>1900 ETB	1		1		1	
Electricity access						
No	1.1 (0.5-2.3)	.837	1.4 (0.7-2.6)	.353	0.6 (0.2-1.3)	.191
Yes	1		1		1	
Ownership of land						
No	1.7 (0.8-3.4)	.127	0.6 (0.3-1.1)	.138	1.2 (0.6-2.2)	.534
Yes	1		1		1	
Ownership of livestock						
No	1.0 (0.5-2.0)	.866	0.6 (0.4-1.2)	.172	1.5 (0.8-2.7)	.224
Yes	1		1		1	
Latrine						
Not improved pit latrine	0.8 (0.4-1.6)	.671	1.2 (0.7-2.2)	.455	2.2 (1.2-4.1)	.011
Improved pit latrine	1		1		1	
Water source						
Public water	0.5 (0.1-1.9)	.361	1.4 (0.6-3.4)	.360	1.7 (0.7-3.9)	.223
Piped in to dwelling	1		1		1	
Iron supplementation during last pregnancy						
No	3.1 (1.5-6.5)	.002	1.2 (0.6-2.1)	.558	3.7 (1.8-7.4)	.000
Yes	1		1		1	

Paper III

PLOS ONE

RESEARCH ARTICLE

Efficacy of processed amaranth-containing bread compared to maize bread on hemoglobin, anemia and iron deficiency anemia prevalence among two-to-five year-old anemic children in Southern Ethiopia: A cluster randomized controlled trial

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OPEN ACCESS

Citation: Orsango AZ, Loha E, Lindtjørn B, Engebretsen IMS (2020) Efficacy of processed amaranth-containing bread compared to maize bread on hemoglobin, anemia and iron deficiency anemia prevalence among two-to-five year-old anemic children in Southern Ethiopia: A cluster randomized controlled trial. PLoS ONE 15(9): e0239192. https://doi.org/10.1371/journal.pone.0239192

Editor: Timothy J. Green, South Australian Health and Medical Research Institute, AUSTRALIA

Received: December 5, 2019

Accepted: September 1, 2020

Published: September 28, 2020

Peer Review History: PLOS recognizes the benefits of transparency in the peer review process; therefore, we enable the publication of all of the content of peer review and author responses alongside final, published articles. The editorial history of this article is available here: https://doi.org/10.1371/journal.pone.0239192

Abstract

Background

Few studies have evaluated iron-rich plant-based foods, such as amaranth grain, to reduce anemia and iron deficiency anemia. Amaranth is rich in nutrients, but with high level of phytate. The objective of this trial was to evaluate the efficacy of home processed amaranth grain containing bread in the treatment of anemia, hemoglobin concentration and iron deficiency anemia among two-to-five year-old children in Southern Ethiopia.

Method

Children with anemia (hemoglobin concentration <110.0g/L) (N = 100) were identified by random sampling and enrolled in a 1:1 cluster randomized controlled trial for six months in 2017. The amaranth group (N = 50), received 150g bread containing 70% amaranth and 30% chickpea, the amaranth grain was processed at home (soaking, germinating, and fermenting) to decrease the phytate level. The maize group (N = 50), received 150g bread, containing processed maize (roasted and fermented) to give a similar color and structure with amaranth bread. Hemoglobin, ferritin, and CRP were measured at baseline and at the end of intervention. Hemoglobin and ferritin values were adjusted for altitude and infection, respectively. Generalized estimating equation and generalized linear model were used to analyze the data.

Result

In the last follow-up measure anemia prevalence was significantly lower in the amaranth group (32%) as compared with the maize group (56%) [adjusted risk ratios, aRR: 0.39 (95% CI: 0.16–0.77)]. Hemoglobin concentration estimate of beta coefficient was significantly

Data Availability Statement: All relevant data are uploaded to the OSF database and publicly accessible via the following URL: https://doi.org/10.17605/OSF.IO/EKUWZ.

Funding: Norwegian Program for Capacity Development in Higher Education and Research for Development/ South Ethiopia Network of Universities in Public Health (NORHED/SENUPH) provided funding for this study in the form of a grant awarded to BL (ETH-13/0025). The funders had no role in study design, data collection and analysis, decision to publish or preparation of the manuscript.

Competing interests: The authors have declared that no competing interests exist.

higher in the amaranth group compared with the maize group [aβ 8.9g/L (95%CI: 3.5–14.3)], p-value <0.01. The risk of iron deficiency anemia is significantly lower in the amaranth group [aRR: 0.44 (95%CI: 0.23–0.83)] in the intention to treat analysis but not significant in the complete case analysis. There was no significant difference between groups in iron deficiency [aRR: 0.81 (95%CI: 0.55–1.19)].

Conclusion

Processed amaranth bread had favorable effects on hemoglobin concentration and has the potential to minimize anemia prevalence.

Clinical trial registration

Trial registry number: PACTR201705002283263 https://pactr.samrc.ac.za/TrialDisplay.aspx?TrialID=2283

Introduction

Chronic malnutrition is a leading health problem among children under five years of age in Ethiopia [1]. Anemia is a chronic type of malnutrition and a major contributor to childhood morbidity and mortality, limited physical and cognitive development, and decreased productivity later in life. In Ethiopia, 57% of children under five years old suffer from anemia [2]. The major cause of anemia is inadequate intake of micronutrients, mainly iron containing foods. These nutrients are largely found in animal-source food, which is generally unaffordable for families living in poverty. The staple food comprising the vast amount of energy in the diet of Ethiopians is dominated by starchy cereals, which are low in proteins, minerals, and vitamins [2].

Studies indicate that the amaranth plant has the potential to reduce food insecurity and resultant undernutrition [3,4]. Amaranth is a pseudo-cereal with high grain yield and capable of withstanding extreme climate and soil conditions. Amaranth also possesses good nutritional qualities with a high level of protein, minerals, and fat as compared to commonly utilized cereals, such as maize. Amaranth grain contains 16/100 grams (g) protein, 173 milligrams (mg)/100 g calcium, 35 mg/100 g iron, 3 mg/100 g zinc, and higher potassium, phosphorous, magnesium, manganese, vitamins A and E, and folic acid levels than cereal grains [5,6]. Consequently, it can constitute a cheaper alternative for poor households to address nutritional anemia.

Even though amaranth contains quality amino-acids, high iron and other micro-nutrients, it also has a high concentration of phytic acid that can reduce the bio-availability of nutrients, especially iron, protein, and zinc [6,7]. One study from Kenya indicated that amaranth has no significant effect on the anemia and iron status of children, and this could be due to a high iron to phytate molar ratio [6]. However, prior to this study, research had been performed on methods to decrease the phytate level of amaranth grain by utilizing home processing, such as soaking, germinating, and fermenting, using locally available materials to create acidic media. The research identified methods to decrease phytate level to an acceptable level of iron to phytate molar ratio of less than one [8,9] but its effectiveness has not yet been investigated *in vitro*. Thus, the present study aimed to evaluate the efficacy of processed amaranth grain compared to the commonly consumed maize on hemoglobin concentration and on treatment of anemia prevalence among two-to-five year-old children in Southern Ethiopia.

Methods

Ethics approval and consent to participate

The Institutional Review Board of Hawassa University (IRB/098/08) provided ethical approval on September 06, 2016 and the Regional Ethical Committee West of Norway (No. 2016/2034) on December 15, 2016. Local administrative and health post authorities also granted official permissions for the study. Informed written consent was obtained from the mothers. The trial was registered (Pan African Clinical Trials Registry number: PACTR201705002283263). The authors confirm that all related trials for this food were registered.

The justification for delayed trial registration was a delay we faced when trying to register it at ClinicalTrial.gov. After getting the ethical approval, we immediately applied for the registration of the Clinical Trials on February 6, 2017, to ClinicalTrial.gov. Their response was unfortunately delayed, and we therefore dropped it on May 11, 2017. Then, we applied to Pan African Clinical Trial Registry (PACTR) on May 11, 2017 and shortly afterwards; we got the trial registration on May 19, 2017. During the time, we started the survey to maintain the study schedule. After the survey, we identified children with anaemia which required immediate interventions as per the ethical approval agreement.

Protocol available

Protocol will be available after publication at: https://dx.doi.org/10.17504/protocols.io.bhzbj72n.

Study area

The study was carried out in the Cheffe Cotte Jebessa kebele (lower administrative unit), in the suburban parts of Hawassa city, in southern Ethiopia. The altitude of the study area ranges from 1708 to 1920 meters above sea level. The annual average temperature is 12–27˚C with an average rainfall of 500 to 1,800 millimeter. The main crops cultivated in the area include maize, haricot beans, *enset*, and Irish potatoes. Amaranth grows as a wild crop in the study area, and its grains and leaves are used as food. Furthermore, the grain part is utilized for both medicinal purposes and alcoholic beverage production in the study area and other parts of the country.

Study design

The study design was a community-based food intervention with a 1:1 cluster randomized controlled trial.

Sample size

Prior to the trial, a census was conducted and 1689 children two-to-five year-old were identified in the study area. By using simple random sampling we selected 340 children and a cross-sectional study carried out to identify children with mild and moderate anemia. The sample size of 340 was estimated to identify anemic children, assuming a 33% mild and moderate anemia prevalence in South Ethiopia, precision of 0.5, and 95% confidence intervals. The sample size of the trial was calculated considering a mean difference of 0.7 in hemoglobin level [10], 1 standard deviation, power of 90%, alpha level of 0.05 with two sided test and an attrition rate of 12%. The total sample size of the trial was 100, with 50 children in each arm.

We conducted the survey from February 15, to March 30, 2017 and identified 100 children with anemia. The identified children were sparsely distributed in the study area and thus, we clustered them in to eight clusters by their geographic locations to ease the distribution of breads and for close follow-up. The eight clusters were randomly allocated into four clusters in the maize arm and four clusters in the amaranth arm, and each arm had 50 children. The random allocation was done by external body without knowing about the cluster. Then, the intervention (feeding of breads) was started for both arms on April 14, 2017 and continued for the next six months.

Inclusion and exclusion criteria

Caregiver-child pairs were included in the study, in which the mother was the targeted respondent. The children included in the study were two-to-five year-olds with hemoglobin levels from 70 to <110 g/L, who lived in the area, and planned to live in the area for the next one year. Children were excluded if they were living with chronic illnesses, such as HIV/AIDS, tuberculosis, or cancer. Children who were taking iron supplements, received a blood transfusion in the last six months, had repeated malaria at least three times in the last three months, or were unwilling to participate in the study were also excluded.

Outcome variables

The primary outcome was hemoglobin concentration and anemia prevalence, and the secondary outcome was iron deficiency anemia and iron deficiency.

Intervention

In both groups, the intervention and the control, 150 g bread was provided to the children under the researcher team's supervision on a daily basis for the period of six months. The children in the experimental and control groups labelled 'amaranth group' and 'maize group', respectively. The amaranth group was supplied with bread containing 70 mass % amaranth grain and 30 mass % mashed chickpea. The maize group was supplied with the same size of bread containing 100% maize. All participants in the amaranth and maize groups were treated with albendazole 400 mg single dose, irrespective of their last dosage prior to the start of the feeding intervention and at the end of feeding intervention.

Recipe preparation

Home level processing was applied to the amaranth grain to reduce phytate level. Amaranth grain was soaked in water by adding 5 ml of lemon juice per 100 ml of water for 24 hours and germinated for 72 hours. After sun drying, it was roasted and milled with a local electrical mill then fermented bread was prepared [9]. At the same time, maize grain was also roasted and fermented in order to make the recipe similar in color and structure with amaranth bread. The recipe was prepared based on the recommended dietary allowance (RDA); according to RDA, 150 g of bread (70% amaranth with 30% chickpea) had 22.3 mg of iron contained in the bread, which can fulfill 50% of RDA considering 15–20% iron absorption. The acceptability of this combination was tested in the community in the previous study and it was acceptable [9,10] "Table 1".

Distribution and masking

The study participant, bread distributer, and data collector were uninformed about arm allocation and the content of the bread. Both grains, amaranth and maize, were processed (roasted

Table 1. The nutrient content of amaranth and maize bread.

100 g	Protein mg/100g	Fat mg/100g	Iron mg/100g	Calcium mg/100g	Zinc mg/100g	Energy Kcal
100% maize	7.58	4.44	4.31	24.82	2.47	386
70% amaranth and 30% chickpea	16.39	7.08	15.16	244.71	2.59	404

mg, milligram; g, gram; Kcal, kilocalorie.

https://doi.org/10.1371/journal.pone.0239192.t001

and fermented) to make the bread similar in color and structure. The bread distributers were different for the amaranth and maize arms, and there were eight caregivers, one for each cluster. Eight boxes, one for each cluster, were packed and labeled with the respective caregiver's name. Each caregiver feed the child at their home every day under her direct supervision. In case of refusals or absenteeism, unopened bread was returned and registered at coordinator level each day.

Blood collection and laboratory methods

Three to five milliliter (ml) of venous blood was collected in test tubes with lithium heparin plasma to measure hemoglobin, serum ferritin, and CRP at the scheduled baseline and last follow-up visits. Hemoglobin concentration was determined immediately using a HemoCue analyser 301 (Angelholm, Sweden). All samples with hemoglobin <110 g/L were tested further for serum-ferritin and CRP. Anemia was classified based on altitude-adjusted hemoglobin concentration as normal (≥110.0 g/L), mild (100.0–109.0 g/L), moderate (70.0–99.0 g/L), or severe (<70.0 g/L) [11,12]. Children with severe anemia (<70.0 g/L) identified in the survey were excluded from the trial, as they were referred for medical treatment (N = 1). Hemoglobin concentrations were corrected for altitudes, according to WHO standards [11,12].

Serum ferritin was analyzed using the Cobas 6000 e601 and CRP using the Cobas 6000 c501 module, both from Roche (Germany). Ferritin was adjusted for inflammation based on CRP measure. The inflammatory state of each individual was classified as "healthy" if CRP<5mg/L and "with inflammation" if CRP >5mg/L. The cut-off value for ferritin level was set at <12 μg/L for those children who had CRP<5mg/L (healthy) and <30 μg/L for those children who had CRP >5mg/L (inflammation). Iron deficiency defined as ferritin concentration <12 μg/L for healthy children and <30 μg/L for children with inflammation [13,14]. Iron deficiency anemia was defined as anemia with concurrent iron deficiency [14,15].

Anthropometry measurements

Anthropometric weight and height were measured to assess nutritional status. Height was measured using a Seca 213 height board (Hamburg, Germany) with a sliding head piece while the child stood straight. Weight was taken using a calibrated Seca 874 electronic flat scale with the child barefoot and wearing light clothing. Anthropometry measurements were analysed according to the Emergency Nutrition Assessment for SMART software 2011 (Toronto, Canada), developed using WHO child growth standards [16]. Weight and height measurements were converted to height-for-age (HAZ), weight-for-age (WAZ), and weight-for-height (WHZ) z-scores, based on WHO reference standards. For WHZ, a z-score <-2 indicated wasting, ≥- 2 to <2 indicated normal, and >2 indicated overweight. For HAZ, a z-score <-2 indicated stunting, and >-2 indicated normal height. WAZ scores <-2 indicated underweight, and >-2 indicated normal. Moderate and severe undernutrition were defined as z-scores <-2 and <-3, respectively [17].

Questionnaire

A 7 days structured food-frequency questionnaire (FFQ) and 24-hour dietary diversity questionnaire were used to collect information about children's diets. The foods were counted in nine food groups (items), in which the food-frequency items were created based on Food and Agriculture Organization (FAO) guidelines. The scores ranged from low (<3 food items), medium (4–5 food items), to high (≥6 food items) [18]. Dietary diversity and iron rich food (red meat, organ meat and fish) 7 days frequency was assessed every month to control confounding. The household food diversity scale was categorized into 12 food types, according to the Food and Nutrition Technical III project, and the answers were scored the same as for the food-frequency items [18]. We used a household food insecurity measure validated in Ethiopia by Gebreyesus [19]. The prevalence of household food insecurity was determined using the nine-component Household Food Insecurity Access Scale (HFIAS). Based on component scores (1 = rarely, 2 = sometimes, 3 = often), household food insecurity was classified as secure, mild, moderate, or severe [20].

Statistical analysis

Data were double-entered and checked using EpiData v. 3.1 (Odense, Denmark), and transferred to IBM SPSS v. 20 (Chicago, IL, U.S.A.) and STATA 15 for analysis [21]. Descriptive statistics, including frequency counts and percentages, were used to summarize the data and chi-square test was used to compare the distribution of categorical variables in the amaranth and maize groups. Means, medians, confidence intervals (CI), and inter quartile ranges (IQR) were used to summarize continuous variables. Before multivariate analysis was performed, we checked for baseline differences of anemia, hemoglobin, and ferritin distributions between the study groups. Intra class correlation coefficients (ICC) was checked prior to running the multivariate analysis among the groups' 1:1 randomly allocated eight clusters: four clusters in the amaranth group and four clusters in the maize group. There was no significant intra class correlation for anemia, hemoglobin, and ferritin among group clusters (S1 Table).

Intention-to-treat analysis and complete case analysis were used to manage the missing value of main variables: hemoglobin, ferritin, and CRP (anemia, Iron deficiency anemia and iron deficiency). Intention-to-treat analysis (ITT) applied by replacing the missing values of the last follow-up measure from the baseline and a complete-case analysis was restricted to available information at the last follow-up measure was used to decrease miss-interpretation. A bivariate and multivariate generalized liner model for binomial family was used for anemia prevalence. Generalized estimating equations (GEE) procedure was used for analysis of repeated measurements (hemoglobin, ferritin and CRP) once at baseline and once at last follow up. Hemoglobin was not normally distributed and included as continuous variable. Adjusted ferritin with CRP was used to categorize iron deficiency anemia and iron deficiency so that it was included as categorical variable. Quasi likelihood under independency model criteria (QIC) with the lowest value was used to select correlation matrix. Child age, child sex, dietary diversity and stunting were included in the multivariate analysis. The estimate of beta coefficient with confidence interval was reported for continuous variables and relative risk ratio was presented for the categorical variables with confidence interval.

Results

Trial participants

Out of the 100 children in the trial, 82 children remained in the trial cohort at the end of the scheduled visits, 41children in the amaranth group and 41children in the maize group "Fig 1".

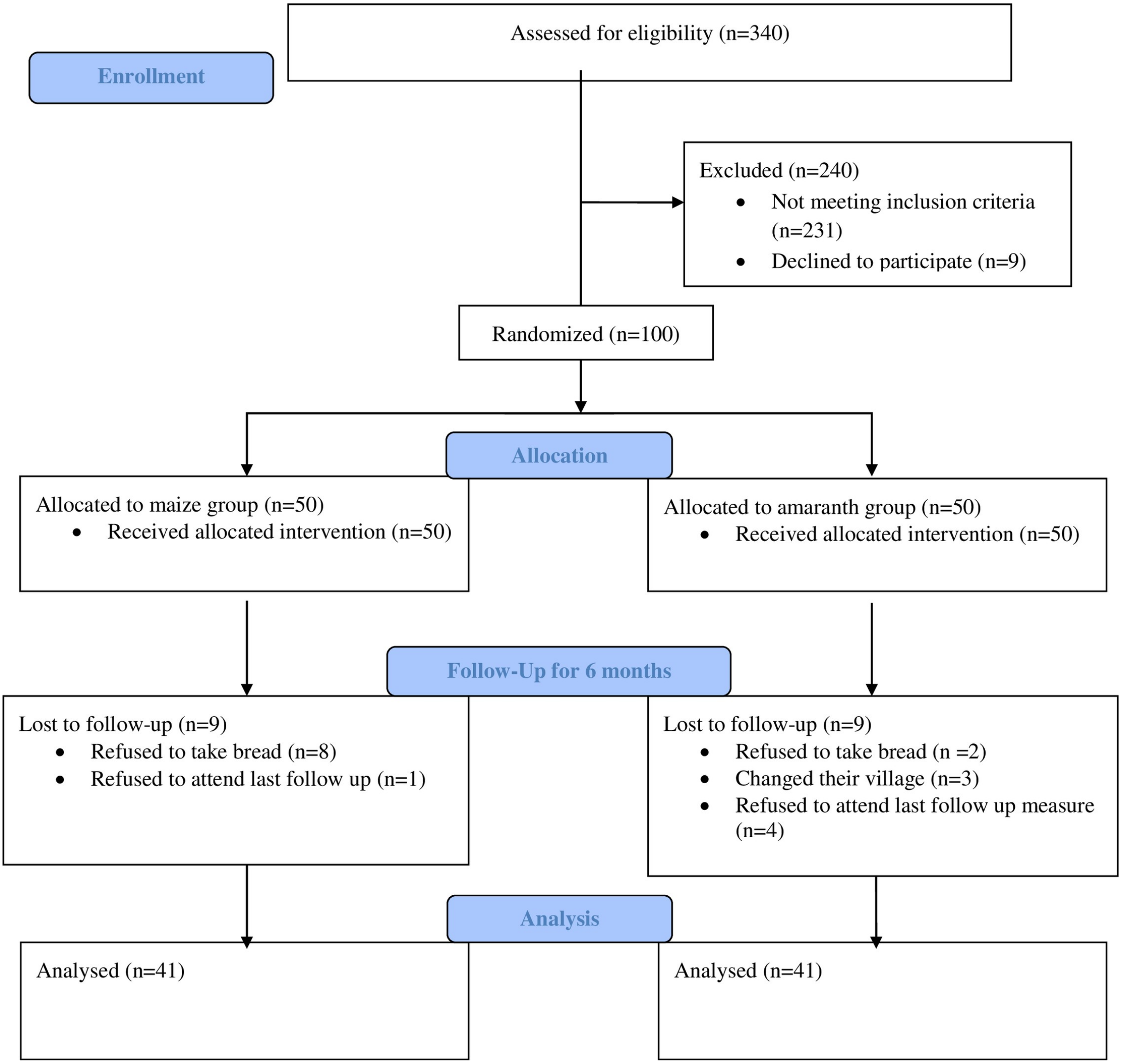

Fig 1. Trial profile.

https://doi.org/10.1371/journal.pone.0239192.g001

There was no statistically significant difference were found in socio-demographic characteristics between discontinued (n = 18) and participated children at last follow-up measure ($P>0.05$) "S2 Table". The six months daily attendance of children with regard to bread consumption indicated that there was no significant mean difference between amaranth and maize groups [-5.32 (95% CI,-20.1; 9.5)] with P-value > 0.5 "Table 2".

Table 2. Mean of bread consumption attendance between groups.

Six month children attendance	Mean (CI)	Mean difference (CI)e	P-value
Maize group	146.9 (135.1–158.6)	-5.32 (-20.1;9.5)	0.5
Amaranth group	152.2 (142.9–161.5)		

https://doi.org/10.1371/journal.pone.0239192.t002

Baseline characteristics of children

From the hundred participants, half were girls and the mean age was 37 months (95% CI: 34.8–39.1). Majority 73 (73%) of children scored the lowest dietary diversity and 4 (4%) were scored high dietary diversity. Child illness history indicated that 38 (38%) had a history of watery diarrhea in the past 15 days preceding the survey. Regarding the nutritional status of the children, 53 (53%) were stunted, and 28 (28%) were underweight. Stunting prevalence was significantly higher in the amaranth arm 31/50 (62%) than in the maize group 22/50 (44%) "Table 3".

Baseline characteristics of mother and household

All caregivers were mothers, and their median age was 26 (IQR: 24–30) years. The median completed school years by mothers were 4 (IQR: 0–7) years and 63 (63%) of the mothers were unemployed. Majority 77 (77%) of the households were earned less than 1.9 USD per day, and 69 (69%) of the households were food insecure "Table 4".

Magnitude of anemia and iron deficiency anemia at baseline

From the total children with anemia, 29/100 (29%) were moderately anemic and 71/100 (71%) were mildly anemic. Children with moderate anemia were 38% in amaranth group and 20% in maze group. Iron deficiency anemia constitutes 30% of the total anemia cases and distributed as 35% in the amaranth group and 24% in the maize group "Table 5".

24 hours dietary diversity score

The mean dietary diversity score (DDV) indicated that there was no significant difference between amaranth and maize arms for each month "Table 6".

Six month follow-up of iron-rich food consumption

Seven days food frequency recall indicated that 59% of children's were not consumed iron-rich food at least one time for the last six consecutive months follow up. There is no significant difference between groups on iron-rich food consumption (p-value >0.5) "Table 7".

The effect of amaranth containing bread on anemia prevalence

At last follow-up measure, the overall prevalence of anemia decreased to 44% as compared with 100% at baseline. Intention-to-treat analysis indicated that, children who received amaranth containing bread had 61% significant reduction in the risk of anemia compared to those who received maize bread [aRR: 0.39 (95%CI: 0.16–0.77)]. The prevalence of anemia was significantly lower (32%) in the amaranth group as compared with the maize group (56%) Similarly, the complete-case analysis result indicated a significant decrease of anemia prevalence in amaranth group (17%) as compared with (46%) in the maize group [aRR: 0.18 (95%CI: 0.05–0.57)], "Table 8".

Table 3. Baseline characteristics of children, maize and amaranth groups.

Characteristics	Total N = 100	Maize	Amaranth
Child age month (mean (CI) (Continuous)	36.9 (34.8–39.1)	36.6 (33.3–39.8)	37.7 (34.9–40.5)
Categorical data	N (%)*	N = 50; N (%)*	N = 50; N (%)*
Child sex			
Boy	50 (50)	22 (44.0)	28 (56.0)
Girl	50 (50)	28 (56.0)	22 (44.0)
Diarrhea in the past 15 days			
No	62 (62.0)	32 (64.0)	30 (60.0)
Yes	38 (38.0)	18 (36.0)	20 (40.0)
Cough in the past 15 days			
No	76 (76.0)	37 (74.0)	39 (81.3)
Yes	22 (22.0)	13 (26.0)	9 (18.8)
Missing	2 (2.0)		
Hospital admission since birth			
No	88 (88.0)	43 (86.0)	45 (90.0)
Yes	12 (12.0)	7 (14.0)	5 (10.0)
Child dietary diversity			
Low	73 (73.0)	38 (77.6)	35 (71.4)
Medium	21 (21.0)	10 (20.4)	11 (22.4)
High	4 (4.0)	1 (1.0)	3 (3.1)
Missing	2 (2.0)		
Height for age (HAZ)			
No stunting	47 (47.0)	29 (58.0)	18 (36.0)
Stunting	53 (53.0)	22 (42.0)	31 (64.0)
Weight for height (WHZ)			
Normal	89 (89.0)	44 (95.7)	45 (88.0)
Wasting	4 (4.0)	1 (2.1)	3 (6.0)
Overweight	4 (4.0)	1 (2.1)	3 (6.0)
Weight for age (WAZ)			
Normal	72 (72.0)	38 (76.0)	34 (68.0)
Underweight	28 (28.0)	12 (24.0)	16 (32.0)

https://doi.org/10.1371/journal.pone.0239192.t003

The effect of amaranth bread on hemoglobin concentration

Last follow-up measure indicated that hemoglobin concentration was increased in both amaranth and maize arms when compared with the baseline results. The estimate of beta coefficient indicated that children who received amaranth bread had significantly higher amount of hemoglobin concentration than maize bread, as the 95% CI does not contain zero [aβ 8.9 g/L (95%CI: 3.5–14.3)] "Table 9".

The effect of amaranth bread on iron deficiency anemia

The overall prevalence of iron deficiency anemia decreased from 29% at baseline to 18% at last follow-up measure. Intention to treat analysis showed that iron deficiency anemia risk was significantly decreased in the amaranth group from 35% at baseline to 15% at the last follow-up [aRR: 0.44 (0.23–0.83)]. However, this difference was not significant in the complete case analysis "Table 10".

Table 4. Baseline characteristics of mother and households.

Characteristics	Total N = 100	Maize	Amaranth
Mother age (Continuous) median (IQR)	26 (20–32)	26 (19–31)	26 (20–32)
Categorical data	N (%)*	N = 50; N %)*	N = 50; N (%)*
Mother's occupation			
Unemployed	63 (63.0)	32 (64.0)	31 (62.0)
Day laborer	23 (23.0)	10 (20.0)	13 (26.0)
Government	14 (14.0)	8 (16.0)	6 (12.0)
Father's occupation			
Unemployed	12 (12.0)	7 (14.0)	5 (10.0)
Daily laborer	49 (49.0)	20 (40.0)	29 (58.4)
Government	39 (39.0)	23 (46.0)	16 (32.0)
Electricity access			
No	27 (27.0)	13 (26.0)	14 (28.0)
Yes	73 (73.0)	37 (74.0)	36 (72.0)
Household food security			
Food secure	31 (31.0)	15 (30.0)	16 (32.0)
Mild food-insecurity	14 (14.0)	6 (12.0)	8 (16.0)
Moderate food-insecurity	18 (18.0)	10 (20.0)	8 (16.0)
Severe food-insecurity	37 (37.0)	19 (38.0)	18 (36.0)
Household income per month			
<1500 (1.9 dollars per day)	77 (77.0)	35 (71.4)	42 (85.7)
>1500 (1.9 dollars per day)	21 (21.0)	14 (28.6)	7 (14.3)
Missing	2 (2.0)		

https://doi.org/10.1371/journal.pone.0239192.t004

The effect of amaranth bread on iron deficiency

In the amaranth group iron deficiency was decreased, from 34% at baseline measure to 26% at last follow-up measure. But the risk of iron deficiency had no significant difference between groups [aRR: 0.81(0.55–1.19)] "Table 11".

Discussion

Research on the nutritional value of amaranth and methods to decrease phytate level has been performed [8,9]. However, few investigations exist focusing on the effect of processed amaranth *in vitro*. This study is the first in Ethiopia to assess the effect of homemade processed amaranth grain on the hemoglobin concentration, anemia and iron deficiency anemia status of children two-to-five years-old.

The findings indicated that hemoglobin concentration increased and the prevalence of anemia decreased significantly after intervention. The mean hemoglobin concentration change

Table 5. Magnitude of anemia and iron deficiency anemia distribution between groups.

	Maize N (%)	Amaranth N (%)	Total N (%)
Iron deficiency anemia N = 91			
Yes	11 (24.4)	16 (34.8)	27.0 (29.7)
No	34 (75.6)	30 (65.2)	64.0(70.3)
Anemia Total N = 100			
Mild	40 (80.0)	31 (62.0)	71.0 (71.0)
Moderate	10 (20.0))	19 (38.0)	29.0(29.0)

https://doi.org/10.1371/journal.pone.0239192.t005

Table 6. The mean dietary diversity score of 24 hours recall.

Month	Group	Dietary diversity mean score (CI)	Mean Difference (CI)	P-value
1	Maize	2.5 (2.6–2.2)	-0.24 (-.82;.33)	0.4
	Amaranth	2.8 (2.3–3.3)	-0.24 (-.82;.33)	0.4
2	Maize	2.5 (2.1–2.9)	-0.39 (-1.15;.36)	0.3
	Amaranth	3.1 (2.5–3.7)	-0.39 (-1.15;.36)	0.3
3	Maize	2.8 (2.4–3.2)	-0.49 (-1.19;.21)	0.2
	Amaranth	3.2 (2.6–3.7)	-0.49 (-1.19;0.21)	0.2
4	Maize	2.9 (2.4–3.3)	-0.04 (-0.66;0.58)	0.8
	Amaranth	2.8 (2.3–3.2)	-0.04 (-0.66–0.58)	0.8
5	Maize	2.9 (2.4–3.4)	-0.31 (-1.01–0.38)	0.4
	Amaranth	3.1 (2.6–3.6)	-0.31 (-1.01–0.38)	0.4
6	Maize	3.4 (2.9–3.9)	-0.26 (-0.94–0.41)	0.4
	Amaranth	3.6 (3.2–4.1)	-0.27 (-0.94–0.41)	0.4

https://doi.org/10.1371/journal.pone.0239192.t006

was significantly higher in the amaranth arm as compared with maize arm. Also the risk of anemia was significantly lower in the amaranth arm as compared with maize arm. There is currently no extant literature in the study area examining the effect of processed amaranth grain on anemia status to which our results can be compared. However, one study conducted on amaranth leaf in Ghana showed similar result to this study. Amaranth leaf dried powder had a significant effect on anemia prevalence. Specifically, anemia prevalence decreased by 28% and hemoglobin concentrations were higher by 8.9 g/L after using amaranth leaf powder [10]. The presence of micronutrients other than iron in the amaranth grain, such as folic acid, copper, and vitamin A may have contributed to the increased hemoglobin [22–24], but changes in those levels were not quantified either in that study or the present one.

In contrast, a study conducted in Kenya demonstrated that amaranth did not decrease anemia prevalence significantly [6]. But the study used raw/unprocessed amaranth grain, which is high in phytate, and suggested that reducing the phytate level of amaranth grain may contribute to increasing the hemoglobin level. It is widely recognized that phytate can impede the absorption of iron and other micro- and macro-nutrients from the gut. This problem can be resolved, however, by applying homemade processing, such as soaking, germinating, and fermenting [8,9]. Therefore, we believe that the result obtained in this study was related with homemade processing applied to amaranth grain that decreased the phytate level and enhanced the absorption of iron and other micro- and macro-nutrients.

Iron deficiency anemia is significantly decreased in the amaranth group in the last follow up measure in the intention to treat analysis. But the change was not significant in the complete case analysis. This difference may be due to the presence of higher inflammation at baseline, confirmed by high CRP level in both arms when compared with last follow up. This is supported by other studies reporting that CRP and ferritin have a positive correlation [25]. This means some of the ferritin level observed as normal at baseline measure may related with

Table 7. Seven days iron-rich food frequency recall of six consecutive months follow-up.

Cumulative Frequency	Maize	Amaranth	Total	P-value
0	27 (65.9)	22 (53.7)	49 (59.8)	0.7
1	8 (19.5)	9 (22.0)	17 (20.7)	
2	4 (9.8)	6 (14.6)	10 (12.2)	
3	2 (4.9)	4 (9.8)	6 (7.3)	

https://doi.org/10.1371/journal.pone.0239192.t007

Table 8. The effect of amaranth bread on anemia prevalence.

	No anemia	Anemia	CRR (95% CI)	ARR (95%CI)
Intention-to-treat analysis				
Maize	22/50 (44.0)	28/50 (56.0)	1	1
Amaranth	34/50 (68.0)	16/50 (32.0)	0.57 (0.35–0.92)	0.39 (0.16–0.77)
Complete-case analysis				
Maize	22/41 (53.7)	19/41 (46.3)	1	1
Amaranth	34/41 (82.9)	7/41 (17.1)	0.37 (0.17–0.78)	0.18 (0.05–0.57)

CRR, crude relative risk; ARR, adjusted relative risk; CI, confidence interval; Adjusted for child age, child sex, and child nutritional status, height for age, dietary diversity. For the intention-to-treat analysis, all missing outcomes were treated as a failure (i.e., considered as anemic without knowing their status). For the complete-case analysis, only individuals with outcome data available were included.

https://doi.org/10.1371/journal.pone.0239192.t008

the presence of inflammation. We adjusted ferritin with CRP level but this may not control the false reading of ferritin by hundred percent [26]. This condition may prevent discernment of the precise effect of amaranth on ferritin level or iron deficiency anemia. In the future, larger sample size and inclusion of additional iron indicator tests may provide more information on the iron status of children [26].

The last follow up measure indicated that anemia prevalence was noticeably decreased in the maize group, as well. This may be attributable to a reduction of helminth burden due to albendazole given to both arms. This result also supported by research conducted on the effect of deworming on anemia prevalence [27,28]. Furthermore, the daily supervision of a bread provider at the household level may have created awareness of sanitation and child-feeding practices, which in turn may have had a positive impact by decreasing the incidence of inflammation and improved dietary diversity. The fermented maize bread itself may also have increased the intake of iron and other micro and macro nutrients.

The acceptability test was not done for this particular research. However, prior to this study a similar combination was tested for acceptability in the research area by the same author and the result indicated the prepared food was acceptable by the community [9]. Non-acceptability of the food was also not reported during supervision and follow-up time.

Regarding the iron intake, there were no other iron supplementation programs in the research area. Further, we did a follow up every month for dietary diversity and iron-rich and bioavailable food consumption such as animal product (red meat, organ meat and fish). The six month follow up indicated that there was low consumption of iron-rich food and there was no significant difference between groups both for dietary diversity score and iron-rich food consumption.

Table 9. The effect of amaranth bread on hemoglobin concentration.

	Baseline mean Hemoglobin g/L	Last follow up mean Hemoglobin g/L	Estimated of beta coefficient (CI)	P-value
Complete case analysis				
Maize	103.5 (100.9–106.0)	110.1 (106.9–113.2)	1	
Amaranth	101.3 (98.7–103.9)	116.2 (112.1–120.3)	8.9 (3.5–14.3)	0.003
Intention to treat analysis				
Maize	103.7 (101.5–105.8)	109.1 (106.4–111.8)	1	
Amaranth	101.0 (98.7–103.4)	113.3 (109.4–117.1)	7.0 (2.1–12.0)	0.006

Hemoglobin was adjusted for time, sex, age, and nutritional status (height for age).

CI: confidence interval; estimated beta coefficient, CI, and P-value analyzed using the GEE linear model.

https://doi.org/10.1371/journal.pone.0239192.t009

Table 10. The effect of amaranth bread on iron deficiency anemia.

	Baseline		Last measurement		Adjusted relative risk ratio (95% CI)
	Iron deficiency anemia		Iron deficiency anemia		
Complete case analysis N = 76	**No**	**Yes**	**No**	**Yes**	
Maize	30 (78.9)	8 (21.1	30 (78.9)	8 (21.1)	1
Amaranth	25 (65.8)	13 (34.2)	31 (81.6)	7 (18.4)	0.54 (0.28–1.03)
ITT analysis N = 91					
Maize	34 (75.6)	11 (24.4)	35 (77.8)	10 (22.2)	1
Amaranth	30 (65.2)	16 (34.8)	39 (84.8)	7 (15.5)	0.44 (0.23–0.83)*

CI: confidence interval; relative risk ratio, CI, and P-value analyzed using the GEE binomial model. Iron deficiency anemia was adjusted for time, sex, age, dietary diversity and nutritional status (height for age).

https://doi.org/10.1371/journal.pone.0239192.t010

Strengths and limitations of the study

This study used a simple random method to identify the study participants and cluster randomization to allocate anemic children into the two arms, so that the results obtained in this research can be reproducible if performed in a similar manner. The bread maker, bread distributor, caregivers, and data collectors were unaware of the maize or amaranth bread allocations. The caregivers fed their children daily under direct supervision. Flour blending was performed carefully considering the ratio of amaranth, and the ratio and size of the bread were checked regularly by a trained supervisor.

This study does not assessed other nutrient-related anemia, such as folic acid deficiency, vitamin A deficiency, and copper deficiency as well as does not considered genetic related hemoglobin disorder even though it is not common in our setup. Further we didn't measured AGP (a-1-acid glycoprotein) concentration to adjust ferritin. In addition, we did not conducted malaria test and stool examination The sample size for the survey was established when individual randomization was planned; however, during the survey period, we understood that children with anemia sparsely distributed and we clustered them by geographical location to effectively manage home based feeding and supervision, and thus clustering was not taken into account during sample size calculations however we managed during analysis using intra class correlation coefficient.

Conclusion

The consumption of processed amaranth-containing bread decreased anemia prevalence, increased mean hemoglobin concentration, and minimized iron deficiency anemia prevalence

Table 11. Iron deficiency distribution between groups at baseline and at last follow-up measure.

Intervention group	Iron deficiency Baseline		Iron deficiency last follow up		Relative risk (95%CI)
	Yes	**No**	**Yes**	**No**	
Complete case analysis N = 76					
Maize	8 (21.1))	30 (78.9)	9 (23.7)	29 (76.3)	1
Amaranth	13 (34.2)	25 (65.8)	10 (26.3)	28 (73.7)	1.12 (0.81–1.55)
Intention to treat analysis N = 91					
Maize	11 (24.4)	34 (75.6)	12 (26.7)	33 (73.3)	1
Amaranth	16 (34.8)	30 (65.2)	13 (28.3)	33 (71.7)	0.81 (0.55–1.19)

CI: confidence interval; relative risk ration and CI; analyzed using the GEE binomial model. Iron deficiency were adjusted for time, sex, age, dietary diversity and nutritional status (height for age).

https://doi.org/10.1371/journal.pone.0239192.t011

of the participated children. Therefore, processed amaranth grain bread has the potential to minimize the prevalence of anemia.

Supporting information

S1 Table. Main outcome variables result of intra class correlation coefficient.
(DOCX)

S2 Table. Comparing the sociodemographic distribution between complete case and missed case.
(DOCX)

S1 Appendix. Questionnaire used to collect data.
(DOCX)

S2 Appendix. Trial protocol.
(DOCX)

S3 Appendix. CONSORT checklist.
(DOCX)

Acknowledgments

The authors are grateful to Mrs. Abebayehu Zebdewos and Mr. Teketel for blood sample collection in the field. Mr. Damte Data Daba and Mrs.Kassaye Zebdewos Orsango were coordinators of field activities. All data collectors performed data collection in the field. We also want to thank the child caregivers, drivers, bread makers, the Cheffe Cotte Jebessa Health Center, all health post workers, Cheffe Cotte Jebessa community leaders, and all study participants.

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Paper III supplementary table 1. Main outcome variables result of intra-class correlation coefficient

Eight groups	Intra class coefficient	Confidence interval	P-value
Anaemia end-line	0.04	0.002-0.51	0.19
Hemoglobin end-line	0.09	0.01-0.58	0.11
Hemoglobin baseline	0.01	8.35E-07	0.41
Ferritin baseline	0.05	0.003 - 0.51	0.16
Ferritin end-line	0.03	0.001 - 0.6	0.25

Paper III Supplementary Table 2. Comparing the distribution between complete-case and missed-case.

		Completed arms 82 (%)		Total	Missed arms 18 (%)		Total
		Maize=41	Amaranth =41		Maize=9	Amaranth =9	
Child sex	Boy	19 (46.3)	25 (61.0)	44 (53.7)	3 (30.0)	4 (50.0)	7 (38.9)
	Girl	22 (53.7)	16 (39.0)	38 (46.3)	7 (70.0)	4 (50.0)	11 (61.1)
Child age in months (constant)		38 (34-42)	36 (33-40)	37 (35-40)	35 (23-41)	46 (28-48)	37(31-43)
Electricity access	No	10 (24.4)	12 (29.3)	22 (26.8)	2 (20.0)	2 (25.0)	4 (22.2)
	Yes	31 (75.6)	29 (70.7)	60 (73.2)	8 (80.0)	6 (75.0)	14 (77.8)
Owns household land	No	25 (61.0)	29 (70.7)	54 (65.9)	7 (70.0)	6 (75.0)	13 (72.2)
	Yes	16 (39.0)	12 (29.3)	28 (34.1)	3 (30.0)	2 (25.0)	5 (27.8)
Owns domestic animal	No	23 (56.1)	24 (60.0)	47 (58.0)	6 (60.0)	7 (87.5)	13 (72.2)
	Yes	18 (43.9)	16 (40.0)	34 (42.0)	4 (40.0)	1 (12.5)	5 (27.2)
Current breast feeding	Yes	16 (39.0)	15 (36.6)	31 (37.8)	5 (55.6)	4 (50.0)	9 (52.9)
	No	25 (61.0)	26 (63.4)	51 (62.2)	4 (44.4)	4 (50.0)	8 (47.1)
Iron supplement during pregnancy	No	26 (63.4)	23 (56.1)	49 (59.8)	8 (80.0)	7 (87.5)	15 (83.3)
	Yes	15 (36.6)	18 (43.9)	33 (40.2)	2 (20.0)	1 (12.5)	3 (16.7)
Vaccine completed (mother's response)	No	7 (17.1)	4 (9.8)	11 (13.4)	1 (10.0)	1 (12.5)	2 (11.1)
	Yes	34 (82.9)	37 (90.2)	71 (86.6)	9 (90.0)	7 (87.5)	16 (88.9)
Diarrhea in the past 15 days	No	27 (65.9)	25 (61.0)	52 (63.4)	5 (50.0)	5 (62.5)	10 (55.6)
	Yes	14 (34.1)	16 (39.0)	30 (36.6)	5 (50.0)	3 (37.5)	8 (44.4)
Cough in the past 15 days	No	32 (78.0)	34 (82.9)	66 (80.5)	6 (60.0)	6 (75.0)	12 (66.7)
	Yes	9 (22.0)	7 (17.1)	16 (19.5)	4 (40.0)	2 (25.0)	6 (33.3)
Hospital admission since birth	No	35 (85.4)	36 (87.8)	71 (86.6)	10 (100.0)	8 (100.0)	18 (100.0)
	Yes	6 (14.6)	5 (12.2)	11 (13.4)	0	0	
Mother aware of iron-rich foods	No	36 (87.8)	40 (97.6)	76 (92.7)	8 (80.0)	7 (87.5)	15 (83.3)
	Yes	5 (12.2)	1 (2.4)	6 (7.3)	2 (20.0)	1 (12.5)	3 (16.7)
Mother's	Unemployed	28 (68.3)	26 (63.4)	54 (65.9)	5 (50.0)	4 (57.1)	9 (52.9)

occupation	Day laborer	5 (12.2)	12 (29.3)	17 (20.7)	4 (40.0)	1 (14.3)	5 (29.4)
	Government /non-government	8 (19.5)	3 (7.3)	11 (13.4)	1 (10.0)	2 (28.6)	3 (17.6)
Father's occupation	Unemployed	6 (14.6)	8 (19.5)	14 (17.1)	3 (30.0)	0 (0.0)	3 (17.6)
	Day laborer	15 (36.6)	21 (51.2)	36 (43.9)	3 (30.0)	4 (57.1)	7 (41.2)
	Government/ non-government	20 (48.8)	12 (29.3)	32 (39.0)	4 (40.0)	3 (42.9)	7 (41.2)
Household dietary diversity	Low	13 (32.5)	13 (31.7)	26 (32.1)	5 (55.6)	1 (14.3)	6 (37.5)
	Medium	19 (47.5)	17 (41.5)	36 (44.4)	2 (22.2)	3 (42.9)	5 (31.3)
	High	8 (20.0)	11 (26.8)	19 (23.5)	2 (22.2)	3 (42.9)	5 (31.3)
Child dietary diversity	Low	32 (80.0)	30 (73.2)	62 (76.5)	6 (60.0)	4 (57.1)	10 (58.8)
	Medium	6 (15.0)	9 (22.0)	15 (18.5)	3 (30.0)	3 (42.9)	6 (35.3)
	High	2 (5.0)	2 (4.9)	4 (4.9)	1 (10.0)	0 (0.0)	1 (5.9)
Household food security	Food secure	15 (36.6)	13 (31.7)	28 (34.1)	0 (100.0)	3 (100.)	3 (16.7)
	Mild food-security	4 (9.8)	6 (14.6)	10 (12.2)	2 (20.0)	2 (25.0)	4 (22.2)
	Moderate food-insecurity	7 (17.1)	8 (19.5)	15 (18.3)	2 (20.0)	1 (12.5)	3 (16.7)
	Severe food-insecurity	15 (36.6)	14 (34.1)	29 (35.4)	6 (60.0)	2 (25.0)	8 (44.4)
HAZ	Normal height	24 (58.5)	16 (39.0)	40 (48.8)	6 (60.0)	3 (37.5)	9 (50.0)
	Stunting	17 (41.5)	25 (61)	42 (51.2)	4 (40.0)	5 (62.5)	9 (50.0)
WHZ	Normal	37 (97.4)	37 (94.9)	74 (96.1)	9 (100.0)	6 (75.0)	15 (88.2)
	Overweight	1 (2.6)	1 (2.6)	2 (2.6)	0 (0)	1 (12.5)	1 (5.9)
	Wasting	0 (0.0)	1 (2.6)	1 (1.3)	0 (0)	1 (12.5)	1 (5.9)
Household income per day	Less than 1.9$	30 (73.2)	35 (85.4)	65 (79.3)	7 (70.0)	6 (85.7)	13 (72.2)
	Greater than 1.9$	11 (26.8)	6 (14.6)	17 (20.7)	3 (30.0)	1 (14.3)	4 (27.8)

Consent form and Questionnaire

Locality Name	Village Name
Name of Household Head	Name of mother
Name of Child_	Cluster Numbe
Household Identity Number	Name of Supervisor

How are you? My name is ______________________Your house is selected randomly for this interview .The purpose of this interview is to collect base line data for the next experimental study for the evaluation of amaranth grain on the iron status of children in the age of 24-59 months and which factors that are related to that. The eligible volunteer will be engaged in the study if they fulfil the criteria. Decision on your involvement will be made by you and only you. If you are willing to participate, you will be expected to realize the following requirement you are required to give us information about you, your house hold and about child, the child weight and height will be measured and blood will be collected from your child to know the child anaemia status. Should you here say that around 1/3 from the survey will be asked to participate in a nutrition study? Or should you wait?

Risk the child will fell mild pain during blood collection and there is no further physical or psychological risk expected being involved in the study

Benefit You have the right to know the finding of the study, you will be given experts advice about anaemia and iron rich foods. The result which produced from your participation will help to fight iron deficiency anaemia in the nation.

Confidentiality Yours and your Childs information will only be used for the purpose of the study. You and your child will not be personally identified in the study report without your will.

Participation You have to know that your participation is largely based on your willingness and approval. You have the right to say “no” and not participate in the study. You will not be penalized if you decide not to participate. If you wish to withdraw from this study you can do that at any time

Questions: you can ask any question about this study

Confirmation of agreement: I have read the consent form /the interviewer has read the consent form. I have understood the aim of the study and the things that I have to do if I agreed to participate in the study. I know that my participation is based on my will and have right not to do so, if I do not want to participate.

Please tell us if you agree or not

Yes ______________

No______________

Mothers name _________________signature _____________

Interviewer signature ______________________________

Date _____________________________

Thank you for your willingness to participate in this study

Consent form translated to local language

Suffetto 2: Xaphoomuna babaxitino xiinxello sumuumme

Teessote Su'ma	Olluu summa
Mini maate anni su'ma	Amate Su'ma
Qaaqquu su'ma	Qachu Kiiro
Minu Kiiro	Sooreessu Suma

Keereholla ? Heeshsho hittoti? Summa'yayaamameemo.

Ate mine xinxallote qorsha assate doodhinomo tenne xiinxallo qorshi korkaati. Raffote yinayi shaanina gumisi giddo afentano ayironete yinayiti Qaaqqulleho yaano 24-59 agani daaimira uyitanno horronna mannu tenna horo uyitanno sagale horonsirate roso xiinxallateeti .Xiinxallonkera mahoye yaanohuna.Hasiisannore wonshannohu assineemo xiinxallora summa yateni kaalannonke.Tenne yoo mahoye yaattohu umikki fajjonni atenna ate callaho. Sumuu yiittoro koonni aantino xa'mora helaale ikkitino dawaro dawaratonkehu minikki maate lainohunnina qaaqqoki lainnohuniti. qaaquullu ayirrena hajjassi seendille keenneemo. anemiyu xissonni fayyo ikassina looposi buuxate shiima mundee haa'nemo.

Hullullo: Mundee haaneyi woyite qaaqqoho xissote gederi maccishamawo .ikkollana kayiinni bisisi ananna angoolessi aana kalaqamemo qarri dino.

Horo: Tenne xiinaxaallo hassato maatiro afete mafte (feceleeqo) noohe. Aneemiyu xisso lainohunnina ayirene uyitano sagale lainohunni rosicho uyiinannihe. Ati ledonke loosakinni kainohunni qaaqquullu ana iilitanota ayirenete anji mundeete anje gargarate dandineemo.

Afa nooheti : Atena qaaqqikki dawaro horo uyitennohu Tenne xiinxallo callateeti. Atenna qaaqqikki fajo nookiha ki'ne ayima ayiino afara didandaanno.

Beeqqa: Ate beqqa tenna xinxallora uyiitanno horo buuxxe affe ‘’mahoooye’’ woy “gimbi” yaate dandaatota. Maahoye yaa giwittoro gadadishshuni/wolqateni mahoye yiisiisannohehu dino . Debeeqqeeemo yaa dandaatto .Hanaffeno urrisemmo yiitoro ayii yannaranno uurisa dandaatto.

Xamo: Xiinxallo lainohunni ayii xa’mono xa’ma dandaatto

Sumuu yaate buuxo: Xiinxallote borro mabawoomo xiinxallote borro xa’maanchu seeke mabawinoe. Xiinxallote heddonna assinayire baala buuxxoomo. Beeqqate summuu yoommo. Xiinxallo assineyihu umiya fajjonnina beeqqonni ikino dafira tashi yiiero beeqqanna tashi yaa hoogiero agura dandemota buxoommo.

Kulie henni mahoyenso gimbi yaato?

Mahooye ____________________________Gimbi_______________________________________

Amate su’ma _________________________________Malaate_____________________________

Xamaanchu su’ma___________________________Barra________________________________

Tenne xiinxallo beeqqate unikki fajjonni sumuu yoottohure lowo geeshsha galaxineemohe.

QUESTIONNAIRE - PREVENTING IRON DEFICIENCY ANAEMIA IN ETHIOPIA

DATE OF INTERVIEW |___|___| ***Day*** |__|__| ***Month*** |__|__||__|__| ***Year***

TIME STARTED |__ |__| ***Hour*** |__|__| ***Minutes***

TIME ENDED |__ |__| ***Hour*** |__ |__ | ***Minutes***

INTERVIEWER NAME ____

SUPERVISOR NAME ____

CHECKED BY ____

ENTERED BY ____

LOCALITY NAME____

VILLAGE NAME ____

NAME OF HOUSEHOLD HEAD____

NAME OF MOTHER ____

NAME OF CHILD ____

HOUSEHOLD IDENTITY NUMBER____

KEBELE CODE____

Part I

Socio demographic characteristics of the mother or care giver of the child

Ask the caregiver to find the vaccination card (cross-check all information with the card)

እናትየውን የክትባት ካርድ ጠይቀህ/ሽ የምትሰ ጠውን መልስ ከካርዱ ጋር አስተያይ/ዪ

No	Question			
101	**How long have you been living here as household/family? Put in year**			
	Tenne mini maate ledo megegshi yanna heerita? Diro KUlie			
	ቤተሰብህ/ሽ ለምን ያህል ጊዜ እዚህ ቆያችሁ; በአመት አስቀምጥ/ጪ (ከ 6 ወር በታች ከሆነ =0 ፤ ከ 6 ወር በላይ ከሆነ =1 አስቀመጥ/ጭ)			
102	**Did your household planed to live here for the next one year?**			
	Danno diro konne heerate hedo noohe?	0. [_] No	0. [_] Dinoe	0. [_] የለውም
	ቤተሰብሽ ለሚቀጥለው 1 አመት እዚህ የመቆየት እቅድ አላችሁ?	1. [_] Yes	1. [_] Nooe	1. [_]አለው
103	**How old is this child? (age in month) and date of birth in GC and EC**			
	3a. Qaaqqu ilamino barra(Itiyopiyu kiironni)		_____,______,____	____,______,____
	ህጻኑ የተወለደበት ቀን (በኢትዮጲያ አቆጣጠር)			
	3b. Illamino barra (aroyopu kiirronni)		______,______,____	______,______,___ _
	ህጻኑ የተወለደበት ቀን (በአውሮፓ) አቆጣጠር			
	3c. Ilamino barra afamannoki haikkiro qaaqqu diro agaanunni		__________	____________
	የተወለደበት ቀን የማይታወቅ ከሆነ የልጁ እድሜ በወር			
104	**How old are you? (mother age in years)**			
	Dirikki meeho? Kiirotenni kulie.			
	የእናትየው ዕድሜ ስንት ነው ? (በአመት)			
105	**Is the child a boy or a girl?**			
	Daaimu Koo/tee? Meyatenso labaho	1. [_] Boy	1. [_] Labaaho	[_] 1.ወንድ
	ሕፃኑ ወንድ ነው ሴት?	2. [_] Girl	2. [_] Meyaate	[_] 2.ሴት
106	**What is your relationship with the child?**			
	Daaimuua ate fixoomi maati?	1. [_] Mother	1.[_]Amate	1[_] እናት
	አንቺ ለህፃኑ ምኑ ነሽ?	2. [_] Father	2.[_]Annaho	2[_] አባት
		3.[_] Step mother /father	3.[_]Buddeenu amate	3[_] እንጀራ እናት/ባት

		4.[_] Grand mother/Father	4.[_]Ahaahete/ hoho	4[_] አያት
		5. [_] Sister/brother	5.[_]Rodoote/Rodooho	5[_]እህት/ወንድም
		6. [_] uncle/aunt	6.[_] Aboho/halamete	6[_] አጎት/አክስት
		7. [_] other specify	7.[_]Welere	7[_]ሌላ/ዘርዝር
107	**What is your religion**			
	Anunoki meati?	1.[_]Orthodox	1.[_]Orthodoksete	1[_] አርቶደክስ
	ሀይማኖትሽ ምንድን ነው?	2. [_] Protestant	2.[_] Protestantete	2[_] ጴንጤ
		3. [_] Catholic	3.[_] Katoolikete	3[_] ካቶሊክ
		4. [_] Muslim	4.[_] Isilamaho	4[_] ሙስሊም
		5. [_] Traditional	5.[_] Budu amanooti	5[_] ባህላዊ
		6. [_] Other specify	6.[_] Wolere	6[_] ሌላ/ዘርዝር
108	**What is your marital status?**			
	Mine kalaqirooto/ta?	1.[_]Single	1.[_]Minaamedti	1[_] ያገባ
	የጋብቻ ሁኔታሽ/ህ ምን ይመስላል?	2. [_] Married	2.[_]Qeedhichchaho	2[_] ያለገባ
		3. [_] Divorced	3.[_] Fateherawase	3[_] ተራርቆ የሚኖር
		4. [_] Widowed	4.[_] Tiroomo/ma	4[_] የተፋታ/ች
		5. [_] Separated	5.[_]Shirroomo/ma	5[_] የሞተባት/በት
109	**How many years of completed school does the mother have?**			
	Rosu aana mee diro sayisootta			
	በትምህርት ላይ ምን ያህል አመት አሳልፈሻል?			
110	**How many years of completed school does the father have?**			
	Annu rosu mine mee dirosayisino			
	የህፃኑ አባት በትምህርት ቤት ምን ያህል አመት አሳልፏል?			
111	**What is the total number of people living in your household?**			
	Tenne mini maate eiddo moo manni kirro meeho?			
	በዚህ ቤት ውስጥ ምን ያህል ሰው ነው የሚኖረው?			
111A	**A. How many are younger than 5 year?**			
	Ontu dirii worihu meeho?			
	ከ5 አመት በታች ምን ያህል ናቸው?			
111B	**How many are 5 and older but younger than 18year?**			

	Ontu dirii alihuna 18 dirii worihu nee mannati?			
	በ5 አመትና በ18 መከከል ምን ያህል ናቸው?			
111C	**How many are 19 up to 59year?**			
	19-59diri geesha nee mannaati?			
	ከ19-59 አመት መካከል ምን ያህል ናቸው?			
111D	**How many are older than 60 year?**			
	60 diri alihu meeho?			
	ከ60 አመት በላይ ምን ያህል ናቸው?			

Part 2 Question on Economic status of the house hold

Kirro 2 Mini manni maatete jirote bikka

ክፍል 2. የቤተሰብ ምጣኔ ሀብት መለኪያ መጠይቅ

201	**What is the mother occupational status?**			
	Amote loosi hiitooho?	1.[_] Unemployed	1.[_]Loosu dinooho	1[_]ሰራ የሌለው
	የእናትየው ዋና ስራ ምንድን ነው?	2. [_] Day laborer	2.[_]Barru loosasicho	2[_]የቀን ሰራተኛ
		3. [_] Farmer	3.[_] Baatote looso	3[_]ገበሬ
		4. [_] Merchant	4.[_] Dadalanchoho	4[_]ነጋዴ
		5. [_] NGO employed	5.[_]Mengistati looso	5[_]የድርጅት
		6.[_]Government employed	6.[_]Manaisete looso	6[_]የመንግስት
		7. [_] Student	7.[_]Rosaanchoho	7[_]ተማሪ
		8.[_]Other specify	8.[_]wolere xawisi	8[_]ሌላ ካለ ግለፅ
202	**What is the Father occupational status?**			
	Annu Loosi hiittooho?	1.[_] Unemployed	1.[_]Loosu dinooho	1[_]ሰራ የሌለው
	የአባትየው የስራ ሁኔታ?	2. [_] Day laborer	2.[_]Barru loosasicho	2[_]የቀን ሰራተኛ
		3. [_] Farmer	3.[_] Baatote looso	3[_]ገበሬ
		4. [_] Merchant	4.[_] Dadalanchoho	4[_]ነጋዴ
		5. [_] NGO employed	5.[_]Mengistati looso	5[_]የድርጅት
		6. [_] Government employed	6.[_]Manaisete looso	6[_]የመንግስት
		7. [_] Student	7.[_]Rosaanchoho	7[_]ተማሪ
		8.[_]Other specify	8.[_]wolere xawisi	8[_]ሌላ ካለ ግለፅ
203	**What was last month income?**			
	Sai aganira megeeshi afidhinoonni			
	ባለፈው ወር ቤተሰቡ ያሰገባው ወራዊ ገቢ ምን ያህል ነበር?			
	What is the average yearly income of the household?			
204	Dirunnit mereerima eone megeeshshaatic?			
	በአማካይ የቤተሰቡ አመታዊ ገቢ ምን ያህል ነበር?			
205	**How much do you save yearly?**			
	Dirruni megeeshshi woxe suuqisidhinenni?			
	በአመት ምን ያህል ትቆጥባላችሁ?			
206	**House hold facility : Do you have any of the following**			

	Mini meate injo konniaane nori gido kinera noori noo'ne?	1. [_] Electricity	1[_] Maabraate	1[_] ማብራት
	በቤት ውስጥ ከሚገኙ መገልገያ ቁሶች የትኞቹ አሉሽ?(የሌላትን 0 ሙላ)	2. [_] Radio	2[_] Radoone	2[_] ራዲዎ
		3. [_]Mobile telephone	3[_]Silke	3[_] ሰልክ
		4. [_]Non mobile telephone	4[_] Teevizhiine	4[_] ቴሌቪዥን
		5. [_] Television	5[_]Qiissaasinchu	5[_] ፍሪጅ
		6. [_] computer	6[_]Maabiraatete	6[_] የማብራት ምድጃ
		7. [_] refrigerator	7[_] Compitere	7[_] ኮምፒውተር
		8. [_] electric stove	8[_] Sayiikile	8[_] ሳይክል
		9. [_] Motor bike	9[_]Motorete sayikle	9[_] ሞተር ሳይክል
		10[_] Car	10[_] መኪና	10[_] መኪና
		11[_] Other specify	11[_] ሌላ ካለ ግለፅ	11[_] ሌላ ካለ ግለፅ
207	**Does the household own any agricultural land?**			
	Konni mini maatera umi'ne baato loosidhinenniti noo'ne?	0. [_] No	0. [_] Dinoe	0[_] የለውም
	የዚህ ቤት ባለቤት የእርሻ መሬት አለው?	1. [_] Yes	1. [_] Nooe	1[_] አዎ
208	**How many (LOCAL UNITS) of agricultural land do this household own?**			
	Allanne halaligne mepeeshite (Kine keeninni) Akine umine baattoti?			
	የእርሻ መሬት ካለው ምን ያህል መሬት ነው ያለው (በባህለዊ መለኪያ አስቀምጥ)			
209	**Does the house hold produce any yield with this land**			
	Baatone loosine guma afidhinenni?	0. [_] No	0. [_]Diafineeno	0[_] የለውም
	በዚህ መሬት የምታመርቱት ምርት አለ?	1. [_] Yes	1. [_]Afineeno	1[_] አዎ
210	**If yes what do you produce and how much (put zero if not)**			
	Afidhinennihe ikkino baattoneana loosiinennihu maati? Mageeshshua afidhinanni?			
	ከሚከተለው ምርት ውስጥ ምን ያህል ታመርታላችሁ (ካላመረቱ 0 አስቀምጥ)			
	Inset? (root)		Weese	እንሰት
	Maize? (quintal)		Badala	በቆሎ
	Cabbage?(lood)		Shaana	ጎመን
	Potato? (quintal)		Dinnichha	ድንች

	Chilli? (quintal)		Shama barbare	ቃሪያ
	Sugercan?(load)		Shoonkoora	ሸንኮራ ዐገዳ
	Tomato? (box)		Timatime	ቲማቲም
	Banana? (load)		Muuze	ሙዝ
	Avocado? (quintal)		Awukado	አቮካዶ
	Mango? (quintal)		Maango	ማንጎ
	Kchat? (load)		Chaate	ጫት
	Other specify		Welere	ሌላ ካለ ግለፅ
211	**Does your house hold own any domestic animal**			
	Konni mini maatera mini saado no?	1. [_] Yes	0. [_] Dinoe	0 [_].አይደለም
	የቤት እንስሳት አላችሁ?	2. [_] No	1. [_] Nooe	1 [_].አዎ
212	**How much of the following do you have?** Afidhinoonniha ikkiro hiite saada? Megessha?	Chiken	1.Lukkicho____	1. ዶሮ ____
	የቤት እንስሳ ካለችሁ ከሚከተሉት ውስጥ ምን ያህል ዐላችሁ?**(የሌለውን 0 አስቀምጥ)**	Goat	2. Meicho_____	2. ፍየል____
ዝለል Skip		Sheep	3.Gereewo____	3. በግ ____
		Ox	4.Bootta_______	4. በሬ ________
		Cow	5.Lalo______	5. ላም ____
		Donkey	6.Harricho____	6. አህያ ____
		Other specify	7.wolere xawisi_______	7. ሌላ ዘርዝሪ/ር____
213 ዝለል Skip	**If yes for question number 211 Do you feed animal products for your children**			
	Saedate winni afidhi nannire daaimaho uyitinnan?	0. [_] No	0. [_] Dinoe	0. [_] አላበላም
	የራሰሽን የከብቶች ተዋፅኦ ለልጆችሽ ታበያለሽ?	1. [_] No	1. [_] Nooe	1. [_] አዎ
214	**What type of latrine do you have**			
	Shumate mini hittoohu noone?	1. [_] no facility/bush/field	1[_]Dinoe	1[_]የለንም
	ምን አይነት ሽንት ቤት ነው ያላችሁ?	2. **[_]** composting toilet	2[_]Irshu giddo	2[_]የማዳ ውስጥ
		3. [_] open pit	3[_]Haqqunnabushshunni calla tu'nooni	3[_]ክፍት ጉድጓድ
		4. [_] pit latrine with slab	4[_]Simmintoteni loonsoonnishuma mine.	4[_]የወለል ልባስ ያለው የጉድጓድ ሽንት ቤት

		5. [_] ventilated improved pit latrine (vip)	5[_]Tuubbotenni foolanno shuamte mine.	5[_]የተሻሻለ የጉድጓድ ሽንት ቤት
		6.[_] flush or pour flush toilet	6[_]Wiyiinni loosanno shumate mine.	6[_]በውሀ የሚሄድ ሽንት ቤት
215	**What is the main source of drinking water for your house hold**			
	Waa horonsidhinonnihu maminniti?	1. [_]unprotected well/spring	1[_]Huxxinoonnkki buichcho	1[_]ካልተከለለ የከርሰ ምድርውሀ
	የመጠጥ ውሀ በዋናነት ከየት ነው የምትጠቀሙት?	2. [_]protected spring/well	2[_]Huxxinoonni buichcho	2[_]ከተከለለ የከርሰ ምድር ውሀ
		3. [_]tanker truck	3[_]Rottote giddo kuusanino waa	3[_]ከማጠራቀሚያ
		4. [_]public tap/standpipe	4[_]Olluu horons’rano 5[_]waa Gibbete giddo	4[_]ከህዝብ ቧንቧ
		5. [_]piped into dwelling	5[_]Waa Gibbete giddo	5[_]ግቢ ከገባ ቧንቧ

Part 3 child feeding practice
Kiiro 3 Qaaqqu sagalate gara
ክፍል 3 የህፃኑ አመጋገብ ሁኔታ

No	Question	Answer		መልስ
301	**Do you feed breast milk to your child?**			
	Daaima unuuna qansata?	1. [_] Yes	1. [_] Qanseema	1[_] አዎ
	ህፃኑን ዐሁን ጡት ታጠቢዋለሽ?	2. [_] No	2. [_]Diqaanseema	2[_] አላጠባውም
302	**If the mom is not breastfeeding now:** **Did you ever breastfeed your child?**			
	Ama unuunna qansitahakkiha ikkiro Qaaqqokk unuuna qansootta	1. [_] Yes	1. [_] Qanseema	1[_] አዎ
	እናትየው ዐሁን ጡት ማታጠባ ከሆነ ልጅሽን ጡት አጥብተሽው ነበር(በፊት)	2. [_] No	2. [_]Diqaanseema	2[_] አላጠባውም
303	**For how long did you breastfeed your child exclusively before giving any other feeds including water**			
	ዐጥብታው ከሆነ ለምን ያህል ጊዜ ነው ጡት ያጠባሽዉ (በወር ዐስቀምጭ/ጥ)			
	When did you start feeding your child with other foods apart from breast milk			
304	Wole segela karsitakinni unuuna calla mageeshi yanna qansita?			
	ለልጅሽ ምንም ምግብ ሳትሰጪ ውሀን ጨምሮ ለምን ያህል ጊዜ ነው ጡት ብቻ የሰጠሽው			
305	**When did you start feeding your child with other foods apart from breast milk**			
	Daaimaho sagale aameu dirin hanafita?			
	ለህፃኑ ተጨማሪ ምግብ በሰንት ወሩ ጀመርሽለት?			
306	**With what food do you start feeding your child?**	1[_] Cerial porrage	1[_]Gidu shirku	1[_]ከእህል ገንፎ
	Daaimikkira itisa hanafootta sagale maati?	2[_] Cow milk	2[_]Saddate adonni	2[_]በከብት ወተት
	ለህፃኑ ምግብ የጀመርሽለት በምንድን ነው?	3[_] Fruit juice	3[_]Muroteni huanlinoni	3[_]በፍራፍሬ ጭማቂ
		4[_] Other specify	4[_]Wole kuli	4[_]ሌላ ጥቀስ
307	**What is the number of meals normally taken by the child per day?**			
	Daaaimikki barrunni meu dani sagale itanno?\			
	ህፃኑ አሁን በቀን ምንያህል ጊዜ ነው የሚመገበው			
308	**Does child can feed himself/herself**			

	Daaimu sagale umisinni saga'la dandaanno?	1[_]Care giver	1[_]Manncholaatisannohu	1[_]ተንከባካቢ ያበላዋል
	ህፃኑ በራሱ መመገብ ይችላል?	2[_]With assistance	2[_]Irkotenniteitanohu	2[_]በዕገዛ ይበላል
		3[_]Self feeding	3[_]Umosit itannohu	3[_]ህፃኑ በራሱ ይበላል
	Who feeds the child?			
309	Daaimaho sagale ayi itisanno?	1. [_] Mother	1. [_] Ama	1[_] እናት
	ህፃኑን በቋሚነት ማን ነው የሚመግበው?	2. [_] Father	2. [_] Annu	2[_] አባት
		3. [_] Ant /Ancle	3. [_] Abbu/La'lama	3[_] እንጀራ እናት/አባት
		4. [_] grandfather /mother	4. [_] Ahahaahu/he	4[_] አያት
		5. [_] other specify	5. [_] Woleho xamisi	5[_] አህት/ወንድም
		6. [_] Do not know	6. [_] Diafoommo	6[_] አጉት/አክስት
310	**Does the child had food allergy**			
	Daaimaho lagaabbino sagale no?	1. [_] Yes	0. [_] Dinoe	0[_] አዎ
	ህፃኑ የማይስማማው የምግብ አይነት አለ?	2. [_] No	1. [_] Nooe	1[_] የለም
311 ዝለል Skip	**What is the food allergic for?**			
	Qaaqqoho (daaimaho) allifatowokki sagalete dani maati			
	ህፃኑ የማይሰማማው የምግበ አይነት ምንድን ነው?			
312 ዝለል Skip	**What was the manifestation of allergic condition?**			
	Allifatawokki sagala leelli shashanno malaati maati	1. [_] Rashes	1.[_]Hafuro	1[_] ሽፍታ
	የማይስማማዉን ምግብ ሲመገብ የሚያሳየው ምልክት ምንድን ነው?	2. [_] Vomiting	2.[_]Tushiishanno	2[_] ትውከት
		3. [_] Diarrhea	3.[_]Deeiishshanno	3[_] ተቅማጥ
		4. [_] Pain	4. [_]Xissannosi/se	4[_] ቁርጠት
		5. [_] Swelling	5. [_]Fuugisanno	5[_] እብጠት
		6. [_] Do not know	6. [_]Welere	6[_] ሌላጥቀሰ
313 ዝለል Skip	**What is the nutrition action taken for the allergic condition?**			
	Daaimu Lagaabbino sagale itiro woyyeessate uyitinanni sagale maati?			
	ለማይስማማው ምግብ ምልክት የተወሰደ እርምጃ ካለ ግለጪ			

Part 4: 24 Hour Dietary Diversity Questioner

Gaamo 4: 24 Sagalete danixamo

ክፍል:4:በ24 ሰአት ዉስጥ የወሰደውን የምግብ አይነት የሚገልጥ ጥያቄ

No	Type of food				
401	**I am going to ask you questions about what you fed your baby from the time you woke up yesterday morning till you woke up this morning either separately or combined with other foods.**				
	Bero soodo qoxootto wiinni kaitto yannanni hanaffe techo soodo geeshsha aante noo segalla giddo daaimaho itisootto sagale no? Itisoottaha ikkiro me’e higge itisoottoro kulattoe? Itisootto segale no ዐሁን ህፃኑ ከትላነት ጠዋት ዕስከ ዛሬ ጠዋት የተመገበውን የምግብ ዐይነት ዕጠይቅሻለሁ (ህፃኑ የተመገበውን ቁርስ ፣ምሳ ዕራት ዕንዲሁም በየመሀል የተመገበውን በመጠየቅ የሚስማማውን ቦታ ጥቀስ)				
402	**Did your child eat any porridge or gruel (from what it made)**				
	Daaimu sherko woy axmiite saga’lino (mayinni qixxeessinoonniha)?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ልጅሽ ከማንኛውም የእህል ዘር የተሰራ ገንፎ ወይም ሙቅ ተመግቧል?	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
403	**Bread, pasta, rice, noodles, biscuits, cookies or any other food made from ,oats, maize, barley, wheat, sorghum millet, or other grain? Specify**				
	Daabbo,paarta,ruuze,koshoro raino sagale woy ajjunni ,badalatenni ,hayixunni,qamadetenni,bashanqunniy loonsoonni sagale woy wolu quminni,xawisi	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ዳቦ፤ ፓስታ፤ ሩዝ፤ ብስኩት፤ ኩኪስ፤ ወይም ማንኛውም ነገር ከአጃ ከበቆሎ ገብስ፤ ሰንዴ፤ ማሽለ፤ወይም ሌላ አህል ዘር የተሰራ	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
404	**Any food made from teff , like injera ,kita or porridge ?**				
	Gaashetenni qqxxeessinoonnic segale buddeena, tima,woy sherko lawinore saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ማንኛውም ምግብ ከጤፍ የተሰራ (እንጀራ፤ ቂጣ፤ ገንፎ)	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
405	**Any white potatoes, white yam?**				
	Maxaaxeesla, diinicha, boyina,lawinore saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ማንኛውም ነጭ ድንች፤ ቦይና፤ እንሰት (ማንኛውም ነጭ ስራስር)	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
406	**Any foods made from beans, peas, lentils or pulses**				

	Aye segale baqeluuni, atarunni ,qibaatete qumma shumburunni qixxeessinoonni sagale saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ባቄላ፤አተር፤ ምስር ወይም ሌላ ጥራጥሬ	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
407	**Any nuts or seed such as peanut, sesame or sun flower seeds?**				
	Ayee qumma ocholoone coommadda gumma saga’lino	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ኦቾሎኒ (ከኦቾሎኒ የተሰራ ማንኛውም ምግብ)	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
408	**Any butter,oil**				
	Zayitetenni woy buurumi loonsoonni sagala	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	በዘይት ወይም በቅቤ የተሰራ ምግብ	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
409	**Any dark green, leafy vegetables like kale, spinach or amaranth leaves?**				
	Haanjarino, daraame ataakiltete daronna xu’naayye lawinore saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ማንኛውም ጥቁር አረንጓዴ አታክልት፤ ቅጠላማ አታክልት እንደ ጎመን፤ ራፎ ወይም ሌላ (ዘርዝር)	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
	Any pumpkin ,carrot, squash or sweet potatoes that are yellow or orange inside				
410	Giddo bica woy haanjirino baaqulaa, kaaroote woy maxaaxeesha saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ማንኛውም ዱባ፤ ካሮት ቢጫ ስኳር ድንች ሌላ (ዘርዝር)	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
411	**Any ripe mangoes, papayas?**				
	Le’ado mango woy paapaayya saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	የበሰለ ማንጎና ፓፓያ	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
412	**Any other fruit or vegetables**				
	Wole aye gumma woy akaakilte saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	ሌላ ማንኛውም አታክልትና ፍራፍሬ	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
413	**Commercially fortified foods.**				
	Ashshagantinota daaimu segale?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	
	በምግብ በልፅገው የሚሸጡ ምግቦች	1[_] Yes	1.[__]Saga’li no	1[_] ተመግ ቧል	
414	**Any cheese or yogurt?**				
	Ayibe/geinto saga’lino?	0[_] No	0.[__]Disaga ’lino	0[_] አልተመ ገበም	

	አይብና እርጎ	1[_] Yes	1.[__]Saga’lino	1[_] ተመግ ቧል	
	Frresh milk				
415	Iibbado ado	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	ትኩስ ወተት	1[_] Yes	1.[__]Saga’lino	1 [_] ተመግ ቧል	
	Any eggs?				
416	Ayee quuphe	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	እንቁላል	1[_] Yes	1.[__]Saga’lino	1[_] ተመግ ቧል	
	Any liver ,kidney, heart or other organ meats				
417	Afale,mule,wodana,woy wole godowu giddo maalla saga’lino?	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	ጉበት፤ ኩላሊት፤ ልብ ወይም የእንሰሳ የውስጥ ሰውነት ክፍል	1[_] Yes	1.[__]Saga’lino	1 [_] ተመግ ቧል	
	Any beef, pork, lamb, goat, rabbit (wild game meat such as antelope or deer)?				
418	Bootu maala, mancheemete maala, gereewo,mellenna hilleessa (wole dubbu saada,goljanna,guru’me lawinore segalino?	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	የበሬ ስጋ፤ የበግ/የፍየል ስጋ የአሳማ ስጋ የጥንቸል ወይም ሌላ እንስሳ	1[_] Yes	1.[__]Saga’lino	1[_] ተመግ ቧል	
	Any chicken ,duck or other birds				
419	Lukko, daakiyye, woy wole cea maala saga’lino	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	ዶሮ፤ እርግብ ወይም የወፍ ስጋ	1[_] Yes	1.[__]Saga’lino	1[_] ተመግ ቧል	
	Any fish				
420	Ayee qilxi’me	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	ማንኛውም አሳና የአሳ ምርት	1[_] Yes	1.[__]Saga’lino	1[_] ተመግ ቧል	
	Any soft drink specify				
421	Ayee shota agatto agino? Aginoha ikkiro xawisi	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	ማንኛውም ለስላሳ መጠጦች	1[_] Yes	1.[__]Saga’lino	1[_] ተመግ ቧል	
	Coffee and Tea				
422	Buna woy shae agino	0[_] No	0.[__]Disaga’lino	0[_] አልተመ ገበም	
	ቡናና ሻይ	1[_] Yes	1.[__]Saga’lino	1[_] ተመግ ቧል	
423	Alcoholic drink				

	Diribisanno ago (birra,xesiixella,areqe)	0[_] No	0.[__]Disaga 'lino	0[_] አልተመ ገበም	
	አልኮል መጠጦች (ቢራ፤ አረቄ፤ ጠጅ፤ጠላ)	1[_] Yes	1.[__]Saga'li no	1[_] ተመግ ቧል	
424	**Other specify**				
	Welere				
	ሌላ ካለ ጥቀስ				

Part 5: 7-Days Food frequency questionnaire for iron rich foods
Sufetto 5: 7 barri giddo ayirenetenni lattino sagale marri marro
ክፍል 5፤በ7ት ቀን ውስጥ ህጻኑ የወሰደው የምግብ አይነት

		Answer the number of the day you eat
No	**Question**	**መልሱን በቀናት ቁጥር ዐስቀምጥ**
501	**Now I am going to ask you if you gave the following items at all the last week ending yesterday morning. Please answer yes if you gave it and no if you did not give it And if you did, will you please tell how many times you gave it**	
	Xa xa’meemohehu sa’u lamala jeefonni kayise be’ro soodo geeshsha saga’lino sagaleeti.itinno sagale “saga’lino” itinnoki sagale ’’di’saga’lino’’ yite me’e marro itinoro xawasi.	
	እሁን የምጠይቅሽ ልጅሽ ባለፉት 7ቀን ውሰጥ አሰከ ዛሬ ጠዋት የበላውን የምግብ አይነት ነው ለአያንደንዱ መልሰ ቁጥሩን ግለጪ ካልተመገበ ዜሮ መፃፍን አትርሳ(ሺ)	
502	**Food made from false banana (Kocho,kita,bula,omolicho,genfo)**	
	Inset?	
	እንሰት(ቆጮ፣ ቡላ...)?	
503	**Cereal group (maize, barley, wheat, oats, ….)**	
	Weese (badala,hayiixe	
	የእህልዘር(በቆሎ፤ ገብስ፤ ስንዴ፤ አጃ፤ ዳጉሳ፤.......	
504	**Pulse group (bean,pea,chickpea,….)**	
	Qixxeessinoonni (baqeluuni, atarunni ,qibaatete qumma shumburunni?)	
	የጥራጥሬ ዘር(አተር፤ ሽንብራ፤ ባቄላ፤ምስር……)	
505	**Teff (ingera,bred,porrage)**	
	Gaashe	
	ጤፍ	
506	**Peanut**	
	Ocholoone	
	ኦቾሎኒ	
507	**Dark green vegitables (Kale,green paper,qosta)**	
	Haanja daro ataakilte shaana, qaariya,Xu’naaye, raafote daro	
	ጥቁር አረንጎዴ አታክልቶች(ጎመን፤ ቃረያ፤ ጥቁር ጎመን፤ ራፎ ……..)	
508	**Tomato, Carrot, watermelon, pumpkin**	
	Timaatime,karoote, Baaqula	
	ቲማቲም፣ ካሮት፣ ዱባ	
509	**ripe mango, papaya**	
	Lino mango,Lino pappaayya	
	የበሰለ ማንጎ፤ የበሰለ ፓፓያ	

510	**orange, lemmon,**	
	Burtukanenna loome	
	ብርቱካንና ሎሚ	
511	**Amarnth leaf or grain**	
	Raffote daronna guma	
	የራፎ ቅጠል ወይም ፍሬ	
512	**Milk and milk product**	
	Ado geinto burbuxxo	
	ወተት፣ እርጎ፣ አይብ	
513	**Any food contains butter or oil**	
	Zayitetenni woy buurumi loonsoonni sagala	
	በዘይት ወይም በቅቤ የተሰራ ምግብ	
514	**Egg**	
	Quuphphe	
	እንቁላል	
515	**Red meet (sheep, goat, ox)**	
	Dummo maala (bootunniha meichchunniha, gerechch unniha)	
	ቀይ ስጋ (የበሬ፣ የፍየል፣ የበግ)	
516	**Chicken meet**	
	Lukkichchu maala	
	የዶሮ ሰጋ	
517	**Fish**	
	Qulxume	
	አሳ	
518	**Internal organ (liver, kidney, heart)**	
	Giddoodi mannimma gaamo maalal fale(kulalitite, wodana)	
	የውሰጥ የሰውነት ክፍል ሥጋ (ጉበት፣ ኩላሊት፣ልብ)	
519	**Coffe or tea**	
	Shaenna buna	
	ሻይ ወይም ቡና	
520	**Soft drink**	
	Shaffado ago	
	ለስላሳ መጠጦች	
521	**Any alchol**	
	Kajjado ago	
	ዐልኮል መጠጦች	
522	**Others specify**	
	Wole kunni assi	
	ሌላ ካለ ግለፅ/ጪ	

Part VI Supplementation question

Kifile 6 Ledishu sagale xa'mo

ክፍል 6፤ ተጨማሪ ምግብ ስለመውሰድ የሚገልጽ መጠይቅ

	Question			Code
601	**Do you feed the child any foods made with oil, fat or butter?**			
	Zayite, buuronna,coomu sagale saga'lano?	0. [_] No	0.Disagalino[__]	0[__]አይደለም
	ልጅሽን ዘይት፤ ጮማ ወይም ቅቤ ያለበትን ምግብ ትመግቢዋለሽ	1. [_] Yes	1.Sagalino[__]	1[__]አዎን
	If yes for question number 601 do you feed every day			
602	Barrunni daaimu me'e marro saga'lano	0. [_] No	0.Disagalino[__]	0[__]አይደለም
ዝለል Skip	ከመገብሽው በየቀኑ ትመግቢዋለሽ	1. [_] Yes	1. Sagalino[__]	1 [__]አዎን
603	**Has your child ever received Iron supplementation**			
	Daaimu ayirenete lattinota ledishu sagale afiranno?	0. [_] No	0. Disagalino[__]	0[__]አይደለም
	የደም መሙያ አንክብል ወሰዶ ያውቃል?		1. Sagalino[__]	1[__]አዎን
	If yes for question number 603 Why he or she received?			
604	19kki xamo saga'lanoha ikiro Mayirra sagalanno?			
ዝለል skip	የደም መሙያ አንክብል ከወሰደ ለምንድን ነው የወሰደው			
	From where do you get?			
605	Maminni afidhinonni?	1.[_] From health institution	1[_]Fayyimateuur ishinni	1 [_]ከጤና ተቋም
ዝለል skip	የደም መሙያ አንክብል ከየት ነው የተሰጠው?	2.[_] Food aid program	2[_]Sagallte qixawonni	2 [_]ከምግብ ፕሮግራም
		3.[_]Pharmacy	3[_]Farmasetenni	3 [_]ፋርማሲ
		4. [_]Other specify	4[_]Wolekuli	4 [_]ሌላጥቀስ
606 ዝለል Skip	**Does the child taking now?**			
	Qaaqqu xaano mundee abbitawoota xagicho adhayno?	0. [_] No	0[__].Disagalino	0[__]አይወስድም
	ህፃኑ አሁንም የደም መሙያ እንክብል እየወሰደ ነው?	1. [_] Yes	1[__].Sagalino	1[__]አዎ
607 ዝለል Skip	**For how long he /she take supplementation?**			
	Ladishshu sagale mageshi geeeshsha sagalonno?			
	የደም መሙያ ዕንክብል ከወሰደ ለምን ያህል ጊዜ ወሰደ በወራት ጥቀሽ/ስ?			

608	**Has your child ever eat iron fortified food**			
	Daaimikki Ayiirenetenni kaajjinshoonni sagle sagalanno?	0. [_] No	0[__]. Disagalino	0[__]አይወስድም
	ህፃንሽ ደምን በሚሞላ የብረት ንጥረ ነገር የበለፀገ ምግብ በልቶ ያውቃል?	1. [_] Yes	1[__]. Sagalino	1[__]አዎ
609 ዝለል Skip	**If yes for question number 608**			
	From where do you get			
	Hiikiinni afiranno?	1.[_] From health institution	1[_]Fayyimateuur ishinni	1[_]ጤናተቋም
	ከበላ ከየት ነው ያገኘሽው?	2.[_] Food aid program	2[_]Sagallte qixawonni	2[_]የምግብ ኘሮ ግራም
		3.[_]Pharmacy	3[_]Farmasetenni	3[_]ፋርማሲ
		4. [_]Other specify	4[_]Wolekuli	5[_]ሌላ ካለ (ዘርዝር/ሪ)
610 ዝለል Skip	**Is the child taking now?**			
	Mamoote saga'lanno?	0. [_] No	0[_]Disagalino	0[_]አይደለም
	አሁንም ህጻኑ እየተመገበ ነው(ዕየተመገበ ከሆነ ዐሳዪኝ)?	1. [_] Yes	1[_]Sagalino	1[_]አዎን
	What was the food?			
611 ዝለል Skip	Sagale maati?			
	የሚወስደው የበለጸገ ምግብ ምን ነበር(ያሳየችህን የምግብ ዐይነት ፃፍ?)			
612 ዝለል Skip	**For how long she/he take?**			
	Mageeshshi geeshe saga'lano?			
	የበለጸገ ምግቡን ለምን ያህል ጊዜ ወሰደ(ወሰደች) በወራት ጥቀሽ/ስ?			
613	**Has the child ever been given a Vit A capsule?**			
	Daaimu vitaamine A kinine afire egennino?	0. [_] No	0. [_]Disagalino	0. [_]አያውቅም
	ህፃኑ የቫይታሚን A እንክብል ወሰዶ ያውቃል?	1. [_] Yes	1. [_]Sagalino	1. [_] አዎ
614 ዝለል Skip	**If yes for question no 613 how often did he/she get**			
	kki xa'mo afirinnoha ikkiro ,me'e marro afirino?			
	ቫይታሚን A ከወሰደ በአመት ምን ያህል ግዜ ያገኛል?			
615	**Since you were pregnant Have you taken any Iron supplements?**			
	Godowii heedhe ayiirenete lattino sagale afiroota?	0. [_] No	0. [_]Disagalino	0. [_]አያውቅም
	እርጉዝ ሆነሽ ደም የሚሞላ እንክብል ወስደሽ ታውቂያለሽ?	1. [_] Yes	1. [_]Sagalino	1. [_] አዎ
616 ዝለል Skip	**If yes for question number 21 for how long did you taken the supplement**			
	21kki xa'mo afirootana ikkiro mageeshi geeshsha afiroota?	1. [_] Less than month	1. [_] 1 agani woro	1 [_] ከአንድ ወር በታች

	ደም የሚሞላ ዕንክብል ከወሰድሽ ለምን ያህል ጊዜ ነው የወሰድሽው?	2. [_] 1 up 2 month	2. [_] 1-2 agani geesha	2. [_] 1-2ወር
		3. [_] 2up3month	3. [_] 2-3 agani geesha	3. [_] ከ2-3 ወር
		4. [_] More than 3 month	4. [_] 3 apani ale	4. [_] ከ3 ወር በላይ
		5. [_] For 6 month	5. [_] 6 adani geesha	5. [_] ለ6ወር
		6. [_] Do not know	6. [_] Diafooma	6. [_] ዐላውቅም

Part 7. Baby health question

Kifile 7 Daaimu fayimu xa'mu

ክፍል 7 የህፃኑን ጤና የሚገልፅ መጠይቅ

Now I am going to ask you questions which are related to your baby's health				
Xa xa'meemohehu daa'mu fayima ledo xedooshu noo xa'mooti				
ዐሁን የህጻኑን ዐጠቃላይ የጤና ሑኔታ የሚገልጽ ጥያቄ ዕጠይቅሻለሁ				
	QUESTION		**CODE**	
701	**Does the baby sleep in your bed?**			
	Daaimu ate daalasira goxanno?	0. [_] No	0. [_]Dee'ni	0. [_]አይ
	ህፃኑ ከአንቺ ጋር ነው የሚተኛው	1. [_] Yes	1. [_]Eewa	1. [_]አዎ
702	**Do you use bed net to your child?**			
	Daaimu goxenno woyite agobere horonsiranno?	0. [_] No	0. [_]Dee'ni	0. [_]አይ
	ህፃኑ በአጎበር ተሸፍኖ ነው የሚተኛው	1. [_] Yes	1. [_]Eewa	1. [_]አዎ
703	**Does the child vaccinated?**			
	Daaimu kittibaate garunni gudino?	0. [_] No	0. [_]Dee'ni	0. [_]አይ
	ህፃኑ ክትባት ወስዷል?	1. [_] Yes	1. [_]Eewa	1. [_]አዎ
704	**Does the child had vaccine card?**			
	Kittibaate gudino kaarde noosi?	0. [_] No	0. [_]Dee'ni	0. [_]አይ
	ህፃኑ የክትባት ካርድ አለው?	1. [_] Yes	1. [_]Eewa	1. [_]አዎ
	Could you show me vaccine card (please find which vaccine the child take from the card?)			
705	BCG kitibaate dasirino anga lea hasireemona lao?	[_]BCG?	[_]BCG?	[_]የሳንባ ምች
	Dasirinohu qinniticho angara dasahootic?	[_]Polio 0?	[_]Polio 0?	[_]የህፃናት ልምሻ 0
		[_]Polio1?	[_]Polio1?	[_]የህፃናት ልምሻ 1
	የክትባቱን ካ ርድ ታሳዩኛለሽ (በክትባት ካርድ ላይ የወሰደውን X ያልወሰደውን 0 አድርግ/ጊ)	[_]Polio2?	[_]Polio2?	[_]የህፃናት ልምሻ 2
		[_]Polio3?	[_]Polio3?	[_]የህፃናት ልምሻ 3
		[_]DPT- 1	[_]DPT- 1	[_]ዲፒቲ 1
		[_]DPT- 2	[_]DPT- 2	[_]ዲፒቲ 2
		[_]DPT- 3	[_]DPT- 3	[_]ዲፒቲ 3
		[_]PCV1	[_]PCV1	[_]ፒሲ ቪ1
		[_]PCV2?	[_]PCV2?	[_]ፒሲ ቪ 2
		[_]PCV3?	[_]PCV3?	[_]ፒሲቪ3
		[_]Rota1?	[_]Rota1?	[_]ሮታ 1
		[_]Rota2?	[_]Rota2?	[_]ሮታ 2
		[_]Measealse	[_]Measealse	[_]ኩፍኝ

706	**Can I see the BCG scar please?**			
	BCG kitibaate dasirino anga lea hasireemona lao?	1.[_] BCG – lesion seen:	1.[_] BCG- bassa leellanno	1 የክትባቱ ጠባሳ ይታያል
	Dasirinohu qinniticho angara dasahootic	2.[_] BCG – lesion not seen	2.[_] BCG- bassu dileellanno	2 የክትባቱ ጠባሳ አይታይም
	የሳንባ ነቀርሳ ክትባት ቦታውን ማየት እችላለው?			

Part 7.1 Morbidity,

Diarrhea episode two weeks recall

ክፍል 7.1: ባለፉት 2 ሳምንታት ስለ ህፃኑ የተቅማጥ ህመም መጠይቅ

	QUESTION		Code	
707	**During the last two weeks that ended yesterday morning, did the child have diarrhea?**			
	Sau leme lamalanni henafe bero soodo geeshsha deuu malaati noosi?	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	ባለፉት 2 ሳምንታት ህፃኑ የተቅማጥ (የሆድ) በሽታ አሞት ነበር?	1. [_] Yes ↓	1. [_]Noosi	1. [_]አዎ
708 ዝለል Skip	**Did the child pass any watery stools?**			
	Waa lawanno deiinoosi?	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	ህፃኑ ውሀ የሚመስል ተቅምጥ አሰቀምጦታል	1. [_] Yes↓	1. [_]Noosi	1. [_]አዎ
709 ዝለል Skip	**The day had most loose or watery stools, how many loose or watery stools did pass?**			
	Wayi gedee deoo deeanni hoosino barri no?			
	ብዙ ባሰቀመጠው ቀን ምን ያህል ጊዜ አሰቀመጠው?			
710 ዝለል Skip	**Did any of the stools contain blood?**			
	Daaimu sagari aana mundee karsantewo?	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	በተቅማጡ ውስጥ ደም ታይቷል	1. [_] Yes ↓	1. [_]Noosi	1. [_]አዎ
711 ዝለል Skip	**Were the stools of different consistency than before fell ill with diarrhea?**			
	Deeiishanno yannara wole yanawiinni baxxitino xisso maciishshenno?	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	ሲያሰቀምጠው ህመም ይሰማው ነበር	1. [_] Yes ↓	1. [_]Noosi	1. [_]አዎ
712 ዝለል Skip	**Did the illness interfere with ability to drink or eat?**			
	Sagalanno woyiitena wala aganno wayite xisso maciishantennosi?	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	ህመሙ ምግብ እንዳይበላ ያደርገው ነበር	1. [_] Yes ↓	1. [_]Noosi	1. [_]አዎ
713 ዝለል Skip	**Did you seek treatment for?**			
	Moyyessate wonaaloo?	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	ለህመሙ የህክምና እርዳታ ፈልገሽ ነበር?	1. [_] Yes ↓	1. [_]Noosi	1. [_]አዎን
714 ዝለል Skip	**Was the child admitted to a hospital?**			
	Daaimu hospitalete goxino?	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	ለህመሙ ሆስፒታል ተኝቶ ነበር?	1. [_] Yes ↓	1. [_]Noosi	1. [_]አዎን
715 ዝለል Skip	**How many days did the diarrhea last?**			
	Deuu meu barri geisha keeshino?			
	ተቅማጡ ለምን ያህል ጊዜ ቆየበት/ባት(በቀናትጥቀስ/ሽ)			

716 ዝለል Skip	**During this period of illness you have described, did you change the way you were feeding your child in any way?**			
	Dhibbu yannara hitto garinnit sagale itisatahu	1. [_] More often	1[_] albininniraha raha	[_]1. ከበፊቱ ቶሎ ቶሎ
	በህመሙ ጊዜ በምን አይነት መልኩ ነበር የምትመግቢው?	2. [_] less seldom than before the illness started.	2[_]Albinni asishi	[_]2. ከበፊቱ ቀንሼ
		3. [_] Did not change feeding frequency	3[_]Albinni Soorro dino	[_]3.ከበፊቱ ለውጥ የለውም

Part 7.2. Malaria 3 month recall

kifille 7.2. 3Aganni mereero sheekerete xisso

ክፍል7.2.ላለፉት 3 ወር ውስጥ ስለ ህፃኑ የወባ ህመም ሁኔታ የሚያመላክት መጠይቅ

	QUESTION		**CODE**	መለያ
717	**During the last three months that ended yesterday morning, did the child have fever?**			
	Sau 3 aganninni kayise bero soodo geshsha daaimu biso iibbabbino	0. [_] No	0. [_]Dinosi	0.[_]አይደለም
	ላለፉት 3 ወራት ልጅሽ ትኩሳት አሞት ያውቃል	1. [_] Yes	1. [_]Noosi	1. [_]አዎን
718 ዝለል Skip	**Did you brought the child to health institute**			
	kki xa'mo "iibbabinno" ikkituro daaimu me'e marr iibbabbino?			
	ትኩሳት ካመመው ለምን ያህል ጊዜ ደጋግሞ ዐመመው?			
719 ዝለል Skip	**Did you brought the child to health institute**			
	Daaimakki fayyimmate uurrinsha massooto	0. [_] No	0. [_]Dinosi	0.[_]አይደለም
	ሀኪም ቤት ወስደሽው ነበር?	1. [_] Yes	1. [_]Noosi	1. [_]አዎን
720 ዝለል Skip	**If yes what was the illness**			
	Massootthoa ikkiro xissosi dani maati			
	ከወሰድሽው በሽታው ምን ነበር			
721 ዝለል Skip	**During this period of illness you have described, did you change the way you were feeding your child in any way?**			
	Daaima xissosi sagale saga'lannoki pedenna agannokki gede assitinosi?	0. [_] No	0. [_]Dinosi	0.[_]አይደለም
	ህፃኑን በሽታው ከመብላት ከመጠጣት ከልክሎት ነበር?	1. [_] Yes	1. [_]Noosi	1. [_]አዎን
722 ዝለል Skip	**Was the child admitted to a hospital for the illness?**			
	Daaimu xissame hospitalete aoxino?	0. [_] No	0. [_]Dinosi	0.[_]አይደለም
	ህፃኑ ባመመው ትኩሳት ምክንያት ሆስፒታል ተኝቶ ነበር	1. [_] Yes	1. [_]Noosi	1. [_]አዎን
723 ዝለል Skip	**During the period of illness did you feed your baby more often, more seldom than or just as often as before the illeness started?**			
	Saggala hiitto daagonni Soorritta	1. [_] More often	1[_]Albinin niraha raha	1[_]ከበፊቱ ቶሎ ቶሎ
	በህመሙ ጊዜ አመጋገብ እንዴት ባለ መንገድ ቀየርሽ	2. [_] More seldom than before the	2[_]Albinni asishi	2[_]ከበፊቱ ቀንሼ

		illness started		
		3. [_] Did not change feeding frequency.	3[_]Albinni Soorro dino	3[_]ከበፊቱ ለውጥ የለውም

Part 7.3: Pneumonia 2 Week recall

Kifille 7.3: Lemala mereero saambu michche xisso

ክፍል 7.3 ላለፉት 2 ሳምንታት የሳንባ ምች ህመምን የሚያመላክት መጠይቅ

	QUESTION		**CODE**	
724	**During the last two weeks that ended yesterday morning, did the child have cough?**			
	Sau lame lemale kayisse be'ro soodo geeshsha, Daaimu Buusano Afirino?	0. [_]No	0. [_]Dinosi	0. [_]አይደለም
	ባለፉት 2 ሳምንታት ህፃኑ ሳል አሞት ነበር ?	1. [_]Yes	1. [_]Noosi	1. [_]አዎን
	During the last two weeks that ended yesterday morning, did the child have fast or difficult breathing?			
725 ዝለል Skip	Sau lemalaa kayiise be'ro soodo geeshsha. Daaimu rahe rare fo'lanno woy foo'late rakkatanno?	0. [_]No	0. [_]Dinosi	0. [_]አይደለም
	ላለፉት 2 ሳምንታት እስከ ትላንት ጠዋት ህፃኑ ትንፋሽ ያጥረው ነበር ?	1. [_]Yes	1. [_]Noosi	1. [_]አዎን
726 ዝለል Skip	**Did the illness interfere with the child ability to drink or eat?**			
	Xisso ittennona agannoki gede rakkatisannosi?	0. [_]No	0. [_]Dinosi	0. [_]አይደለም
	የህመሙ ሁኔታ መብላትና መጠጣት ዕንዲያስቸግረው ዐድርጎበት ነበር?	1. [_]Yes	1. [_]Noosi	1. [_]አዎን
727 ዝለል Skip	**Was the child admitted to a hospital for the illness?**			
	Daaimu xissame hospiitalete goxinno?	0. [_]No	0. [_]Dinosi	0. [_]አይደለም
	ህፃኑ በህመሙ ምክንያት ሆስፒታል ተኝቶ ነበር?	1. [_]Yes	1. [_]Noosi	1. [_]አዎን
728 ዝለል Skip	**During the period of illness did you feed your baby more often, more seldom than or just as often as before the illness started?**			
	Konni dhibbinni daaimakki hiittoonniti itisaffahu	1. [_] More often	1. A lbininniraha raha	1[_] ከበፊቱ ቶሎ ቶሎ
	በዚህ ህመም ህፃንሽን እንዴት ነበር የምትመ ግቢው?	2. [_] More seldom than before the illness started	2. Albinni asishi	2[_] ከበፊቱ ቀንሼ
		3. [_] Did not change feeding frequency.	3. Albinni Soorro dino	3[_]ከበፊቱ ለውጥ የለውም

Part 7.4:Hospitalizations
Kiffile 7.4:Hospiitaalete goxinno yanna
ክፍል 7.4:የህፃኑ ሆስፒታል የመተኛት ሁኔታ

	QUESTION			CODE
729	**Since birth has ever been admitted to hospital?**			
	Daaimu ilami yanaa henafe hospiitalete goxe egennino	0. [_] No	0. [_]Dinosi	0. [_]አይደለም
	ህፃኑ ከተወለደ ጀምሮ ሆስፒታል ተኝቶ ያውቃል?	1. [_] Yes ↓	1. [_]Noosi	1. [_]አዎን
730 ዝለል Skip	**How many times has been admitted to hospital?**			
	Hospitaalete me'e marro goxino?			
	ሆስፒታል ለምን ያህል ጊዜ ደጋግሞ ተኝቶዐል?			
731 ዝለል Skip	**What was the maximum days admited t hospital**			
	Barra ikkanno hospitalete seeda yanna goxinohu mee barra ikkanno			
	በሆስቲታል ለረጅም ጊዜ የተኛው ምን ያህል ቀን ነው?			
	What was the reason was in the hospital each time:			
732 ዝለል	Hospiitaalete mayi xinsoonnit	1. [_] 1st	1. [_] 1st	1. [_] 1st
	ሆስፒታል የተኛበት ምክንያት ምን ነበር ለእያንደንዱ ከሚከተሉት ምርጫ አሰቀምጥ/ጭ	2. [_] 2nd	2. [_] 2nd	2. [_] 2nd
	1. Malaria/ወባ/wobahoni	3. [_] 3rd	3. [_] 3rd	3. [_] 3rd
	2. Pnemonia/የሳንባምች/sanbamich	4. [_] 4th	4. [_] 4th	4. [_] 4th
	3. gastrointeritis/ተቅማጥ/gadawa gameetini	5. [_] 5th	5. [_] 5th	5. [_] 5th
	4. any respiratory disease/ማንኛውም የመተንፈሻ አካል በሽታ/ayee foolate bisishibaah	6. [_] 6th	6. [_] 6th	6. [_] 6th
	5. ሌለ ጥቀሰ/woleri kuli	7. [_] 7th	7. [_] 7th	7. [_] 7th
		8. [_] 8th	8. [_] 8th	8. [_] 8th

Part 8.1: Mother knowledge on Anaemia

Kiffille 8.1 Aneemiyu (mundeete anje) aana amate no huwato

ክፍል 8.1. በደም ማነስ ህመም ላይ የናትየው ዕውቀት

No	**Question**			**Remark**
	Did you ever heard about anaemia			
801	Aneemiyu xisso maatiro macciishite egennota?	0.[_] No	0.[_] Dee'ni	0. [_]አይደለም
	ስለ ደም ማነስ ሰምተሸ ታውቂያለሽ?	1.[_] Yes	1.[_] Eewa	1. [_]አዎን
802 Skip ዝለል	**If yes for question number what does it mean?**			
	Aneemiya yaa mayaate?			
	ሰምተሸ ካወቅሽ ምን ማለት ነው?			
803 Skip ዝለል	**How can anaemia are prevented**			
	Aneemiyu xisso hiitoonni gargadhinanni?			
	የደም ማነስ እንዴት መከላከል ይቻላል?			
804 Skip ዝለል	**Do you think anaemia is sever problem**			
	Aneemiyu lowo geeshsha gawajjanota huwatootta?	0.[_] No	0.[_] Dee'ni	0. [_]አይደለም
	የደም ማነስ ከባድ የጤና ቸግር ነው ብለሽ ታስቢያለሽ	1.[_] Yes	1.[_] Eewa	1. [_]አዎን
805 Skip ዝለል	**If no what is your reason**			
	Huwatootta ikkiha ikkiro, korkaatu maati?			
	ከባድ ነው ብለሽ ካልሽ ምክንያትሽ ምንድን ነው?			
806 Skip ዝለል	**What action you will did if you know your child is anemic**			
	Daaimikki aneemiyu xissonni amadamiro adhatta qaafo maati?			
	ህፃንሽ በደም ማነስ ቢታመም ምን ታደርጊያለሽ?			

Part 8.2 Mother knowledge on iron rich food

Kiffile 8.2 ama/lossaancho ayiirene guuttino sagale aana nose huwato

ክፍል 8.2.የናትየው ዕውቀት በብረት የበለፀጉ ምግቦች ላይ

No	Question			
807	**Have you ever heard about iron rich food if yes go to question number 808**			
	72[kki] Kiiro aana ayyirene guutino sagale macciishootta?	0.[_] No	0.[_] Dee'ni	0. [_]አይደለም
	በብረት ስለበለፀጉ ምግቦች ሰምተሽ ታውቂያለሽ?	1.[_] Yes	1.[_] Eewa	1. [_]አዎን
808 Skip ዝለል	**From where did you hear about iron rich food?**			
	Ayirene guuttino sagale mamminni macciishitta?	1.[_]Health staff	1[_] Fayimmate loosasinewinni	[_]1. ከጤና ባለሞያ
	ካወቅሽ ከየት ነው የሰማሽው?	2.[_]Radio	2[_] Raddonetenni	[_]2. ከብዙሀን መገናኛ
		3.[_]NGO	3[_] Mangistawe ikkitinokki uurrinshanni.	[_]3.መንግስታዊ ካለሆኑ ድርጅቶች
		4.[_]Family	4[_] Maate'yawiinni	[_]4.ከቤተሰብ
		5.[_]Don't know	5[_] Diafoomma	[_]5.አላውቅም
		6. [_]Other (specify)	6[_] Wolewiinni (xawisi)	[_]6.ሌላ ግለፅ
809 Skip ዝለል	**Can you list some of iron rich food?**			
	Ayirene guuttino sagale hiikkoreetiro kulattaera dandaatta?			
	በብረት ንጥረ ነገር የበለፀጉ ምግቦችን ጥቀሺ?			
810 Skip ዝለል	**When taken during meals, certain foods help the body absorb and use iron. What are those foods?**			
	Hurbaaxinnemmo yannara bissinke ayirene raha horonsi'ranno gede asisitanno sagale hikkoreeti?			
	አንዳንድ ምግቦች ከምግብ ጋር ዐብረው ሲወሰዱ የብረት ንጥረ ነገር በቀላሉ ሰውነታችን እንዲጠቀመው ይረዳሉ እነዚህ ምግቦች ምንድን ናቸው?			
811 Skip ዝለል	**Some beverages decrease iron absorption when taken with meals. Which ones?**	1.[_]Tea	1[_] Shae	1[_]ሻይ
	Mite mite agatto sagalete ledo anganni woyiite ayiirene ajanno gede asitannori hiikkoreeti?	2.[_]Coffee	2[_] Bunu	2[_]ቡና
	አንዳንድ መጠጦች የብረት ንጥረ ነገር ሰውነታችን ዕንዳይጠቀም ይከለክላሉ ዕነዚህ የምግብ ዐይነቶች አነማን ናቸው?	3.[_]Other specify	3[_] Diafoomma	3[_]አላውቅም
		4.[_]Don't know	4[_] Wolere (xawisi)	4[_]ሌለ ጥቀሰ

812 Skip ዝለል	**How good do you think it is to prepare meals with iron-rich foods such as beef, chicken or liver?**			
	Ayirene guuttino sagale danchu garinni hiitto qixxesinanniro afootta? Lawishaho bootu maala,lukkichu maalanna salto lawinore.	1.[_]Not good	1[_] Didanchaho	1[_]ጥሩ አይደለም
	በብረት የበለፀጉ ምግቦቸን ማለትም የበሬ ስጋ የዶሮ ሰጋና ጉበት፤ ኩላሊት ከህፃናት ምግብ ጋር ቀላቅሎ መስራት ጥሩ ነው ብለሽ ታስቢያለሽ?	2.[_]Good	2[_] dibbuxomma.	2[_]ጥሩ ነው
		3.[_]You're not sure	3[_]Danchaho	3[_]እርግጠኛ አይደለሁም
		4.[_]I don't know		4[_]አላውቅም
813 Skip ዝለል	**If it is not good. Can you tell me the reasons why it is not good?**			
	Dancha ikkinokiha ikkiro:Dancha ikkinokki Korkaati maati?			
	ጥሩ ካልሆነ ምክንያትሽ ምንድን ነው			
814 Skip ዝለል	**If it is not good. Can you tell me the reasons why it is not good?**			
	Dancha ikkinokiha ikkiro:Dancha ikkinokki Korkaati maati?			
	ጥሩ ከሆነ ምክንያትሽ ምንድን ነው			
815 Skip ዝለል	**How difficult is it for you to prepare meals with iron-rich foods?**	[_]Not difficult	[_] injaannoe	1ከባድ አይደለም
	Dancha ikkinokiha ikkiro: Dancha ikkinokki Korkaati maati?	[_]So-so	[_] hageeshi geeshaati.	2ከባድ ነው
	ላንቺ በብረት የበለፀጉ ምግቦችን ማዘጋጀት ምን ያህል ከባድ ነው?	[_]Difficult	[_] Di injannoe	3በጣም ከባድ ነው
816 Skip ዝለል	**If difficult. Can you tell me the reasons why it is difficult?**			
	Injaannohe ikkiha ikkiro: Korkaatu maati?			
	ከባድ ከሆነ ምክንያቱ ምንድን ነው			
817 Skip ዝለል	**If it is not difficult. Can you tell me the reasons why it is difficult?**			
	Injaannohe ikkiha ikkiro: Korkaatu maati?			
	ከባድ ካልሆነ ምክንያትሽ ምንድን ነው			

Part 9: Household Food Insecurity Access Scale (HFIAS) Measurement:
Now I would like to ask you few questions regarding your household food security situation in the past four weeks.
ክፍል 9 ላለፉት 4 ሳምንታት የቤተሰቡን የምግብ ዋስትና የሚያሳይ መጠይቅ

Rule: We, will not give "timing" instructions and force answers. Reported frequency in correct category according to this list: 0. No/never, 1. Sometimes: 1-2 days last month, 2. Often: 3-10 days/month, 3. Very often/usually: More than 10 days last month		በሚከተለው ገለፃ መሰረት የዕናትየውን መልስ ዐስቀምጥ/ጪ □1ወ ይም 2 ጊዜ ላለፉት 4 ሳምንታት (ጥቂት ጊዜ) □ከ3 -10 ጊዜ ላለፉት 4 ሳምንታት (ዐንዳንድ ጊዜ) □ከ10 ጊዜ በላይ ላለፉት 4 ሳምንታት (ብዙ ጊዜ)		
ተ/ቁ	**ጥያቄ (questions)**			
901	**In the past four weeks, did you worry that your household would not have enough food?**			
	Sau shoole lamalara miniki manni ikkado sagala afirewokkhura qarranten egenootta	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት ዐራት ሳምንታት ለቤተሰብሽ በቂ ምግብ ካለመኖሩ የተነሣ ተጨንቀሽ ታውቂያለሽ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
	How often did this happen in the past four weeks?			
902 skip ዝለል	Qarrante egendottara shoole lamala giddo mageeshshi yanna ikkanno	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda yanna	[_]3. ብዙ ጊዜ
	In the past four weeks, were you or any household member not able to eat the kinds of foods you preferred because of a lack of resources?			
903	Sau shoole lamalara hidha hoogate kainohunni ati woy minikki maate sagala hasidhinoonni sagalete dano ita hooggine egentinoonni	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት ዐራት ሳምንታት መግዛት ካለመቻል የተነሳ ዐንቺ ወይም ከቤተሰብሽ ዐባላት መብላት የፈለጋችሁትን የምግብ ዐይነት ሳትመገቡ ቀርታችሁ ታውቃላችሁ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
904 Skip ዝለል	How often did this happen in the past four weeks?			
	Hoogginihunni mageeshshi barra ikkanno xaadenehu	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ

	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ
	In the past four weeks, did you or any household member have to eat a limited variety of foods due to a lack of resources?			
905	Sau shoole lamala hoongunni kainohunni at way minikki maate mitte bikka callo sagalete dana sagalitinoonni	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት ዐራት ሳምንታት መግዛት ካለመቻል የተነሳ ዐንቺ ወይም ከቤተሰብሽ ዐባላት ውስጥ ዐንድ ዐይነት ምግብ ብቻ ተመግባችሁ ታውቃላችሁ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
906 Skip ዝለል	**How often did this happen in the past four weeks?**			
	Ikkiro sau shoole lamalara mageeshshi geeshshooti	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ
907	**In the past four weeks, did you or any household member have to eat some foods that you really did not want to eat because of a lack of resources to obtain other types of food?**			
	Sau shoole lamalara hoongunni kainohunni ati woy miniki manni giddo mittu ita hasidhinayikki sagalaittineegentinooni	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት ዐራት ሳምንታት መግዛት ካለመቻል የተነሳ ዐንቺ ወይም ከቤተሰብሽ ዐባላት መብላት የማትፈልጉትን ምግብ በልታችሁ ታውቃላችሁ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
908 Skip ዝለል	**How often did this happen in the past four weeks?**			
	Kuni mageeshshi geeshshaatisau shoole lamalara ikkeuohu	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ

	In the past four weeks, did you or any household member have to eat a smaller meal than you felt youneeded because there was not enough food?			
909	Sau shoole lamalara atway minikki maate giddomitto ikkado sagala heera hoogatenni kainohunni shiima ikkado sagala sagalino	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት ዐራት ሳምንታት መግዛት ካለመቻል የተነሳ ዐንቺ ወይም ከቤተሰብሽ ዐባላት መብላት ከሚገባው መጠን በታች በልታችሁ ታውቃላችሁ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
	How often did this happen in the past four weeks?			
910 Skip ዝለል	Sagalino sau shoole lamalara mageshshi yanna geeshshati xaadinohu	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ
	In the past four weeks, did you or any other household member have to eat fewer meals in a day because there was not enough food?			
911	Sau shoole lamalara ikkado sagale heera hoogatenni kainohunni at woy minikkki maate giddo mittu barru giddo sagala noosi yanna sagalinokki yanna no	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት 4 ሳምንታት በቂ ምግብ ካለመኖሩ የተነሳ ዐንቺ/ከቤተሰብሽ መካከል ምብላት ባለበት ሰዐት ያልተመገበ ዐለ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
912 Skip ዝለል	**How often did this happen in the past four weeks?**			
	Sau lamalara mee yanna geeshshaati xaadinonehu	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2.ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ
	In the past four weeks, was there ever no food to eat of any kind in your household because of lack of resources to get food?			

913	Sau lamalara anjetennikainohunni sagale minigiddo hooge epentewo	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት 4 ሳምንታት መግዛት ካለመቻል የተነሳ ማንኛዉም ዐይነት ምግብ ከቤትሽ ጠፍቶ ያውቃል	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
	How often did this happen in the past four weeks?			
914	Ikkiromageshshi yanna geeshshaati	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
ዝለል	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ
915	**In the past four weeks, did you or any household member go to sleep at night hungry because there was not enough food?**			
	Sau shoole lamalara ati woy minnikki maate giddo mittu sagalete anjoninni kainohunni hudiisannassi gaxinohu no	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት 4 ሳምንታት ዐንቺ/ከቤተሰብሽ መካከል ከምግብ ዕጥረት የተነሳ ዕየራበው ሳይበላ የተኛ ዐለ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
	How often did this happen in the past four weeks?			
916 Skip ዝለል	Ikkiro mageeshshi yanna geeshshaati sau shoole lamalara	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ዐጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ
917	**In the past four weeks, did you or any household member go a whole day and night without eating anything because there was not enough food?**			
	Sau shoole lamalara at way minikki maate giddo mittu sagalete anjenni kainohunni sagalikkinni 24 saate keeshshinohu no	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፉት 4 ሳምንታት ዐንቺ/ከቤተሰብሽ መካከል ከምግብ ዕጥረት የተነሳ ሳይመገብ 24 ሰዐት የቆየ ዐለ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን

918 Skip ዝለል	**How often did this happen in the past four weeks?**			
	Ikkiro mageeshshi yanna geeshshaati kalaqaminohu	[_]1.Rarely (once or twice in the pastfour weeks)	[_]1.Harancho hanna	[_]1.ጥቂት ጊዜ
	ባለፉት 4 ሳምንታት ለምን ያህል ጊዜ ነው ያጋጠመሽ	[_]2.Sometimes (three to ten times inthe past four weeks)	[_]2.Sae sae	[_]2. ዐንዳንድ ጊዜ
		[_]3.Often (more than ten times the past four weeks)	[_]3.Seeda	[_]3. ብዙ ጊዜ
919	**Have you received any food support in the past month?**			
	Sai aganira ayita sagalete kaalo adhootta	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ባለፈው ወር ማንኛውም የምግብ ዕርዳታ ዐግኝተሻል	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
920	**How do you cope at times when you are running out of food in the house?**			
	Sagala mini giddo heedhu kkinni gatturo maat assahahu	1[_].Reduce number of meals	1[_]Adhaw sagaleaanohea yeajisheena	1[_]የሚወሰደ ውን የምግብ ድግግሞሽ ቀንሳለሁ
	በቤት ውስጥ የምግብ ዕጥረት ሲያጋጥምሽ ምን ዐማራጭ ነው የምትወስጂው	2[_].Reduce meal size	2[_]Sagalete geesha ajiplema	2[_]የምግብ መጠን ቀንሳለሁ
		3[_].Borrowing	3[_]Liqiirema	3[_]ዕበደራለሁ
		4[_].Petty trade	4[_]Hirary sagale hasileme	4[_]የሚሸጥ ነገር ፈልጋለሁ
		5[_].Consume stored food (seed)	5[_]Gootam giddo nosagale harunsitmo	5[_]በጎተራ ያለ ምግብ ዕጠቀማለሁ
		6[_].Migration for labour	6[_]Loosoho walekachcha horeema	6[_]ለስራ ወደ ሌላ ዐካባቢ ሔዳለሁ
		7[_].Sell of farm tools	7[_]Hwurayi udene hirana	7[_]የማረሻ ዕቃዎችን ሸጣለሁ
		8[_].Sale charcoal/fire wood	8[_]Ishine hirema	8[_]ቆሻሻ ሸጣለሁ
		9[_].Daily labor	9[_]Barulooso losema	9[_]የቀን ሥራ ሰራለሁ
		10[_].Safety Net	10[_]Kaalo hasireema	10[_]ዕርዳታ ዐገኛለሁ
		11[_].Sell of farm animals	11[_]Ishine adhe itema	11[_]ከቆሻሻ ወስጀ በላለሁ
		12[_].Other(specify)	12[_]Welu noose keeli	12[_]ሌላ ካለ ጥቀስ/ሽ

Section 10: Household dietary diversity
Kiffile 10. Mini mate sagalete danisagala
ክፍል 10. የቤተሰቡ የምግብ ዐመጋገብ
Now I would like to ask you about the types of foods that you or anyone else in your household ate yesterday during the day and at night either separately or combined with other foods.
ቤተሰቡ በ 24 ሰዐት ዉስጥ የተመገበውን የምግብ ዐይነት የሚገለፅ መጠይቅ (ቤተሰቡ ቁርስ፣ምሳ፣ዕራት የሰራውን ወይም የተመገበውን ምግብ ጠይቅ የሚስማማውን ቦታ ዐመልክት)

1000	**Were there any foods that were not prepared for the in the house because it was a fasting day?**			
	Qatume ikkewo daafira loonsoyikki sagale no	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ጾም ስለሆነ ያልተሰራ ምግብ ዐለ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1001	**Could you tell me the types of foods that were prepared in the house and that you or anyone else in your household ate?**			
	Loonsoyi sagale kulatoe	[_]Breakfast		[_]ቁርስ
	ለቤተሰቡ የተሰራውን የምግብ ጊዜ ትነግሪኛለሽ	[_]Lunch		[_]ምሳ
		[_]Dinner		[_]ዕራት
		[_]Others		[_]ሌላካለ ጥቀስ/ሽ
1002	**Any bread, rice, pasta, biscuits, or any other foods made from millet, sorghum, maize, rice, wheat?**			
	Daabbo,paarta,ruuze,koshoro raino sagale woy ajjunni ,badalatenni ,hayixunni,qamadetenni,bashanqunniy loonsoonni sagale woy wolu quminni,xawisi	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ዳቦ፣ ፓስታ፣ ሩዝ፣ ብስኩት፣ ኩኪስ፣ ወይም ማንኛውም ነገር ከአጃ ከበቆሎ ገብስ፣ ሰንዴ፣ ማሽላ፣ወይም ሌላ አህል ዘር የተሰራ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1003	**Any potatoes, bulla, kocho or any other food made from roots or tubers?**			
	Maxaaxeesla, diinicha, boyina,lawinore saga'lino?	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም ነጭ ድንች፣ ቦይና፣ እንሰት (ማንኛውም ነጭ ስራስር)	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1004	**Any vegetables?**			
	Wole aye ataakilte saga'lino?	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም አታክልት	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1005	**Any fruits?**			
	Wole aye gumma	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም ፍራፍሬ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1006	**Any beef, pork, lamb, goat, rabbit wild game, chicken, duck, or other birds, liver, kidney, heart, or other organ meats?**			

	Bootu,mancheemete,gereewo,mellenna hilleessa,Lukko,daakiyye, Afale,mule,wodana,woy wole godowu giddo malla?	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	የበሬ፤ የበግ/የፍየል የአሳማ,የጥንቸል ,ዶሮ፤ እርግብ/ወፍ (ጉበት፤ ኩላሊት፤ የልብ ወይም ማንኛውም የውስጥ ሰውነት ክፍል)	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1007	**Any eggs?**			
	Ayee quuphe	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም እንቁላል	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1008	**Any fresh or dried fish or shellfish?**			
	Ayee qilxi'me	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም አሳና የአሳምርት	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1009	**Any foods made from beans, peas, lentils, or nuts?**			
	Aye segale baqeluuni, atarunni, qibaatete qumma shumburunni qixxeessinoonni sagale saga'lino?	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም ምግብ ከ ባቄላ፣አተር፤ ምስር ወይም ሌላ ጥራጥሬ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1010	**Any cheese, yogurt, milk or other milk products?**			
	Ayibe/geinto saga'lino?	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም አይብ፣እርጎ፣ወተት ወይም ሌላ የወተት ምርቶች	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1011	**Any foods made with oil, fat, or butter?**			
	Zayitetenni woy buurumi loonsoonni sagala	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	በዘይት ወይም በቅቤ የተሰራ ምግብ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1012	**Any sugar or honey?**	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ማንኛውም ስኩዋርና ማር	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1013	**Any other foods, such as condiments, coffee, tea?**	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ቡናና ሻይ	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1014	**Alchol drink**			
	Diribisanno ago (Birra, Xesiixella,Areqe	0. [_]No	0. [_]Dee'ni	0. [_]አይደለም
	ዐልኮል መጠጦች (ቢራ፣ ዐረቂ፣ጠጅ፣ጠላ)	1. [_]Yes	1. [_]Eewa	1. [_]አዎን
1015	**Others specify**			
	Welere			
	ሌላ ካለ ዘርዝር			

Anthropometry assessment form

Village ____________________________________Date of data collection________________

Name and signature of data collector ________________Supervisor _____________________

HH.No	Child ID	Child name	Child age	Sex	Wt (kg)		Ht (Cm)	
						2		2

Blood sample collection form

Village ________________________________Date of data collection______________

Name of data collector ______________________Supervisor sig_____________________

HH. No	Child ID	Child name	Child age	Sex	Haemoglobin	Ferritin	CRP	Remark

Ethical Approvals

Region:	Saksbehandler:	Telefon:	Vår dato:	Vår referanse:
REK sør-øst	Anette Solli Karlsen	22845522	15.12.2016	2016/2034/REK sør-øst A
			Deres dato:	Deres referanse:
			01.11.2016	

Vår referanse må oppgis ved alle henvendelser

Alemselam Orsango
Universitetet i Bergen

2016/2034 Forebygging av jernmangelanemi: Evaluering av amarant gryn supplementering til 24.0-59.9 måneder gamle barn i sør-Etiopia, et randomisert kontrollert forsøk

Forskningsansvarlig: Universitetet i Bergen
Prosjektleder: Alemselam Orsango

Vi viser til søknad om forhåndsgodkjenning av ovennevnte forskningsprosjekt. Søknaden ble behandlet av Regional komité for medisinsk og helsefaglig forskningsetikk (REK sør-øst) i møtet 01.12.2016. Vurderingen er gjort med hjemmel i helseforskningsloven (hfl.) § 10, jf. forskningsetikkloven § 4.

Prosjektbeskrivelse (revidert av REK)

Formålet med dette prosjektet er å undersøke anemiforholdene blant barn i sør-Etiopia, og videre hvorvidt tilskudd av det lokalt tilgjengelige amarantgrynet kan ha gunstig effekt på helsen til barn sammenliknet med tradisjonelt brukt maismel.

Jernmangelanemi hos barn er et stort problem i fattige land grunnet lite variasjon og mat med lavt innhold av viktige mikronæringsstoffer. For å bøte på mangler i kosten har man mulighet til å berike maten med vitaminer og mineraler, eller å gi supplement med næringsstoffer. Disse to metodene har ikke vist seg å være bærekraftige over tid. Ideelt bør barn ha tilgang på flere proteinrike kilder slik som egg, kjøtt, fisk og meieriprodukter. Da dette for de fleste vurderes som dyrt og uoppnåelig å inkludere i dietten så ønsker man å undersøke om nye, billigere og plantebaserte alternativer er effektive. Amarant har høyt protein- og jerninnhold og er lett tilgjengelig. Amarant har en gunstig profil i forhold av mikronæringsstoffer og inneholder 4 ganger mer jern enn tradisjonelt benyttet maismel.

Prosjektet er todelt. Det skal til en tverrsnittsundersøkelse inkluderes 340 barn mellom 2 og 5 år. Videre planlegges det, basert på funn i tverrsnittsundersøkelsen, å inkludere inntil 100 barn mellom 2 og 5 år, med mild og moderat jernmangelanemi til en kostintervensjon.

Deltakere i kostintervensjonen skal randomiseres til intervensjon med 150 g brød daglig med enten 70 % amarant og 30 % kikerter eller 100 % maismel. Brødene skal bakes i separate bakerier og undersøkelsen skal gjennomføres blindet. Intervensjonsperioden varer i 6 måneder.

Primært endepunkt for intervensjonen er effekt på hemoglobin målt i blod.

Deltakelse i tverrsnittsundersøkelsen innebærer utfylling av et spørreskjema om sosio-demografiske forhold samt spørsmål om næringsinntak, vaksinering, hospitalisering og sykelighet. Det vil bli foretatt klinisk undersøkelse samt tas blodprøver der CRP, Hgb og Ferritin vil bli undersøkt. Kunnskapsspørsmål om kosthold og anemi vil også bli inkludert.

For de 100 barna som selekteres til intervensjon, vil det stilles spørsmål om næringsinntak regelmessig gjennom intervensjonsperioden. Disse barna vil bli undersøkt klinisk hver måned, og blodprøver tas ved inklusjon samt etter tre og seks måneder.

Blodprøver skal inngå i ny, prosjektspesifikk biobank «Ethiopian public health institute (EPHI)», med ansvarshavende Alemselam Zebdewos ved EPHI.

Vurdering

Etter komiteens syn er dette en viktig studie. Man skal her undersøke hvorvidt en rimelig og tilgjengelig matvare kan redusere problemene som følger et kosthold der jernbehovet ikke er dekket. Deltakelse innebærer at barna får utdelt daglige rasjoner av enten brød bakt av amarant og kikerter eller maismel.

Helseforskningloven § 18 omhandler vilkår for person uten selvstendig samtykkekompetanse. Ett av vilkårene er «.. *at det er grunn til å anta at resultatene av forskningen kan være til nytte for den aktuelle personen eller for andre personer med samme aldersspesifikke lidelse, sykdom, skade eller tilstand.*» Etter komiteens syn antas dette vilkåret oppfylt, sett i lys av de resultater som potensielt kan følge av prosjektet. For samtlige deltakere vil i tillegg studien ha nytteverdi, ved energitilførsel gjennom kostintervensjonen.

I helseforskningloven § 18 er det videre gitt at «*For mindreårige kreves det at tilsvarende forskning ikke kan gjennomføres på personer som ikke er mindreårige*.» Når det gjelder tilførsel av jern fra amarantbrødet, antas det at intervensjonen vil ha størst nytteverdi for de minste barna. Av den grunn mener komiteen at det er forsvarlig at prosjektet gjennomføres i denne aldersgruppen.

Sett i lys av hvilke vilkår det for øvrig ville vært stilt, dersom prosjektet skulle vært gjennomført i Norge, har prosjektet enkelte mangler. Dette dreier seg om omfanget av informasjonsskrivet og at det ikke legges opp til at begge foresatt skal samtykke på vegne av barnet. Imidlertid vurderer komiteen det slik at lokale rutiner og tradisjoner må veie tungt, så lenge dette ikke går på bekostning av deltakeren (barnets) velferd og integritet. Prosjektet ansees som lite invasivt med direkte nytte for barnet og er gitt stedlig godkjenning av IRB ved Hawassa University og Tulla sub city Health bureau, Etiopia. Komiteen anser dermed at prosjektet har blitt vurdert i tråd med lokale forhold og dermed vil bli forsvarlig gjennomført.

Prosjektet har relativt kort sluttdato, 03.09.2018. Prosjektperioden omfatter, i tillegg til praktisk gjennomføring av studien, også forskning og publisering av de opplysninger som er innhentet. Det er en vanlig misforståelse at sammenstilling av data og publisering skal skje etter prosjektperiodens utløp og ikke innenfor perioden. Etter prosjektslutt skal altså dataene oppbevares, men ikke forskes på. Av den grunn forlenges prosjektperioden med to år, til 03.09.2020.

Vedtak

Prosjektet godkjennes med hjemmel i helseforskningsloven §§ 9 og 33.

Godkjenningen er gitt under forutsetning av at prosjektet gjennomføres slik det er beskrevet i søknaden og protokollen, og de bestemmelser som følger av helseforskningsloven med forskrifter.

Godkjenningen gjelder til 03.09.2020.

Av dokumentasjonshensyn skal opplysningene oppbevares i 5 år etter prosjektslutt. Opplysningene skal oppbevares avidentifisert, dvs. atskilt i en nøkkel- og en datafil. Opplysningene skal deretter slettes eller anonymiseres.

Forskningsprosjektets data skal oppbevares forsvarlig, se personopplysningsforskriften kapittel 2, og Helsedirektoratets veileder for «Personvern og informasjonssikkerhet i forskningsprosjekter innenfor helseog omsorgssektoren».

Prosjektet skal sende sluttmelding på eget skjema, jf. helseforskningsloven § 12, senest et halvt år etter prosjektslutt.

Dersom det skal gjøres endringer i prosjektet i forhold til de opplysninger som er gitt i søknaden, må prosjektleder sende endringsmelding til REK, jf. helseforskningsloven § 11.

Klageadgang

Komiteens vedtak kan påklages til Den nasjonale forskningsetiske komité for medisin og helsefag, jf. helseforskningsloven § 10 tredje ledd og forvaltningsloven § 28. En eventuell klage sendes til REK sør-øst A. Klagefristen er tre uker fra mottak av dette brevet, jf. forvaltningsloven § 29.

Med vennlig hilsen

Knut Engedal
Professor dr. med.
Leder

Anette Solli Karlsen
Komitesekretær

Kopi til:ingunn.engebretsen@uib.no; post@uib.no

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HAWASSA UNIVERSITY
COLLEGE OF MEDICINE AND HEALTH SCIENCES
Institutional Review Board

Ref. No: IRB/098/08

Date: 06/09/2016

Name of Researcher/s: ***Alemselam Zebdewos, Ingunn Marie, Bernt Lindtjorn, Eskindir Loha***

Topic of Proposal: ***Preventing iron deficiency anemia: Evaluation of amaranth grain supplementation to 24.0-59.9 month old children in southern Ethiopia, a randomized controlled trial***

Dear researcher(s),

The Institutional Review Board (IRB) at the College of Medicine and Health Sciences of Hawassa University has reviewed the aforementioned research protocol with special emphasis on the following points:

1. Are all principles considered?
   - 1.1. Respect for persons: Yes ☑ No ☐
   - 1.2. Beneficence: Yes ☑ No ☐
   - 1.3. Justice: Yes ☑ No ☐
2. Are the objectives of the study ethically achievable? Yes ☑ No ☐
3. Are the proposed research methods ethically sound? Yes ☑ No ☐

Based on the aforementioned ethical assessment, the IRB has:

A. Approved the proposal for implementation ☑

B. Conditionally Approved ☐

C. Not Approved ☐

Yours faithfully,

Ayalew Astatkie (PhD),

Institutional Review Board Chairperson.

Institutional Review Board
Hawassa University College of Medicine & Health Sciences

☎ +046 820 92 90 Fax + 046 2208755
✉ 1560 Hawassa

19 May 2017

To Whom It May Concern:

RE: Preventing iron deficiency anemia: Evaluation of amaranth grain supplementation to 24.0-59.9 month old children in southern Ethiopia, a randomized controlled trial.

As project manager for the Pan African Clinical Trial Registry (www.pactr.org) database, it is my pleasure to inform you that your application to our registry has been accepted. Your unique identification number for the registry is **PACTR201705002283263**

Please be advised that you are responsible for updating your trial, or for informing us of changes to your trial.

Additionally, please provide us with copies of your ethical clearance letters as we must have these on file (via email, post or fax) at your earliest convenience if you have not already done so.

Please do not hesitate to contact us at +27 21 938 0835 or email epienaar@mrc.ac.za should you have any questions.

Yours faithfully,

Elizabeth D Pienaar
www.pactr.org Project Manager
+27 021 938 0835

THE SOUTH AFRICAN MEDICAL RESEARCH COUNCIL
Cochrane South Africa | PO Box 19070, Tygerberg, 7505
Tel: +27 (0)21 938 0438 | Email: cochrane@mrc.co.za | Web: www.southafrica.cochrane.org

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Hawassa University

College of Medicine and Health Sciences

Chief Academic and Research Director

ቁጥር ሀ/ኮጤሳ/ም/3633/09
Ref. No

ቀን 15/06/09
Date

ለ ኢትዮጵያ ህብረተሰብ ጤና ኢንስቲትዩት

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ጉዳዩ፡ ትብብር ስለመጠየቅ

መምህርት አለምሰላም ዘብዲዎስ በሀብ/አካ/ጤ/አ ት/ቤት በማገልገል ላይ የነበሩ ሲሆን ከህዳር 2008 ዓ.ም ጀምሮ የPhD ትምህርት እድል ተሰጥቷቸው ትምህርታቸውን በመከታተል ላይ ይገኛሉ ። አሁን የምርምር ስራቸውን "preventing Iron deficeincy anemia: evaluation of amaranth grain supplementation to 24.0- 59.9 month old children in southern Ethiopia ,a randomized controlled trial" በሚል ርዕስ በመስራት ላይ ይገኛሉ ይህም ምርምር የላብራቶሪ ጥናት ስላለው እንደሚከተለው የዘረዘርነውን አገልግሎት መስሪያ ቤቱ እንዲተባበራቸው ስንል በትህትና እንጠይቃለ ?

የላብራቶሪ አገልግሎት ዝርዝር

- የደም ምርመራ 100 ናሙና ሲሆን የሚያስፈልገው ምርመራ (serum feretine and C-reactive protine (CRP))
- የምግብ ናሙና ብዛት 8 የሚያስፈልገው ምርመራ (protein, carbohydrate, fat, Iron, calcium zinc and phytate level)

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Errata for

Childhood undernutrition and the potential of amaranth for reducing anaemia in southern Ethiopia

By Alemselam Zebdewos Orsango

Thesis for the degree philosophiae doctor (PhD)
at the University of Bergen

13/04/2022 (date and sign. of candidate)

(date and sign. of faculty)

Errata

- Page 1 Missing space: “age(6).” – corrected to “age (6).”
- Page 18 Full stop misplaced: “(paper I.)’’ corrected to “(paper I).’’
- Page 27 Missing words: ‘‘twenty’’ Corrected to ‘’ twenty eight’’
- Page 53 Unmatched words: ‘‘improved ferritin levels among the study participants.’’ Corrected to ‘‘improved iron deficiency anaemia among the study participants.’’

Graphic design: Communication Division, UiB / Print: Skipnes Kommunikasjon AS

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ISBN: 9788230842102 (print)
9788230859827 (PDF)