Dedicated to my mother: Yeshidinber Abebe

For your hard work and inspiration

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THE ROLE OF FEEDING PRACTICES, DIETARY DIVERSITY AND SEASONALITY AND N-3 LONG-CHAIN POLYUNSATURATED FATTY ACIDS IN THE DIET OF ETHIOPIAN INFANTS AND YOUNG CHILDREN

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List of abbreviations

AA	Arachidonic acid
AAP	American Academy of Pediatrics
ALA	α-linolenic acid
AMDR	Acceptable macronutrient distribution range
CI	Confidence interval
CRP	C-reactive protein
CSA	Central Statistical Agency [Ethiopia]
DDS	Dietary diversity score
DFSFQ	Department of Food Safety and Food Quality (Ghent University)
DHA	Docosahexaenoic acid
EDHS	Ethiopia Demographic and Health Survey
EE	Environmental enteropathy
EED	Environmental enteric dysfunction
EHNRI	Ethiopian Health and Nutrition Research Institute
ENI	Ethiopian Nutritoin Institute [Ethiopia]
EPA	Eicosapentaenoic acid
FAO	Food and Agriculture Organization of the United Nations
FDRE	Federal Democratic Republic of Ethiopia
FDRE-PCC	Federal Democratic Republic of Ethiopia, Population Census Commission
FMoH	Federal Ministry of Health
FVS	Food variety score
GGFRC	Gilgel Gibe Field Research Center
HAZ/LAZ	Height-for-age /Length-for-age
ICFI	Infant and child feeding index
IFPRI	International Food Policy Research Institute
IHSR	Institute of Health Science Research [Jimma University]
IQR,	Interquartile range
IYCF	Infant and young child feeding
LA	Linoleic acid

LIC	Low income country
LNS	Lipid-based nutrient supplements
MDGs	Millennium Development Goals
MMDA	Mean micronutrient density adequacy
MPA	Probability adequacy
n-3 LC-PUFA	Omega-3 Long-chain poly unsaturated fatty acids
n-6 LC-PUFA	Omega-6 Long-chain poly unsaturated fatty acids
NDA	Nutrient density adequacy
NNP	National Nutrition Programme
OR	Odds ratio
PAHO	Pan American Health Organization (PAHO)
RCT	Randomized controlled trial
RE	Retinol equivalent
RUSF	Ready to use supplementary foods
RUTF	Ready to use therapeutic foods
SDG	Sustainable Development Goal
SUN	Scaling Up Nutrition
UN	United Nations
UNICEF	United Nations Children's Fund
WHO	World Health Organization
WHZ/WLZ	Weight-for-height /Weight-for-length

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1

Chapter 1: General Introduction

1.1. Global nutrition situation of children

1.1.1. Undernutrition in children

It is known that better nourished population is an essentional part of sustainable development in a given country. Despite considerable achievments towards reducing the rate of malnutrition, it remained an important public health problem in many countries of the world. Eighty eight percent of the countires included in the recent Global Nutrition Report, face multiple burden of malnutrition among children and women in the reproductive age (Development Initiatives, 2017). There has been a decline in the rate of chronic or acute malnutrition in many countries of the world; however, many of them are far behind in the pace of reducing stunting and such level of improvement is not enough to meet the internationally agreed nutrition targets such as Sustainable Development Goal (SDG) target 2.2 to end all forms of malnutrition by 2030(United Nations, 2015b). There is a general consensus that poor nutritional status during childhood (and in utero) can have long-lasting consequences into adulthood, both in terms of health and mortality, and other measures of human capital such as schooling and productivity(Maluccio and Flores, 2005, Glewwe and Miguel, 2007, Ahmed et al., 2014).

Linear growth has been known to be a very good indicator of child wellbeing and it has been widely used to indicate measure of quality of child environment, current and past occurrences of nutritional or health problems (WHO, 1995, Hoddinott et al., 2013b, Bhutta and Salam, 2012, de Onis and Branca, 2016). Linear growth failure is the commonest growth failure (Prendergast and Humphrey, 2014) and a good marker of overall socioeconomic inequality in a given country (de Onis and Branca, 2016). Achieving optimal growth during infancy and early childhood is an important measure towards achieving development objectives (Prendergast and Humphrey, 2014). It impacts on child health, mortality, and future educational attainment (Ramakrishnan et al., 1999, Bhutta and Salam, 2012, de Onis and Branca, 2016).

In 2013, linear growth failure (stunting) was themost prevalent type of child malnutrition affecting an estimated 161 million children globally (de Onis and Branca, 2016) and it is mostly left unrecognized in a community. More than 80% of the world's undernourished children are found in countries with a stunting prevalence ≥20% which shows the coexistence of other forms of growth faltering (Bryce et al., 2008).An earlier estimate of levels and patterns of stunting for the period 1990–2020, predicted a high prevalence of stunting among children 0-5 years of age globally in the year 2010 (27%)(de Onis et al., 2004a). The most affected region remained to be East Africa in which 45% of the children are affected in 2010 followed by Middle (39%) and Western Africa (38%)(Bhutta and Salam, 2012, Prendergast and Humphrey, 2014, de Onis et al., 2012). In 2011, the prevalence of stuntingin sub-Saharan Africa and south Asia was 36% and 27% respectively(Black et al., 2013). Lately, the WHO, UNICEF and World Bank Group estimated that more than half of all stunted children under 5 lived in Asia and more than one third lived in Africa in 2015(UNICEF/WHO/World Bank, 2016)**(Figure 1.1)**. In 2000 and 2015, the absolute number of children who are stunted under the age of five years increased by 8% in low income countries while there has been a reduction by 57% in the number of children malnourished in high income countries(UNICEF/WHO/World Bank, 2016).

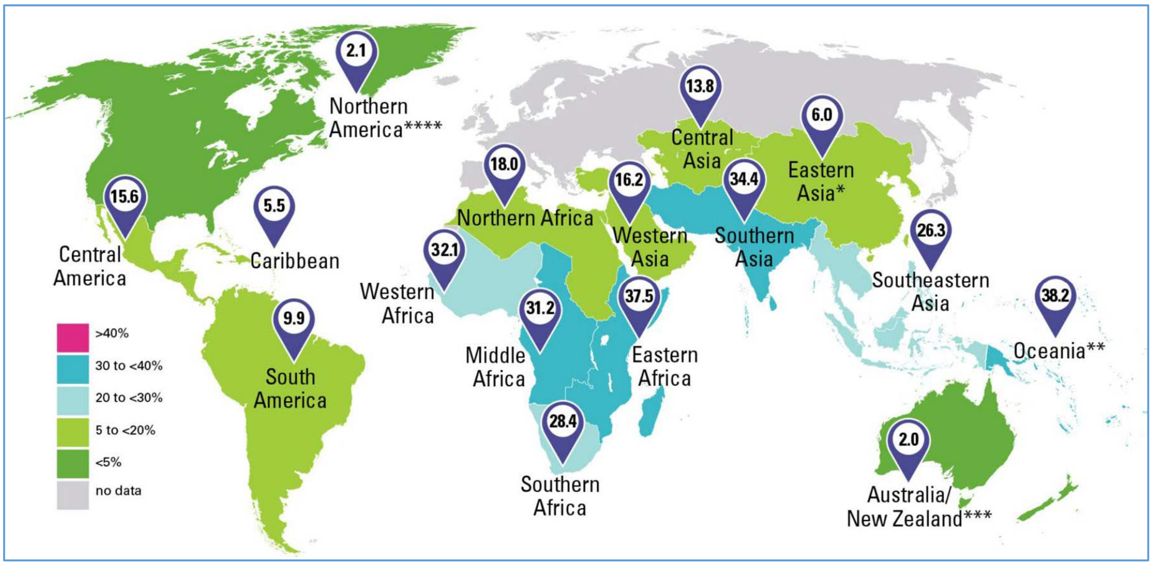


Figure 1.1. : Percentage of stunted children under 5, by United Nations sub-region, 2015;
Source: UNICEF, WHO, World Bank Group joint malnutrition estimates, 2016 edition.
Available at: http://www.who.int/nutgrowthdb/estimates2015/en/

1.1.2. Overview of child growth indicators

The three most commonly used anthropometric indices to assess child growth status are weight-for-height/length, height/length-for-age, and weight-for-age. Currently, the WHO

Child Growth Standard (WHO, 2006) is used to calculate such indices. There are a number of ways of expressing the growth indicators, but z-scores are commonly used. Hence, height/length-for-age z-scores (HAZ/LAZ), weight-for-age z-scores (WAZ), and weight-for-length/height z-scores (WHZ/WLZ) indicate stunting, underweight or wasting respectively when low compared to a median for the same age and sex of the WHO Child Growth Standards(WHO, 2006).

1.2. Timing of growth faltering

1.2.1. The "first 1000 days"

The time from conception to the second year of life ("the first 1000 days") is considered to be the most sensitive period in terms of vulnerability to inadequate nutrition and care, and response to specific and nutrition sensitive interventions(Victora et al., 2010, Prendergast and Humphrey, 2014).This period is also characterized by a significant proportion of physical growth in brain and musculoskeletal and other major organ tissues (**Figure 1.2**). The period determines not only the nutritional situation during the first two years but also during the subsequent periods in the human reproductive life cycle (http://thousanddays.org/). Nutritional interventions and maternal dietary practices during pregnancy are important factors for the development of important endocrine, metabolic, immunological alterations with potentional long term outcomes for health (Agosti et al., 2017). Thisfurther highlights the importance of growth in the first 1000 days and its effect in subsequent stages in the life cycle (Victora et al., 2008).

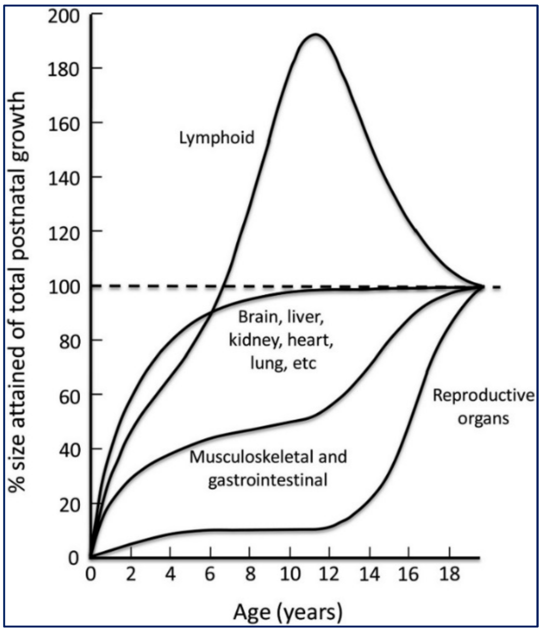


Figure 1.2. : Postnatal growth of body systems in humans by age (Source :(Prentice et al., 2013))

1.2.2. The role of prenatal period

Studies in several countries showed that growth restrictions which happened prenatally contribute significantly to the development of growth faltering in the early postnatal years. The intergenerational links of child growth or undernutrition are well established in which maternal birth weight determines the birth weight of her offspring (Martorell and Zongrone, 2012). The association remains unchanged after adjusting for socioeconomic status. It has been shown that fetal growth rather than gestational age was found to be an important factor for post natal growth and subsequent effect on the offspring (Ramakrishnan et al., 1999). Analysis of data from a large sample of children below 4 years of age at different time points showed that from the total growth faltering that had taken place in the first three years, 44-55% was present at birth (Mamidi et al., 2011) and another analysis of data from 19 birth cohorts showed that 20% of growth faltering that occurred between 12-60 months had intra- uterine origins (Christian et al., 2013).Twenty percent of the growth faltering in the first 3 years in Malawi had intra uterine origins(Dewey and Huffman, 2009).

1.2.3. Growth during the first two years

The possibility of catching up the lost growth during early childhood has been an important issue among the nutrition community for a long time. Fifty percent of the stunting that occurs during the first 3 years happens in the age range 6-24 months in a study among Malawian children (Dewey and Huffman, 2009) as a result of exposure to environmental factors affecting growth and development. Analysis of data from several low and middle income countries demonstrated that there is deterioration in HAZ from early in life to about 24 months of age but there is no change in HAZ beyond this age (Victora et al., 2010)(**Figure 1.3)** despite recent assertion that there is some catch up in linear growth (increase in HAZ) beyond 24 months of age (Prentice et al., 2013). However, 30% of the growth faltering during the first 60 months occurs after the second year of life which shows a potential for growth beyond the first 1000 days (Leroy et al., 2014, Leroy et al., 2015).There is a tendency for HAZ to increase after 2 years due to an increase in the standard deviation of the reference population with age(WHO, 2006, Onis et al., 2007) rather than actual catchup in linear growth.

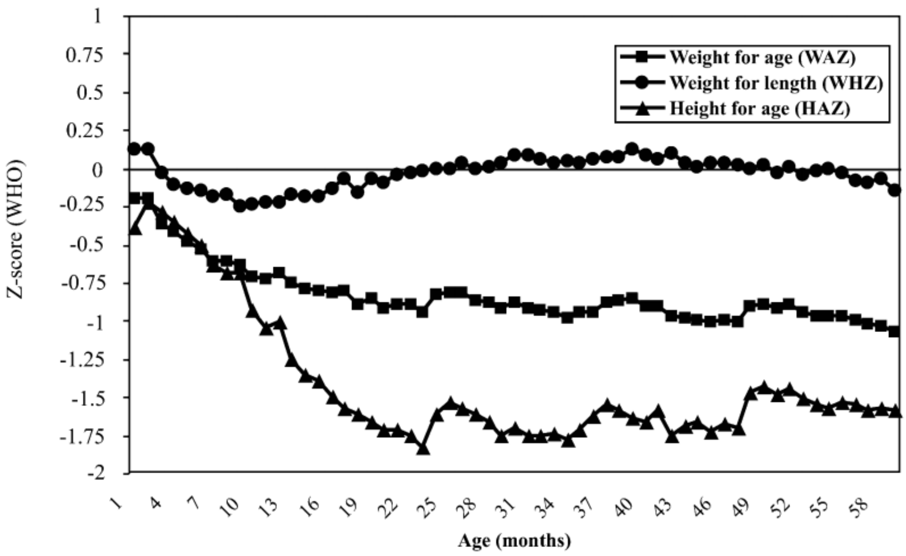


Figure 1.3 Mean anthropometric *z* scores according to age for all 54 studies, relative to the WHO standard (1 to59 months). Analysis was made based on the information available from the WHO Global Database on Child Growth and Malnutrition from nationally representative anthropometric surveys of 54 countries (Ethiopia DHS 2005 data included). Source: (Victora et al., 2010)

1.3. Determinants of childgrowth

The determinants of child undernutrition operate at multiple levels of causation, ranging from the most distal socioeconomic and political variables to the most proximal ones including intake of recommended nutrients and the occurrence of clinical and subclinical childhood infections. There is significant interaction among suboptimal feeding practices, several nutrient deficiencies and infectious diseases in causing childhood growth restriction or stunting (Black et al., 2008, Dewey and Mayers, 2011, Ahmed et al., 2014). Nutritional health or undernutrition is a product of the lack of balance between food intake and utilization nutrients and energy (Black et al., 2008). This balance could be influenced by diseases and mediated by underlying causes of undernutrition such as maternal and child care practices, household food security, and environmental sanitation and health services (**Figure 1.4**).

1.3.1. Genetic and prenatal factors

Human height is known to be a heritable polygenic trait in which genome-wide association studies have identified about 200 loci that have a significant association with height (Berndt et al., 2013, Lango Allen et al., 2010). Even if genetic inheritance explains at least 10% of the variation in stature attained during adulthood (Lango Allen et al., 2010), children from different geographical and ethnic backgrounds experience a similar pattern of growth as long as certain conditions are met (Martorell and Zongrone, 2012). The latter conditions include good maternal nutrition, appropriate child feeding practices, environmental hygiene, accessible health care services and treatment of common childhood illnesses (Dewey and Mayers, 2011, Black et al., 2013). Maternal condition prior to and during pregnancy such asshort stature, poor maternal nutrition and weight gain and inadequate care during pregnancy are important factors for low birth weight and poor postnatal growth(Abu-Saad and Fraser, 2010, Black et al., 2013, Neufeld et al., 2004). Analysis of 109 Demographic and Health Surveys in 54 countries conducted between 1991 and 2008 showed that maternal stature was inversely related with offspring mortality, underweight, and stunting in infancy and childhood(Özaltin et al., 2010). Hence, maternal care prior to and during pregnancy is crucial for the optimal growth and health of the offspring.

1.3.2. Socioeconomic factors

The highest burden of malnutrition is occurring in low-income countries. Based on analysis of data from 80 countries, undernutrition has been shown to be correlated with income inequalities in a given country (Bredenkamp et al., 2014). As part of Young Lives study (the multinational cohort study) in four developing countries including Ethiopia, children from lowest wealth quintile (poorest households) were more likely to be stunted and improving wealth status was associated with reduced probability of being stunted(Petrou and Kupek, 2010). Recent Lancet series also demonstrated that stunting prevalence among children younger than five years was 2.47 times (range 1.00-7.64) higher in the poorest quintile of households compared to the richest quintiles using data from 79 countries with population based surveys(Black et al., 2013). Based on data from the 5 studies of 7630 mother–child dyads in the Consortium on Health Orientated Research in Transitional Societies (COHORTS), children were on average taller than their mothers due to improvements in the socioeconomic situation, nutrition and environmental hygiene(Addo et al., 2013). Progressive growth in GDP over a long period time starting from the great economic reform in 1978 has been also associated with an improvement in growth rate among children and adolescents in China (Zong and Li, 2014); however, a higher rate of undernutrition was observed in regions where there was high-income inequality.

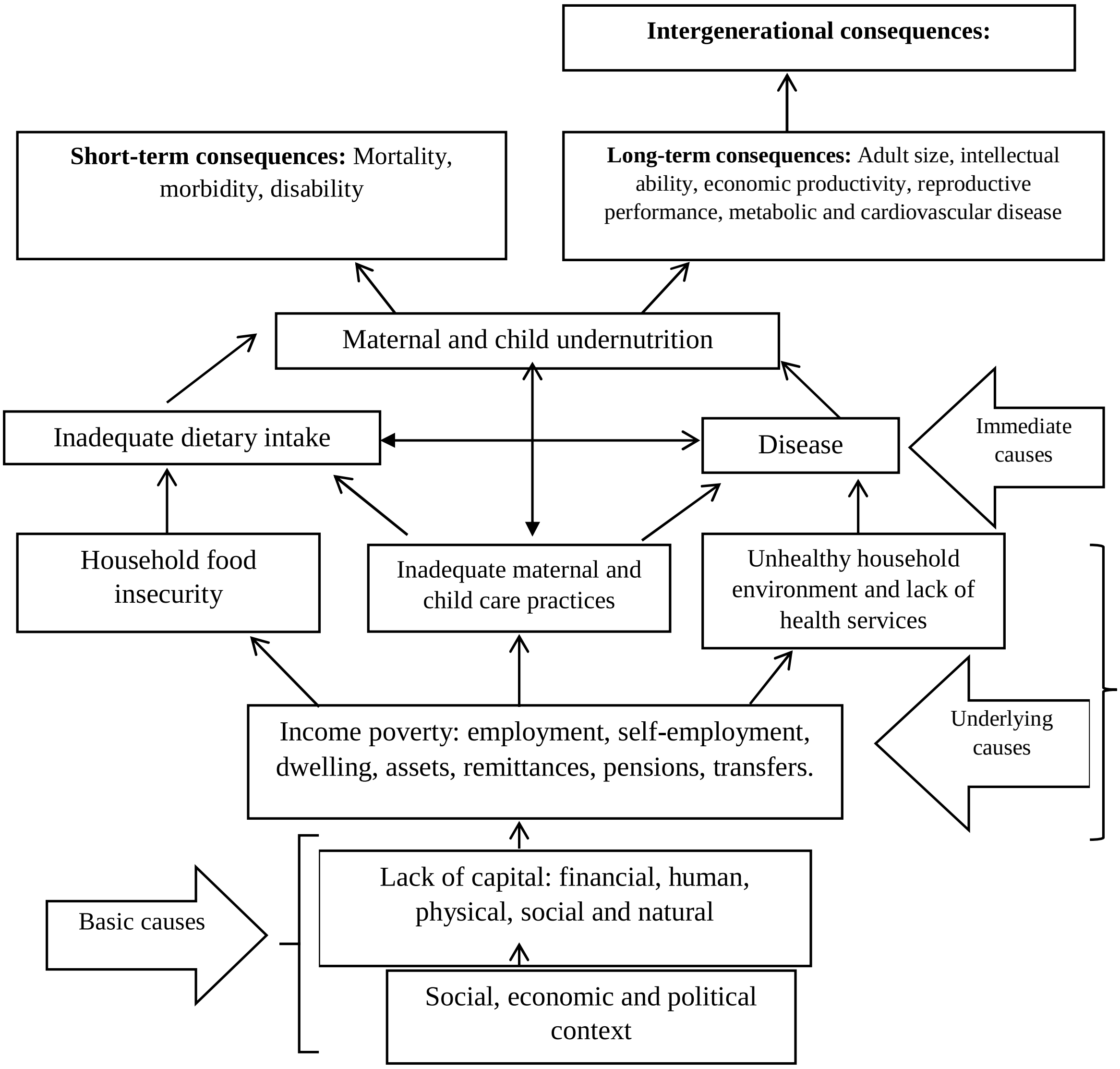


Figure 1.4: Framework of the relations between poverty, food insecurity, and other underlying and immediate causes to maternal and child under-nutrition and its short-term and long-term consequences. **Source: Adapted from (Black et al., 2008)**

1.3.3. The role of infection,environmental enteric dysfunction and the microbiota

Several studies indicate that environmental hygiene /sanitation is associated with the growth of children. Analysis of a longitudinal data on the effect of daily diarrhea on stunting showed that 25% of stunting was explained by five or more episodes of diarrheal before 24 months of age and concluded that recurrent diarrhea could have a deleterious effect on linear growth among young children(Checkley et al., 2008). Similar observationswere made with analysis of another longitudinal data which showed smaller but significant change on linear growth with a typical diarrheal burden compared to those with no diarrheal episodes(Richard et al., 2013).

Apart from symptomatic infectious conditions, subclinical ones are known to have a detrimental effect on the growth of young children (Campbell et al., 2003) and infections with pathogenic organisms can occur without diarrhea (Kotloff et al., 2013). Recently, great attention has been given to subclinical inflammation called environmental enteropathy (EE) or “tropical enteropathy” or environmental enteric dysfunction (EED). It is characterized by altered intestinal architecture (diffuse villous atrophy, crypt hyperplasia), increased permeability, T-cell infiltration and ultimately associated with nutrient malabsorption and growth faltering (Prendergast and Kelly, 2012, Humphrey, 2009, Keusch et al., 2013)as a result of ingestion of fecal bacteria by young children residing in an unhygienic environment (Korpe and Petri, 2012, Prendergast and Kelly, 2012, Dewey and Mayers, 2011) and other responses to environmental factors (exposure to mycotoxins etc.) (Khlangwiset et al., 2011, Egal et al., 2005). It was first identified among travelers from developed countries to developing and tropical countries in which there was exposure to unhygienic conditions. The sustained damage to the intimal mucosa allows the translocation of foreign macromolecules (e.g. bacterial lipopolysaccharides) which constantly activate the systemic immune system (Lunn, 2000, McKay et al., 2010, Korpe and Petri, 2012).

The continuing high prevalence of growth faltering or stunting among children in developing countries has been partly explained by EE(Gordon et al., 2012). The nutritional deficiency associated with EE is subtle and usually manifests itself in the form of chronic undernutrition or stunting (Humphrey, 2009). Two earlier observational studies from The Gambia demonstrated an association of intestinal inflammation with growth faltering starting from early infancy (Lunn et al., 1991, Campbell et al., 2003) and 43% of growth faltering in the first 15 months of life was explainedby the abnormal mucosal morphology and increased permeability (Lunn et al., 1991). Addressing the changes in the intestinal morphology and increased permeability was propsosed to have an effect on improving child growth (Campbell et al., 2003, Campbell et al., 2002). Another study in rural Malawi showed that most of the preschool children investigated had some evidence of EE and it was inversely associated with linear growth (Weisz et al., 2012).Later studies corroborated the link between environmental enteric dysfunction (EED) and linear growth restriction among children (Kosek et al., 2013) and environmental hygiene and growth (Lin et al., 2013) in which children residing in a clean environment are less likely to have parasitic infection and hence growth deficit.

The only available and widely used test of gut permeability is the lactulose: mannitol test (L:M test) or commonly known as dual sugar permeability test (Lunn, 2000). The person is allowed to ingest these two sugars whereby lactulose which is normally absorbed poorly will translocate easily in the presence of EE, while mannitol is absorbed completely in a normal circumstance. Measuring the levels of lactulose and mannitol in urine after two hours of ingesting is a validated and noninvasive marker of gut integrity (Korpe and Petri, 2012, van der Merwe et al., 2013). The L:M test requires a lot of cooperation and understanding from the participants and could be challenging when perfomed among very young children and contamination with fecal material should be avoided(Crane et al., 2015). Elevated lactulose: mannitol ratio indicates increased gut leakiness and malabsorption. However, the lack of local reference values, use of diffrerent assay techniques, specimen collection and data reporting make comparison among the various lietarature on L:M test. Fecal calprotectin (a cytoplasmic protein within neutrophils) has been also used as an indicator of active inflammation in patients with inflammatory bowel diseases but its use in EE is not known.

Environmetnal enteric dysfuction can altermetabolism(Semba et al., 2017) and nutrient absorption (Owino et al., 2016, Korpe and Petri, 2012) which could explain the reported functional abnormalities including growth faltering. A recent study in Malawi reported that there ia altered fatty acid oxidation, amino acid metabolism and deficiency of carnitine(an amino acid derivative and a transporter of long chain fatty acids to the mitochondria for β-oxidation and energy production) (Semba et al., 2017). A combination of interventions involving environmental sanitation and hygienic practices could prevent growth faltering among young children (Humphrey, 2009). However, intestinal mucosal permeability changes were not fully reversed by supplemental feeding or psychosocial stimulation among severely underweight children (Hossain et al., 2010) and it is not known if an alteration in gut morphology and barrier function can be ameliorated by nutritional interventions (Gordon et al., 2012, Dewey and Mayers, 2011). An omega-3 long chain fatty acid supplementation intervention targeting children with possible EE with the aim of improving linear growth showed no effect in other studies (van der Merwe et al., 2013, Smith et al., 2014).The changes in the intestinal wall in environmental enteropathy mimic morphologically the enteropathy seen in zinc deficiency and the relationship could be bidirectional. Zinc deficiency plays a part in altering intestinal barrier, inducing inflammation and infection (Lindenmayer et al., 2014, Prasad, 2009)but there is a lack of interventions combining micronutrient (e.g. Zinc) and other interventions (e.g. n-3 LCPUFA) with the aim of treating or ameliorating EE. Micronutrient supplementation showed a transient amelioration of EE among Malawian children and combining this with other interventions in most affected population was recommended (Smith et al., 2014) and a trial with of a single dose of albendazole and a 14days course of 20mg zinc sulfate or placebo among 1-3 years old Malawian children at high risk of EE showed a marked reduction in L:M ratio in the albendazole and zinc group compared to the placebo group (Ryan et al., 2014).However, another RCT among children 12–35 months old in rural Malawi, in which the intervention group received a single dose of albendazole, a 14 days of zinc at enrollment and after 20 weeks and a daily multiple micronutrient powder throughout the 24 week of study, did not decrease L:M ratio or stunting measured at 12 and 24 weeks (Wang et al., 2017).

Another factor which plays an important role in growth at an early age is the interaction between food, the immune system and the *microbiota/microbiome*in each subject**(Figure 1.5).** Assembly of the gut microbial community are reffered to as *microbiota*wheras the collection of microbial genes is called *microbiome*(Ahmed et al., 2014). The diversity, stability, resilience of the microbiota is affected by the type, timing and duration of of nutritional intervention given to address moderate or severe acute malnutrition and the vice versa (Gordon et al., 2012). Moreover, the gut microbiota differs between malnourished and well-nourished children (Crane et al., 2015). Animal studies demonstrated that microbiome from a Kwashiorkor child transplanted to a sterile gut of a mice which was on typical Malawian diet resulted in the mice to become malnourished (Smith et al., 2013) which demonstrated the relationship betweenperturbed microbiota and malnutrition and the vice versa.

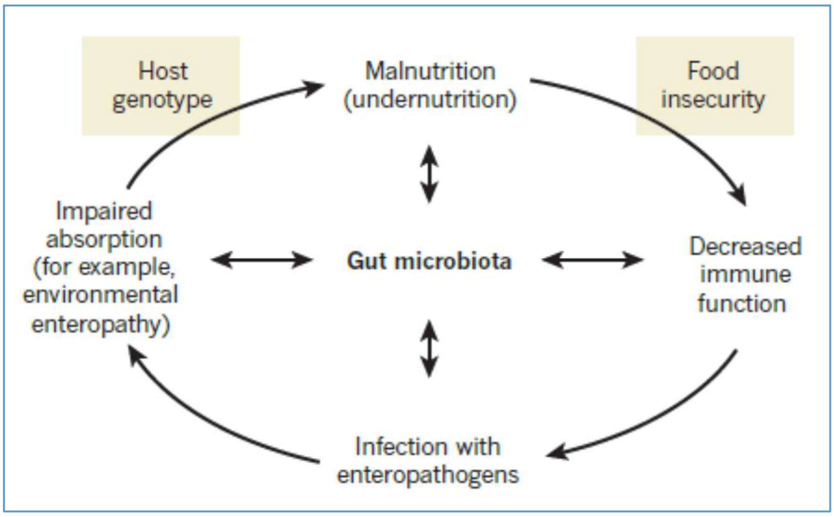


Figure 1.5: A schematic of the proposed relationships between the gut *microbiota*, the immune system and the diet, which underlie the development of malnutrition. Source (Kau et al., 2011)

1.3.4. Child feeding practices and nutritional status

It has been widely reported that child feeding behaviours in terms of caregiver and child interaction in the context of feeding; breast feeding and complementary feeding practices and nutrient intake from complementary foods are important determinans of nutritional status and growth among children. The role of above mentioned factors has been discussed elaborated under section **1.9.**

1.4. Consequences of malnutrition

Despite their limitations, epidemiological studies show that growth faltering starts in utero and continues for the first two years and results in various physical, health and neurodevelopmental consequences (Bhutta, 2013). More than a third of the disease burden (Black et al., 2008) and 45% of death (Black et al., 2013) in children under the age of five years and 11% of global Disability-Adjusted Life-Years (DALYs) can be due to undernutrition which makes undernutrition the largest risk factors for the global burden of disease in any age group (Bryce et al., 2008, Black et al., 2008, Black et al., 2013). Suboptimal breastfeeding alone contributes to 10% of disease burden in children younger than 5 years. Micronutrient deficiencies especially zinc, vitamin A, iron and iodine deficiencies play a significant role in disease burden among under-five children (Black et al., 2008).

During childhood, stunting is associated with impaired immunity, increased risk of mortality from infection (Olofin et al., 2013) through increased susceptibility to pneumonia, meningitis, diarrheal diseases. It affects schooling in children through impaired brain development mainly in cognitive domains but spatial navigation, locomotors skills are also affected(Grantham-McGregor et al., 2007).

Unless routinely measured, stunting is asymptomatic and it can easily be overlooked in communities where short stature is a norm and children who are stunted at an early age are more likely to remain stunted as adults (Coly et al., 2006, Stein et al., 2010). In a pooled data analysis from five birth cohorts (Brazil, Guatemala, India, the Philippines and South Africa), the authors estimated that every 1SD lower height-for-age at 2 years was associated with a 3.2cm lower adult height (Adair et al., 2013). Children who are stunted will have long term consequences which range from short stature as an adult to a reduced immunity, increased mortality, various chronic non-communicable diseases and reduced productivity and income earning capacity(Victora et al., 2008, Dewey and Begum, 2011, Huxley et al., 2000, Whincup et al., 2008). There is an increased risk of perinatal and neonatal mortality for women who are stunted (Lawn et al., 2005, Özaltin et al., 2010). Moreover, such outcome affects not only the individuals and communities directly involved but also the overall socioeconomic development of any nations (de Onis and Branca, 2016, Hoddinott et al., 2013a). The earning capacity of adults is reduced by 20% when they experienced stunting during childhood (Grantham-McGregor et al., 2007) and there is 1.4% loss in productivity for each 1% loss in adult height due to childhood stunting (The World Bank, 2006).

In summary, there is a huge interrelationship between the different causative factors which play at different levels. Underlying determinants lead to food insecurity, poor access to health care, hygienic environment, inadequate care practices which in turn expose children to inadequate dietary intake and illnesses and undernutrition (stunting) with its short and long term consequences. **(Figure 1.6).**

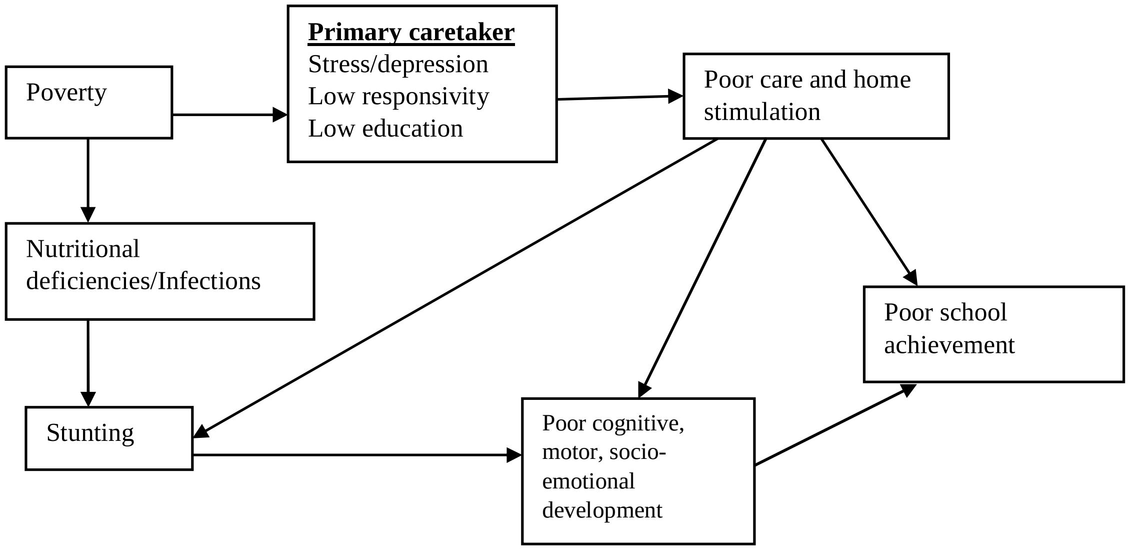


Figure1.6:Hypothesized relations between poverty, stunting, child development, and school achievement. Source: (Grantham-McGregor et al., 2007)

1.5. Child feeding practices andbehaviors

1.5.1. Maternal or caregiver feeding behavior or "style"

This section presents the behavioral or cultural aspect of child feeding or care practice in general. Good nutrition is a balance between food intake and utilization of energy and nutrients. This balance is influenced by diseases and mediated by care practices. The impact of care practice on nutrition is particularly important during the 1000-day window between conception and 2 years of age. The provision of complementary foods to young children according to the recommendation is essential; however, attention has been geared towards on how the foods are provided and fed to children with an implication on food intake, and nutritional status (UNICEF, 1997). Even if dietary intake and illness are immediate determinants of nutritional status in the UNICEF model, care practices are also identified as important factors which directly affect dietary intake and child health (Engle et al., 1999).

Care has been defined by Engle, 1999 (p.133) as"the behaviors and practices of caregivers to provide the food, health care, stimulation, and emotional support necessary for children's healthy survival, growth and development" (Engle, 1999). These practices translate food security and health care resources to child wellbeing(UNICEF, 1997). Caregiver-child interaction or caregiver feeding behavior (style) is one of the broad ranges of care practices affecting food intake, food preferences, weight status and general health status. The responsiveness of the care practices is crucial for child's survival, growth, and development (UNICEF, 1997).

In the past, much of the emphasis has been given to feeding practices without dealing with the behavioral aspect of feeding by parents or caregivers.Feeding practices of caregivers are goal oriented behaviors with the aim of affecting the feeding of their children through adoptions of various feeding practices (Vollmer and Mobley, 2013, Shloim et al., 2015). The issue of care practices (one of which is care during complementary feeding) was picked up starting from the early 90s (Engle et al., 2000, UNICEF, 1990) but incorporating care practice into the child feeding has been a challenge. Care practices are essential in promoting child growth and development in addition to household food security, provision of health care and appropriate hygiene and sanitation (Engle et al., 1999). Both under and over-nutrition may be due to difficulties in the interactions between caregivers and children as well as to a lack of high-quality foods(Moore et al., 2006) and care during feeding should be used as an opportunity for children to develop long-term feeding skills and a healthy appetite and food choices(Moore et al., 2006). Integrating care practices into nutrition or feeding has a potential to decrease the burden of growth impairment and promotion of child development (de Onis et al., 2013).

Care behaviors during feeding episodes (feeding styles) remain poorly investigated in relation to child health and nutritional outcomes. There is large body of literature on parental or caregiver feeding practices but very little is known about specific feeding patterns (feeding styles) of parents or caregivers and its effect on dietary intake, nutritional status and food preferences and considerable interest has been developed (Engle and Ricciuti, 1995, Engle et al., 2000, UNICEF, 1997, WHO, 1999). The conceptual framework developed by UNICEF and adopted by many countries stipulates that care practices are crucial for child growth, development and health (UNICEF, 1997). Child care practices or feeding styles are part of the parental cultural belief systems in which some feeding behaviors or styles are expressed accordingly.

The care practices during feeding often appear together in patterns that can be considered to represent an overall ‘style’ of feeding. Caregiver feeding practices can directly affect child health outcomes; however, feeding styles can influence child health outcomes indirectly through influencing feeding practices (Darling and Steinberg, 1993). The conceptualization of feeding styles or behaviors was first done by Birch and Fisher (1995) in which feeding styles were categorized as controlling, laissez-faire and responsive (active) (Birch and Fisher, 1995, Engle et al., 2000). A controlling feeding style iswhen a caregiver has complete control of when what and how much the child eats and includes force feeding the child. Controlling feeding style (Ventura and Birch, 2008) and nonresponsive feeding (Hurley et al., 2011) have been associated with the development of overweight or obesity among children and adolescents in the Western countries and indicated as one reason for suboptimal intake of energy from complementary foods in developing countries (Brown, 1997).

Cultures in different societies vary in many ways; one of which is on the behavior of caregivers over control of eating by children. Hence, feeding styles are rooted in cultural ethno-theories of parenting and care (Ha et al., 2002, Engle et al., 2000). It could be expressed in the form of total control by the caregiver or the child (Engle and Lhotska, 1999). When children are forced to eat, they are unable to regulate their own intake. Neither of these behaviors is desired for optimal growth and development outcomes and could lead to either underweight oroverweight or obesity depending on the behavior adopted by the caregiver (Brown et al., 1988). When too much control is in the hands of the caregiver, there is a tendency to over consume and that leads to overweight or obesity (Engle and Lhotska, 1999). This style has been observed in the West and is related to pediatric obesity risk (Johnson and Birch, 1994, Faith et al., 2004, Clark et al., 2007). The control of feeding might be also associated with restriction of foods that the caregiver thinks are unhealthy (less nutritious) and such styles were associated with increased urge to consume them and at times it leads to disinhibited eating (Fisher and Birch, 1999a, Fisher and Birch, 1999b, Faith and Kerns, 2005) with its undesirable consequences. Theoretically, too much caregiver control may also lead to high levels of food refusal and a dysfunctional caregiver–child interaction during feeding (Birch, 1998).

The terms parenting and feeding styles are usually used interchangeably even if there have not been studies which directly related the two; however, feeding is one of the earliest parenting practices(Jansen et al., 2012). Parenting style signifies the general and stable traits or behaviors parents have with regards to their interaction with their children (Jansen et al., 2012, Vollmer and Mobley, 2013); while parental feeding style refers to such interaction during feeding times (Ventura and Birch, 2008, Shloim et al., 2015, Hughes et al., 2005). Parenting style generally has two dimensions (responsiveness and demandingness) and there are four specific types of styles within these dimensions (Maccoby, 1992, Darling and Steinberg, 1993, Shloim et al., 2015): 1) authoritative parenting involves high demandingness and high responsiveness; 2) authoritarian parenting encompasses hight demandingness and low responsiveness; 3) indulgent parenting encompasses low demandingness and high responsiveness and; 4) uninvolved parenting in which it is associated with low demandingness and low responsiveness. Demandingness is manifested by high expectations and parental control; while, responsiveness involves support and recognition of cues of different forms (Darling and Steinberg, 1993) and it could be either parent-centered or child-centered (Hughes et al., 2005). In parental feeding styles the same responsiveness and demandingness dimension apply but in a feeding context (Hughes et al., 2005) **(Table 1.1).**

Table 1.1: Major parenting/feeding styles based on responsiveness and demandingness dimensions

Parenting/feeding style dimensions	**Responsiveness**		
		Low	High
Demandingness	Low	Neglectful / uninvolved	Permissive / indulgent
	High	Authoritarian	Authoritative

Source:(Vollmer and Mobley, 2013)

Feeding responsively can be particularly important for young children. Caregivers encourage, cajole, offer more helpings, talk to children while eating, and monitor how much the child eats. Mothers and other caregivers who show or model for children how to eat healthful foods will encourage children's eating, especially when food quality is low. The amount of food that a child consumes may depend as much on the caregiver's active encouragement of eating as on the amount offered (Bentley et al., 1991, Bentley et al., 1992, Engle and Zeitlin, 1996, Gittelsohn et al., 1998). Active feeding may not compensate for low-quality food but encouraging during eating leads to increased intake and might improve nutritional status and other desirable outcomes (Engle and Zeitlin, 1996).

Few studies in developing countries used the three categories of caregiver feeding styles; namely responsive, controlling, laissez-faire. In these contexts, responsive feeding involves adapting feeding methods based on child's needs and abilities, encouraging the child to eat the allotted amount, recognizing feeding cues and responding to them, verbalizing, avoiding distraction during feeding episodes and providing warmth and support (Engle and Ricciuti, 1995, Engle et al., 1999). This feeding style is critical especially at a younger age where children are totally dependent on the caregiver for feeding and other care needs and young children with a difficult temperament manifested by a refusal to eat and erratic appetite (Engle and Zeitlin, 1996, Gittelsohn et al., 1998, Bentley et al., 1991, Engle et al., 2000). On the other hand, controlling style is characterized by a total control of what the child is eating and can be associated with forced feeding or control could be left for the child. This style is not desirable as it may lead to either obesity or lack of regulation of food intake or undernutrition on the other extremity. The third type of feeding style according to Birch and Fisher was the Laissez-faire style (Birch and Fisher, 1995). Laissez-faire style involves a passive approach and low levels of caregiver-child interaction. The caregiver makes little effort to encourage eating, and may often expect children to eat on their own at an early age. These attitudes may reflect the belief that children know best how much they should eat. These attitudes may be detrimental when children have low appetites (Engle et al., 1999). Many of the caregivers in developing countries practice laissez-faire style and they usually interact only when the child refuses to eat (Engle and Zeitlin, 1996).

As a result of diverse conceptualization, definitions and measurement of caregiver feeding styles, the findings from various sources are difficult to compare (Jansen et al., 2012). A study using the three categories of feeding styles (responsive, controlling and Laissez-faire) together with direct observation of feeding episodes demonstrated that caregivers physically support children while eating and verbalization was associated with increased food acceptance (Dearden et al., 2009) whereas child food rejection was associated with pressure or force(Ha et al., 2002, Bentley et al., 2011). Active feeding behaviors has been reported to be associated with good acceptance of food(Bentley et al., 1991, Bentley et al., 1992, Gittelsohn et al., 1998, Engle and Zeitlin, 1996) have been included as research agenda(Engle et al., 2000) and programs(UNICEF, 1997). In general, appropriate feeding style means that caregivers be proactive instead of responding to food rejection or aversion, illness or loss of appetite (Engle and Zeitlin, 1996, Bentley et al., 1991).

Even if the interventions conducted so far regarding the effect of responsive feeding on food acceptance, food intake, and growth lack specificity (Engle and Pelto, 2011), the role of responsive feeding for food acceptance (Bentley et al., 2011, Dearden et al., 2009), food intake and growth (Hurley and Black, 2011), development (Engle and Pelto, 2011, Vazir et al., 2013) and prevention of childhood overweight or obesity in countries undergoing nutrition transition has been highlighted by many organizations in both high, low and middle-income countries(Engle and Pelto, 2011). Nevertheless, little is known in many developing countries about what type of feeding “styles” parents/caregivers practice and their effect on food acceptance, dietary intake, health and nutritional status of children.

1.6. Infant and young child feedingpractices

Optimal infant and young child feeding (IYCF) is essential for child growth during the first two years which is considered a “critical window of opportunity” for the prevention of growth faltering, and subsequent long-term consequences (Mukuria et al., 2006). Feeding in the first two years set a stage for optimal growth and development of young children and several benefits were identified for the mother(Arabi et al., 2012, Victora et al., 2016, Rollins et al., 2016). The first two years of life is the period when the recommended infant and young child feeding practices are implemented (PAHO/WHO, 2002); namely, exclusive breastfeeding for the first 6 months, continued breastfeeding to 2 years or beyond together with adequate, safe, and appropriate complementary feeding from 6 to 23 months (UNICEF, 2013). According to the Lancet Child Survival Series (2003), breastfeeding for the first 12 months and initiation of complementary feeding at six monthscontributed to the prevention of one-fifth of child deaths (Jones et al., 2003). The challenge is that socioeconomic development has not been directly translated to optimal IYCF practices in many developed countries(Arabi et al., 2012).

1.7. Current practice of infant and young child feeding

1.7.1. Initiation and continued breast feeding

The problem of inadequate breastfeeding practices has been common to both developed and developing countries in which the progress for the early initiation of breastfeeding has been very slow (Victora et al., 2016). The scale-up of breastfeeding at universal level could avert significant number of death of both children under the age of 5 years and women (through prevention of breast cancer)(Victora et al., 2016). Currently, only 45% of newborns are started with breast milk within the first hour of birth (UNICEF, October, 2016), and 37% of the infants less than 6 months of age are exclusively breastfed (Victora et al., 2016). Only three-quarter of children aged12-15 months are still breastfeeding and about half the children aged 20-23 months are breastfed(UNICEF, October, 2016). Continued breastfeeding to until 2 years of age is one of the recommendations tracking behind in terms of actual practice in which low-income countries are at an advantage. Exceptfor the prevalence of early initiation of breastfeeding and exclusive breastfeeding, the prevalence of other indicators of breastfeeding are higher among low-income countries (Victora et al., 2016).

1.7.2. Complementary feeding practice

The World Health Organization recommends that infants should begin to eat solid, semisolid or soft foods at 6 months of age to ensure that their nutrient intake is sufficient to fuel their developing brains and bodies as the breast milk cannot provide sufficient amount of energy and much of the micronutrients (Butte et al., 2002, WHO and UNICEF., 2003, PAHO/WHO 2003, Dewey and Brown, 2003). This is also the period when much of the growth faltering is happening (Victora et al., 2010). At this period breast milk remains an important source of energy and micronutrients. Children who were started with appropriate complementary food during 6-8 months have a lower risk of stunting and underweight (Bhutta et al., 2013, Marriott et al., 2012) but many both in high and low-income countries tend to start earlier with nutritionally inadequate food with no added benefit (UNICEF, October, 2016). Such practice exposes infants to infectious agents and leads also to the displacement of breast milk.

While the benefits are clear, about one-third of 6-8 months old infants were not started with solid foods globally(UNICEF, October, 2016) without taking the breastfeeding status into consideration. Several factors came into play for starting solid foods earlier than 6 months such as cultural practices, lack of adequate time by the caregiver, government policy regarding maternity leave and unregulated advertisements from the commercial formula producers (Martin et al., 2008, Alder et al., 2004, Butte et al., 2002). Apart from the timing of initiation of appropriate complementary food, the amount, diversity, and frequency of feeding are equally important as the requirement for energy and nutrients is the highest per body weight in this age group compared to any other age(Butte et al., 2000, FAO/WHO, 2004). To meet the minimum meal frequency breastfed children aged 6-8 months need to eat at least two meals or snacks a day and those 9-23 need at least three meals or snacks a day(WHO, 2010b).

Half of all children are not receiving the minimum meal frequency globally and Eastern and Southern Africa has one of the lowest figures in terms of this indicator (~10%)(UNICEF, October, 2016)**(Figure 1.7)**.It has been shown that consumption of four or more food groups a day is associated with improved linear growth in young children (Onyango et al., 2014). Less than a third of infants and young children in the worldandonly one in five in East and Southern Africa are getting the recommended number of food groups and the problem gets worse among the lowest socioeconomic group(UNICEF, October, 2016).The type of food groups in the diet of infants and young children are equally important as the diversity in which food groups from animal origin are more likely to provide the required amount of micronutrients(PAHO/WHO 2003, Dror and Allen, 2011); Dewey, 2013 #839}. The other aspect of complementary foods is whether they are given appropriately or not at each feeding. This implies frequency of feeding, diversity and appropriate caregiver-child interaction (responsive feeding). Minimum acceptable diet is a composite indicator of feeding for children 6-23 months including both minimum meal frequency and dietary diversity(WHO, 2008). Only one in six and one in ten children 6-23 months of age receive a minimum acceptable diet globally and in Easter and Southern Africa(UNICEF, October, 2016).

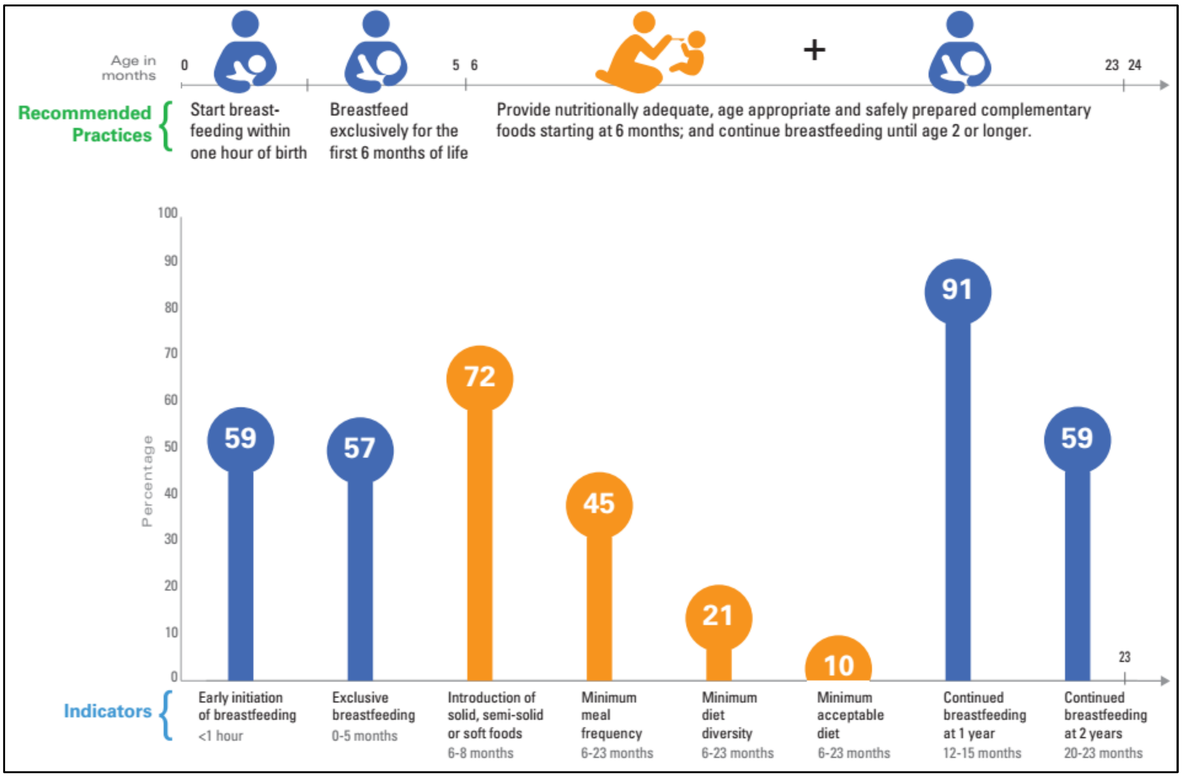


Figure 1.7. Percent of children: put to the breast within one hour of birth, exclusively breastfed (0-5 months); introduced to solids (6-8 months), with a minimum meal frequency, minimum diet diversity and minimum acceptable diet (6-23 months) and continued breastfeeding at 1 year (12-15 months) and 2 years (20-23 months), 2015 in Eastern and Sothern Africa.

Source: UNICEF global databases, 2016, based on MICS, DHS, and other nationally representative surveys, 2010-2016. Available at: **https://data.unicef.org/topic/nutrition/infant-and-young-child-feeding/#**

1.7.3. Hygienic preparation and storage

Safe preparation and storage are as important as providing adequate nutrition to the growing child. The World Health Organization estimates that infections associated with unsafe water, poor sanitation and unhygienic practices are contributing to 50% child undernutrition (Prüss-Üstün et al., 2008, Prüss-Üstün and Corvalán, 2006); diarrheal diseases are the second leading cause of death in under-five children globally (Liu et al., 2012). Hygienic preparation and storage of complementary foods are hampered by a very limited safe water availability and hand washing practices in many African countries. Less than 20% of the 11 countries studied in Eastern and Southern Africa have hand washing facilities with soap and water(UNICEF, October, 2016).

1.8. Measurement of infant and young child feeding

1.8.1. Overview of the guiding principles of infant and young child feeding

Child feeding is known to be a continuum and multidimensional practice consists mainly of breastfeeding and complementary feeding (Arimond and Ruel, 2003b, Arimond and Ruel, 2002a)**(Figure 1.8)**. Pan American Health Organization and the World Health Organization published theGuiding Principles for Complementary Feeding of the Breastfed Childin 2003(PAHO/WHO 2003). The 10 guiding principles include: “(1) duration of exclusive breastfeeding and age of introduction of complementary foods; (2) maintenance of breastfeeding; (3) responsive feeding; (4) safe preparation and storage of complementary foods; (5) amount of complementary food needed; (6) food consistency; (7) meal frequency and energy density; (8) nutrient content of complementary foods; (9) use of vitamin-mineral supplements or fortified products for infant and mother; and (10) feeding during and after illness”.

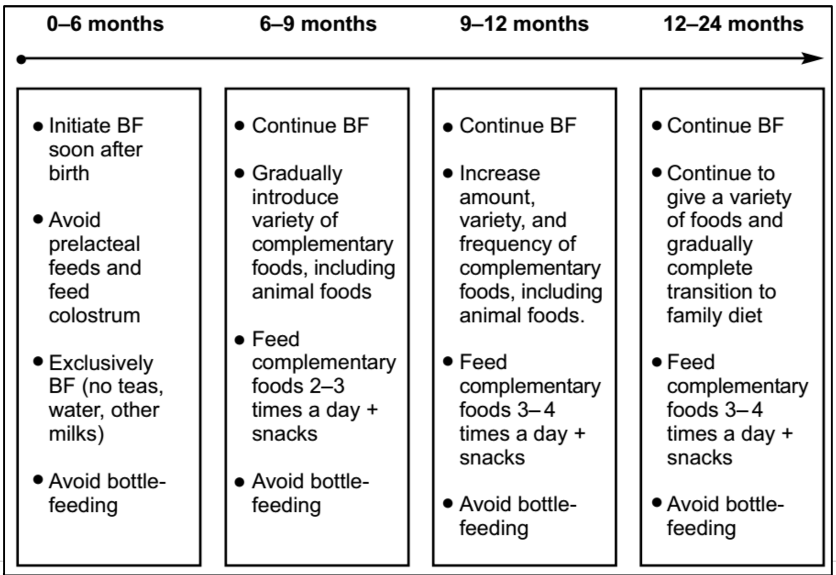


Figure 1.8. The continuum of child feeding. Source: (Arimond and Ruel, 2003b); BF= breast feeding

Measuring child feeding practices has been an issue of investigation for some time and as a result, different sets of indicators of measurement have been proposed (PAHO/WHO 2003, Dewey et al., 2006, WHO, 2008). For breastfed children, the guiding principles for the feeding of infants and young children suggested by WHO include exclusively breastfed from birth to 6 months and should continue breastfeeding on demand until 24 months or beyond (PAHO/WHO 2003). Complementary food should be started at 6 months of age and fed with increasing amount and density as the child grows.

1.8.2. Indices of infant and young child feeding practices

Despite the development of the global guidelines and recommendations for feeding, the adaptation of simple and valid indicators for assessment, monitoring, and evaluation of programs is needed. The development and further adaptation and validation of feeding indices are crucial in addressing the problem of the measurement of different feeding patterns and modes. Indices can be composed of different feeding dimensions and practices so that wider aspects of care practices can be captured (Ruel and Menon, 2002). Feeding practices are rather a spectrum, hence there is a minimum such practice which leads to certain outcome (Arimond and Ruel, 2003a). The clustering of feeding practices can also be easily captured using composite indices of child feeding practices (Arimond and Ruel, 2003a, Arimond and Ruel, 2003b). Moreover, the composite indices are age specific and allow for the determination of sample size and later analysis depending on the specific subgroup (Ruel and Menon, 2002). Analysis of the association of indices of feeding practice and child outcomes can be done when sample size takes into account the age group under consideration.

Several attempts had been made to construct child feeding indices by various authors and the earliest index created was by Ruel, et al (1999) using child feeding practices and the use of preventive health care services(Ruel et al., 1999). The authors used breastfeeding practices and preventive health services such as growth monitoring and immunization to develop the “care index”. Multiple child feeding practices or modes were used by Ruel et al (2002) for the first time to construct Infant and Child Feeding Index (ICFI) from children starting from 6 months of age using DHS data from

Ethiopia(Arimond and Ruel, 2002b)and later on several Demographic and Health Survey data were used as data sources (age of children 6-36 months) (Ruel and Menon, 2002).Additional complementary food related practices were added to the index created addressing the earlier limitation of use of only breastfeeding related practices.

Analysis of data from eleven developing countries used a dietary diversity score calculated from seven food groups instead of six (Arimond and Ruel, 2004). The seven-point dietary diversity score included: 1) starchy staples (foods made from grain, roots, or tubers); 2) legumes; 3) dairy (milk other than breast milk, cheese, or yogurt); 4) meat, poultry, fish, or eggs; 5) vitamin A-rich fruits and vegetables (pumpkin; red or yellow yams or squash; carrots or red sweet potatoes; green leafy vegetables; fruits such as mango, papaya, or other local vitamin A-rich fruits); 6) other fruits and vegetables (or fruit juices); and 7) foods made with oil, fat, or butter. Those foods/groups consumed for ≥3 times per week were given a score of 1 otherwise, 0 was assigned. Then age specific terciles of the dietary diversity were used to categorize children as having low, average or high diversity (Arimond and Ruel, 2004).

A set of feeding indicators were developed to monitor complementary feeding practices globally mostly commissioned by WHO. The first such indicators were developed in 2001 where breastfeeding for 6 months was recommended instead of the 4-6 months earlier (WHO, 2001). In order to address the complementary feeding adequacy and frequency of feeding, WHO organized working groups to develop indicators of complementary feeding for both breastfed and non-breastfed mainly in developing countries(Ruel et al., 2003, PAHO/WHO, 2002, WHO, 2005, Dewey and Brown, 2003). Another Working Group on infant and young child feeding indicators was established in 2004 and worked on various indicators and definitions. Later onseveral indicators and their validation was reported by Dewey et al (Dewey et al., 2006); which were then used as an input for the WHO's Infant Young Child Feeding Indicators definition, measurement guides for core and optional indicators (WHO, 2008, WHO, 2010b)**(Table 1.3).**Different recommendations were made for breastfed and nonbreastfed children regarding complementary feeding.

Table 1.3: Core indicators of infant and young child feeding used in measurement of IYCF practices*

S.N	Indicators	Definitions
1.	Timely initiation of breastfeeding (children 0-23 months)	Proportion of children 0-23 months who were put to the breast within one hour
2.	Exclusive breastfeeding under 6 months	Proportion of infants 0-5 months of age who were fed exclusively with breast milk in the past 24 hours
3.	Timely complementary feeding	Percent of infants 6-9 months of age who receive breast milk and a solid or semi-solid in the previous 24 hours. Solid, semi-solid and soft foods are defined as mushy or solid foods, not fluids
4.	Introduction of solid, semi-solid or soft foods	Proportion of infants 6-8 months who receive solid, semi-solid or soft foods
5.	Continued breastfeeding at 1 year	Proportion of children 12-15 months old who are fed breast milk
6.	Minimum dietary diversity	The proportion of children 6-23 months who received food from 4 or more food groups in the past 24 hours : The 7 food groups used to calculate this indicator are: 1. Grain, roots tubers; 2. Legumes and nuts; 3. Dairy products (milk, yogurt, cheese); 4. Flesh foods (meat, fish, poultry, liver/organ meats); 5. Eggs; 6. Vitamin A rich fruits and vegetables; 7. Other fruits and vegetable
7.	Minimum meal frequency	The proportion of breastfed and non-breastfed children 6-23 months of age who receive solid, semi-solid or soft foods the minimum number times or more. Minimum is defined as — 2 times for breastfed infants 6–8 months — 3 times for breastfed children 9–23 months — 4 times for non-breastfed children 6–23 months
8.	Minimum acceptable diet	The proportion of children 6-23 months of age who receive a minimum acceptable diet (apart from breast milk). **Note**: • For breastfed children, see indicators 6 and 7 above for “Minimum dietary diversity” and “Minimum meal frequency” definitions. • For nonbreastfed children, see indicator 7 above for definition of “Minimum meal frequency”. The definition of “Minimum dietary diversity” is similar to the definition for indicator 6, but milk feeds are excluded from the diversity score for nonbreastfed children when calculating “Minimum acceptable diet”.
9.	Consumption of iron-rich oriron-fortified foods	Proportion of children 6-23 months old who receive an iron-rich food or iron-fortified food that is specially designed for infants and young children or that is fortified in the home

*Source: WHO indicator guide (WHO, 2008)

1.9. Infant and young child feedingpractices and child outcomes

1.9.1. Overview

It is acknowledged that infant and child feeding is are important factors for child growth and development outcomes(Arabi et al., 2012) and the first two years are especially important as optimal nutrition during this period leads to improved growth and development (WHO, 2008) and reduced risk of mortality, morbidity, and chronic non-communicable diseases (Black et al., 2008).However, how strong of infant and child feeding practice in a given childis related to growth and health outcomes remains to be an important area for research for the last decades or more. The development of infant and young child feeding indicators is crucial to monitor the changes in the practice in a given community or country at large (WHO, 2008). Most assessments used one indicator at a time which results in a loss of information and lack of knowledge about the association of overall feeding and single feeding practice/dimension and association of a feeding indicators with child health, growth, and other outcomes. A single behavior of infant and child feeding practice is usually is not robust enough to be able to indicate child nutrition and health outcomes; hence, the need for a validated composite index to monitor changes in care practices. Moreover, feeding practice recommendations are age dependent and there is a problem of age censoring in which child feeding practice questions are asked about a child who is still too young for that feeding mode (Arimond and Ruel, 2003b). The other dimension that was advocated to be included has been the child-caregiver interaction during feeding (Arimond and Ruel, 2002a). Moreover, child feeding or care practices tend to cluster in which a caregiver who has a favorable practice in the early age of the child will most likely have a favorable practice in child feeding at a later age due to better awareness and knowledge(Arimond and Ruel, 2003b).

1.9.2. Child feeding practices and growth

The relationship between child feeding practices and child outcomes has been mixed. Among other care practices, child feeding practices have been associated with child growth and nutrient adequacy and other outcomes (Arimond and Ruel, 2003a, Arimond and Ruel, 2003b). Ruel & Menon (2002) demonstrated for the first time that infant and child feeding index was positively associated with height-for-age based on data from seven Latin American countries (Ruel and Menon, 2002). The same association was observed among the rural population in Ethiopia (Arimond and Ruel, 2002b). Arimond & Ruel demonstrated using DHS data that dietary diversity was associated with HAZ as a main effect or in an interaction in10 of the 11 countries whose data were analyzed (Arimond and Ruel, 2004).Food variety scores (mean number of different food items consumed from all possible items eaten) and dietary diversity scores (mean number of food groups out of nine possible groups) were correlated with HAZ and WHZ among 1-8 years old South African children(Steyn et al., 2006).

In another analysis of samples from 14 low-income countriesthe WHO feeding indicators (WHO, 2008) were examined for their association with child stunting or underweight. The introduction of solids at 6–8 months, consumption of solid foods, meeting the WHO guidance for minimum acceptable diet, iron-rich foods, and dietary diversity intake were associated with reduced risk of stunting and underweight (Marriott et al., 2012); moreover,early initiation of breastfeeding was associated with reduced risk of underweight. In the latter analysis maternal education was found to be an important factor in the reduction of underweight and stunting. A similar analysis was performed based on DHS data from Ethiopia and Zambia, and it was shown that dietary diversity score was associated with HAZ in both countries (Disha et al., 2012). However, there were inconsistencies in other indicators of infant and young child feeding. Several studies showed that dietary diversity/variety is a good predictor of nutritional status or growth; however, one of the reasons discussed was the lack of consistency in term of calculation of dietary diversity, control of confounding factors (Arimond and Ruel, 2004) and considering interaction with other factors such as breastfeeding. In the latter case, some researchers found astronger association between dietary diversity and HAZ among children who were not breastfed (Marquis et al., 1997a, Marquis et al., 1997b).

Using an adapted version of the ICFI in Burkina Faso, the authors found that there was a significant association between ICFI and HAZ among children 6-36 months old (Sawadogo et al., 2006); on the contrary the study in Senegal by Ntab et al, did not demonstrate an association between ICFI and HAZ or linear growth velocity among 12-

42 months old children (Ntab et al., 2005). In another study in Madagascar among children from 6-23 months, the authors found no association of ICFI with LAZ when ICFI was cut into categories (Moursi et al., 2009) and adjusted for covariates but another study in China found an association of cross-sectional ICFI with WAZ and WLZ. In the latter study, the authors found an association between ICFI and LAZ when the breastfeeding was removed from the index. Mixed results were found for the associations of components ICFI with the growth of children in different settings (Moursi et al., 2008b, Ma et al., 2012).

The stability of ICFI was analyzed based on three cross-sectional ICFI data collected from the same children over a period of time(Moursi et al., 2008b). It was shown thatgoing from low to high longitudinal ICFI (constructed from repeated cross-sectional ICFIs), there was a highly significant difference in mean LAZ. The stability of ICFI over time was also observed and the finding suggested a better precision obtained by using ICFI calculated from repeated measurements (Moursi et al., 2008b). The use of longitudinal ICFI and its stability over time has been replicated in studies in other settings. A follow-up of children of 5-7 months in China demonstrated that ICFI at 6 months was associated with growth at 18 months and the index was stable over time (Ma et al., 2012). A study in Ethiopia used longitudinal ICFI based on data from HIV-exposed infants (Haile et al., 2014). The latter study showed that the longitudinal ICFI was associated with LAZ and WAZ after controlling for measured confounding variables.

1.9.3. Association of child feeding practice and micronutrient intake

The use of simple indicators of dietary status has been proposed to be indicative of energy and nutrient intake among children in many developing countries (Hatloy et al., 1998, Onyango et al., 1998, Tarini et al., 1999, Moursi et al., 2008a).

The relationship between feeding practice indicators and dietary quality (adequacy) in terms of micronutrient intake has been studied by various authors. The different studies attempted to validate dietary diversity or food variety scores as an indicator of dietary quality through sensitivity and specificity analyses. A food variety score (FVS) and dietary diversity score (DDS) were found to be correlated with mean adequacy ratio (the mean of nutrient adequacy ratio calculated for each nutrient) among children 1-8 years from South Africa (Steyn et al., 2006) and among 13-58 months old children in urban area in Mali (Hatloy et al., 1998). Based on the trade-offs between sensitivity and specificity, a DDS of 4 and an FVSof 6 were the best indicators of mean adequacy ratio (MAR) of less than 50% (Steyn et al., 2006). Moursi et al (2008) found that dietary diversity, calculated from 7 food groups was a good predictor of micronutrient density adequacy (the mean of micronutrient density adequacy of 9 or 10 nutrients depending on the age and breastfeeding status and further capped at 100% to avoid bias by some high consumers).

In conclusion, despite the reported association of dietary diversity with HAZ, it has been quite inconsistent as a result of the limitation of measurement of dietary diversity, the use of dietary diversity calculated from food groups consumed over24 hours or one week period. Moreover, identifying cut-offs indicating nutrient adequacy and nutritional status remains to be investigated in different settings where there are diverse food consumption patterns and staple diet. The investigation about the importance of simple feeding practice indicators as predictors of growth or micronutrient intake or adequacy is far from complete.

1.10. Seasonality of food intake and child outcomes

It is widely acknowledged that dietary quality is associated with improved child growth, yet there had been limited emphasis given to seasonal changes in dietary intake(Savy et al., 2006) and the need to consider seasonal variation in food and nutrient intake when planning interventions aiming at addressing micronutrient deficiency in developing countries. Especial attention has been given to pre-harvest (lean or rainy) seasons in which the produce from the previous harvest season got depleted and food prices are high on the market and people's diet tends to be less nutritious in terms of the content of essential micronutrients(Vaitla et al., 2009, Abay and Hirvonen, 2016). This season is characterized by increased risk of infectious morbidity especially among the most vulnerable groups (young children).

Based on a nationwide estimate using consumption data collected at a household level, cereals and roots and tubers are the major source of energy in both rural and urban population in Ethiopia while animal source foods contribute to 1.7 and 3% in rural and urban settings respectively (Hirvonen et al., 2015). Cereals only contribute to 60% of the energy intake in both urban and rural settings in Ethiopia; however, seasons play a role in the proportion of cereal or non-cereal sources of energy in which less non-cereal sources (only around 30%) are used during the lean season (Hirvonen et al., 2015). Based such representative data from households over one year period in Ethiopia, the authors also found a drop in household dietary diversity in both urban and rural households during the two fasting periods of the Orthodox followers in December and March (Hirvonen et al., 2015). However, the household dietary diversity in July was comparable with the post-harvest period (February) and it was not affected by the lower energy consumption **(Figure 1.8 A).** There was also a drop in the per capita energy intake of individuals studied in the month of June to July (lean season) among rural households compared to the case in February (post-harvest season) (Hirvonen et al., 2015) (**Figure: 1.8 B**).

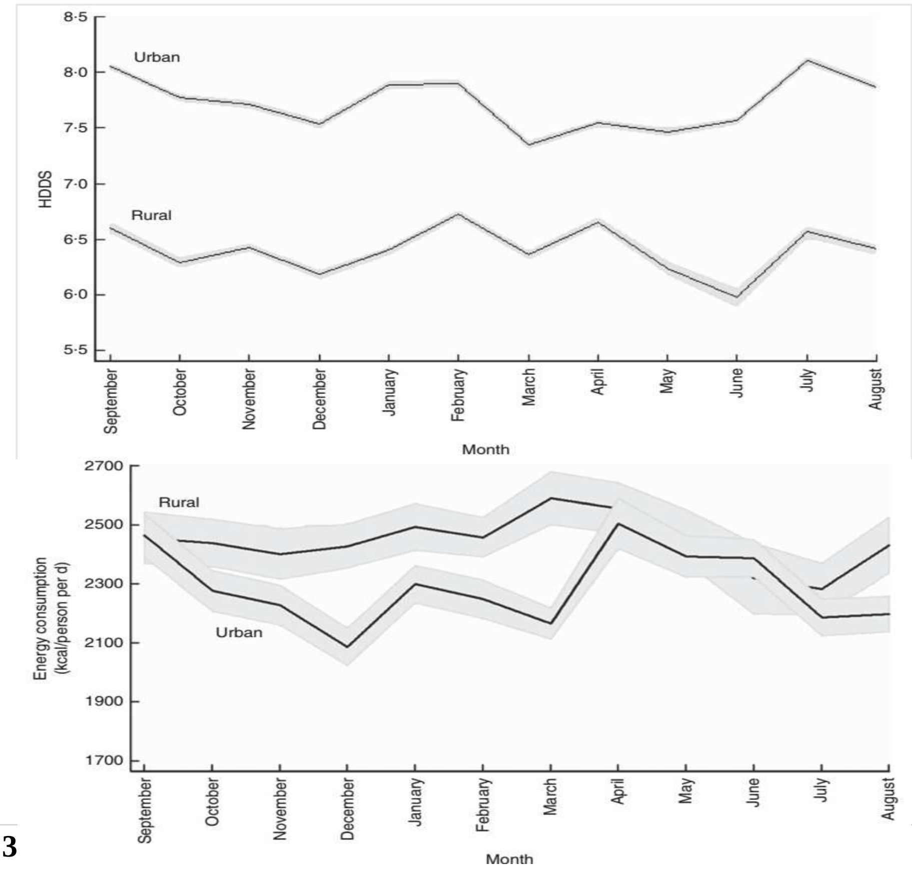


Figure: 1.8 Seasonal patterns in meanhousehold dietary diversity score (HDDS) (**A**) Seasonal patterns in mean daily per capita energy intake in kcal (**B**), by rural/urban setting, among 27 835 households from eleven regions of Ethiopia;

The solid line gives the mean for each calendar month and the grey area represents the 95 % confidence interval

Adapted from: (Hirvonen et al., 2015)

Hence, the use of dietary diversity as an indicator of dietary quality should be interpreted cautiously and should be used at individual level (Hirvonen et al., 2015). Seasonal fluctuations in food intake and dietary diversity can be partly offset by market integration or by being closer to a market where food can be purchased or sold in Ethiopia(Hoddinott et al., 2015, Abay and Hirvonen, 2016).

Similar observations were made in other countries. There was an increase in the average dietary diversity score of women at the end of a lean season by around 11% in Burkina Faso as a result of consumption of diverse food items which are cheaper and accessible(Savy et al., 2006). Dietary quality expressed in the form of mean probability adequacy (MPA) was assessed among women and children in Burkina Faso and it was reported that intake of most micronutrient was slightly higher in the post-harvest season (Arsenault et al., 2014).

1.11. Interventions during the complementary feeding period

The period of complementary feeding (6-24 months) is characterized by high rate of growth faltering, high nutrient gap, and consumption of relatively small amount due to the size of their stomach. Hence, complementary foods need to be nutrient dense (high amount per 100kcal) (PAHO/WHO 2003, Dewey and Vitta, 2013). However, evidence from complementary feeding interventions conducted in the past provided the weakest link in terms of supporting healthy growth in children (de Onis et al., 2013). According to a systematic review of 29 efficacy and 13 effectiveness trials of complementary feeding interventions in developing countries, provision of complementary foods resulted in mean effect sizes of 0.26 (range: -0.02 to 0.57) for weight (WAZ) and 0.28 (range: -0.04 to 0.69) for linear growth (HAZ). The presence nutrition education intervention in addition to the provision of complementary food resulted in an effect sizes of 0.35 (range: 0.18 to 0.66) for weight and 0.17 (range: 0 to 0.32) for linear growth. Improving the energy density of the usual complementary foods had mean effect sizes of 0.35 (range: -0.13 to 1.37) for weight and 0.23 (range: -0.25 to 0.71) for linear growth (Dewey and Adu-Afarwuah, 2008). Such effect has been considered modest(Dewey and Adu-Afarwuah, 2008). Micronutrient supplementation trials reviewed did not provide an effect on growth and the use of milk products and essential fatty acids were recommended. The effect of complementary food provision (± nutrition counseling) was higher in settings where there was a high rate of malnutrition and food insecurity (Imdad et al., 2011, Bhutta et al., 2008) and more significant effect observed if nutrition specific and sensitive interventions are combined (Bhutta et al., 2013). Using a linear programming techniques(Briend et al., 2003), the use of the hypothetical complementary diets in Ethiopia, Vietnam, and Bangladesh did not meet the requirements for “problem nutrients” such as zinc and iron for the age groups 6-8 and 9-11months(Vitta and Dewey, 2012).

1.12. Fats in the diets of young children

1.12.1. Enrichment of complementary foodswith fats

Fats are important energy sources for young children and half of the energy from breast milk is contributed by fats. However, the quality of fats in the diet of infants was not given much attention until specific deficiency symptoms (e.g. skin changes) and poor growth were noted among those infants consuming skimmed milk based formula devoid of linoleic acid(Hansen et al., 1963). Later on, a case study on the consumption of high linoleic acid and low in linolenic acid lead to the development of neurologic symptoms in a 6 years old child which was corrected with replacement with a diet containing linolenic acid (Holman et al., 1982).

As a result of the difficulty of meeting the demands of young children for essential nutrients through typical complementary foods in many developing countries (Dewey, 2013), enriching complementary could be the only available solution (Dewey and Vitta, 2013). Such improvement can be made through fortified blended foods, micronutrient powders and complementary foods supplements (e.g. LNS). The latter ones could be used to add essential fatty acids to traditional complementary food or blended products as intake of essential fatty acids is extremely low in many developing countries (Michaelsen et al., 2011).

Linoleic acid (18:2 n-6) and α-linolenic acid (18:3 n-3) (ALA) are the two parent essential fatty acids (EFA) which are not synthesized in the body and need to be consumed in the diet (Koletzko et al., 2008, Michaelsen et al., 2011). The n-6 fatty acid is converted to its longer chain metabolites such as arachidonic acid (AA) and n-3 fatty acid ALA is converted to eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (**Figure 1.9**). LA and ALA are not inter-convertible but compete for the ∆6-desaturase in the synthesis of long-chain PUFA (Gibson et al., 2011). The disruption of the normal ratio of n-6 to n-3 PUFA is key for the production of either proinflammatory or anti-inflammatory metabolites (Simopoulos, 2002); in which an increased intake of n-6 promotes the proinflammatory state.

The rate of conversion of ALA to EPA (5%), and to DHA (0.5%) is very low (Harris et al., 2009); and lower for children compared to adults (Uauy and Castillo, 2003, Uauy and Dangour, 2009). The presence of iron, zinc, vitamin B6 and E are important for the conversion process of ALA to EPA and DHA and LA to AA (Smuts et al., 1995, Smuts et al., 1994, Agostoni et al., 2007). Iron and zinc are important in fatty acid desaturation while the others could be used as antioxidants. Inareas with high rate of micronutrient deficiency (mainly iron, zinc, selenium) conversion of LA and ALA to their respective metabolites could be limited (Smit et al., 2004).

The potential role of essential fatty acids in growth has been a topic of discussion in the past decade (Lauritzen et al., 2001, Lauritzen et al., 2005a, Lauritzen and Carlson, 2011, Makrides et al., 2011, Prentice and van der Merwe, 2011) and it is a subject of continuing research at present in terms their roles for promoting healthy growth and development in the first few years of life, reducing morbidity and subclinical inflammation in children.

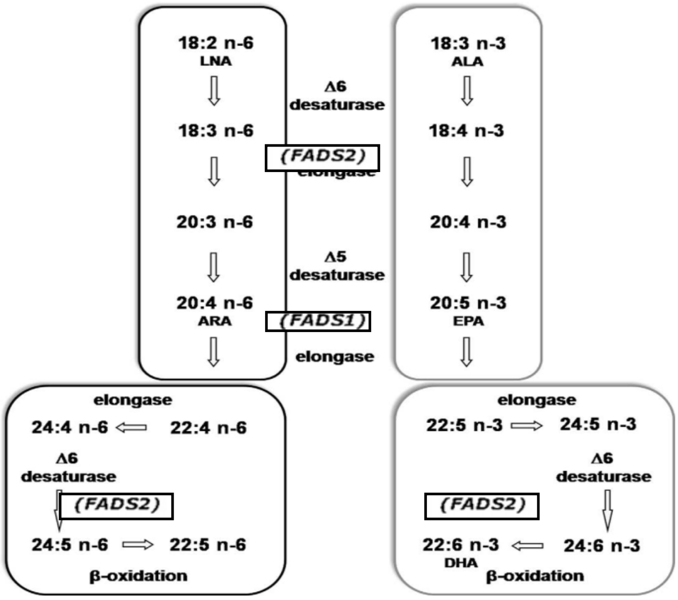


Figure 1.9. A mammalian pathway of the n-6 and n-3 fatty acid elongation and desaturation.

LNA = linoleic acid; ALA = α-linolenic acid; ARA = arachidonic acid; DHA = docosahexaenoic acid; EPA = eicosapentaenoic acid

(Adapted from: (Barceló-Coblijn and Murphy, 2009)&(Koletzko et al., 2011).

Inclusion of essential fatty acids in the complementary foods of young children has been recommended in the previous reviews of the efficacy and effectiveness of complementary feeding interventions in developing countries (Dewey and Adu-Afarwuah, 2008); and there has been growing interest in providing fats into the diet of young children in the form of lipid-based nutrient supplements (LNS) with special attention to essential fatty acids, improving energy and micronutrient intake(Arimond et al., 2015, Iannotti et al., 2014, Mangani et al., 2014) and promoting growth (Adu-Afarwuah et al., 2007, Phuka et al., 2008, Phuka et al., 2009) and development of young children. Moreover, the high content of omega 6 in the standard LNS has been a concern in the growing use of LNS in the form of ready to use therapeutic foods (RUTF) or ready to use supplementary foods (RUSF) and improving the n-3 LC PUFA content have been proposed to be feasible by use of a “high-oleic” RUTF instead ofLA in which a four weeks supplementation for Malawian children with severe acute malnutrition resulted in increased plasma level of EPA and DHAcompared to the starndard RUTF and recovery rate was comparable (Hsieh et al., 2015, Brenna et al., 2015); moreover, addition of fish oil capsule to a RUTF with elevated n-3 PUFA in comparison to standard RUTF or a PUFA with elevated short chain n-3 only resulted in increased erythrocyte long-chain n-3 PUFA among rural Kenyan children with severe acute malnutrition (Jones et al., 2015).

1.12.2. Essential fatty acids in the diet

There is a growing understanding over the last half of the century that the fatty acid consumption of our diet has changed in which there are more n-6 fatty acids in the diet as a result of consumption of refined vegetable oils (van Goudoever et al., 2011, Innis, 2011); which in turn increased the ratio of n-6 to n-3 up to 20-25:1(Teitelbaum and Allan Walker, 2001)and the farming of fish has led to decreased level of n-3 fat composition(Simopoulos, 2002). In situations where the source of essential fatty acids include sunflower, safflower, corn and peanut oils (which are very good source of LA), the conversation of ALA to EPA and DHA could be significantly reduced depending on the amount of consumption of these LA sources(Briend et al., 2011). Hence, the complete fatty acid profile of the diet needs to be known for a better understanding of the status of conversion of ALA to DHA. For infants who are breastfed, breast milk is the main source of DHA and EPA; however, DHA concentration of breast milk depends on the mother’s diet. It is also reported that there is a reduction in DHA content shortly after delivery and there is a need to consider supplementing the mother during lactation to promote normal visual and cognitive development (Campoy et al., 2012, Koletzko et al., 2008).

Supply and intakes of n-3 LC-PUFAs are far below the recommendation in children in most developing countries (Michaelsen et al., 2011, FAO, 2010 , Golden, 2009) and there is no sufficient evidence for the ratio of LA to ALA(FAO/WHO, 2008, Smit et al., 2004). Ideally, the ratio of n-6 to n-3 LC-PUFA should be kept low. In an area where sea foods are not widely consumed, the n-3 LC-PUFA composition of breast milk can be much lower than that of high-income countries and those in developing countries with access to sea foods(Michaelsen et al., 2011) and breast milk remains the main source of n-3 LC-PUFA for children in the first two years in these areas. Based on analysis of availability data of FAO food balance sheet, Ethiopia has one of the lowest figures in term of percentage of energy from fat (<20 E%), the lowest n-3 fatty acid supply (<0.4 E%) and n-6 to n-3 LC-PUFA ratio was also one of the highest in Africa (14) while the recommendation is between 5-15:1(Michaelsen et al., 2011) **(Figure 1.10).** Nearly 70% of the fat is also coming from cereals with very low PUFA values and has the lowest value of energy supply from n-3 fatty acids (Michaelsen et al., 2011). Cow milk, commonly consumed as additional drink starting early age in Ethiopia (CSA [Ethiopia] and ORC Macro, 2011) and has very low in LA but no DHA or EPA (Huffman et al., 2011 560).

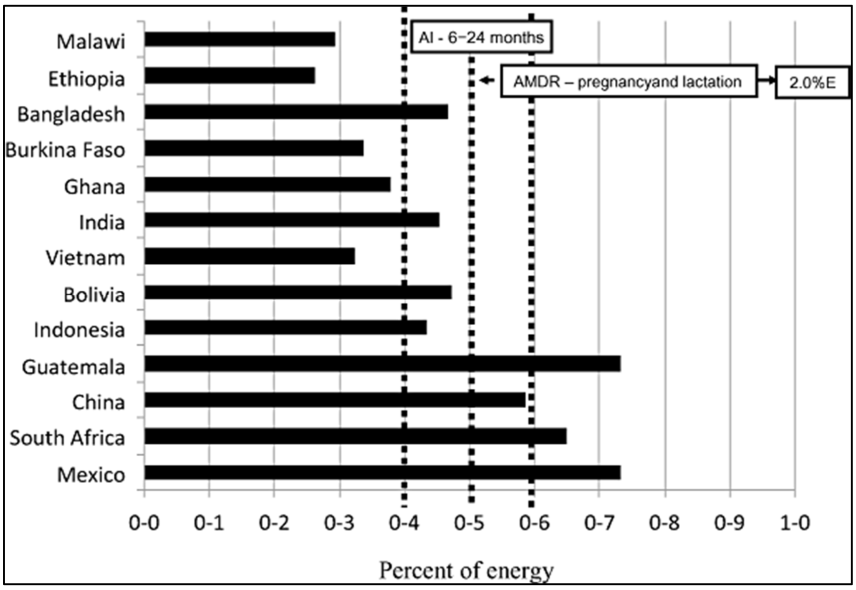


Figure 1.10: The n-3 fatty acid supply [percentage of energy supply (%E)] in 13 countries ranked according to gross domestic product. The AMDR for pregnant and lactating women and the AI for the 6–24 months age groupare shown (FAO/WHO 2008). AMDR, acceptable macronutrient distribution range; AI, adequate intake(Source:(Michaelsen et al., 2011))

In general, there is a low intake of n-3 fatty acids compared to AI in many developing countries (Huffman et al., 2011). Very good sources of ALA include flaxseed, soybean oil (Harris et al., 2009, Michaelsen et al., 2011). Tilapia which also commonly found fish in freshwaters in Africa is poor in n-3 PUFA (Harris et al., 2009, Racine and Deckelbaum, 2007) compared to other species such as mackerel, herring and salmon, and tuna.

1.12.3. Recommendations for essential fatty acid

Currently, there is no established dietary reference intake (DRI) for the intake of EPA and DHA for children and adults (Flock et al., 2013). Much of the current recommendations are derived from expert group reports and from different organizations and it is mostly established for the ALA and LA. The AI for n-3 and n-6 fatty acids recommended by the Food and Nutrition Board of Institute of Medicine (IOM) is commonly used (**Table 1.4**) and the requirements are based on the amount required for the prevention coronary heart diseases (CHD) (Trumbo et al., 2002). World Health Organization recommends acceptable macronutrient distribution range (AMDR) of 6-11%

and 0.5-2% of energy for intake of n-6 and n-3 fatty acids respectively based on secondary prevention of CHD (FAO/WHO, 2008).

Table 1.4: Intake requirements for n-6 and n-3 fatty acids for the different life stage groups

Life stage group	Age	AI for n-6 fatty acids (FA)			AI for n-3 fatty acids(FA)		
		Source	Males (g/day)	Females (g/day)	Source	Males (g/day)	Females
Infants	**0-6 months**	**LA***	**4.4**	**4.4**	**ALA¥**	**0.5**	**0.5**
Infants	**7-12**	**LA**	**4.6**	**4.6**	**ALA**	**0.5**	**0.5**
Children	1-3 years	LA	7	7	ALA	0.7	0.7
Pregnancy	all ages	LA	-	13	ALA	-	1.4
Breast-feeding	all ages	LA	-	13	ALA	-	1.3

***LA= linoleic acid;¥ALA= α-linolenic acid;**

AI= Adequate intake: "The recommended average daily intake level based on observed or experimentally determined approximations or estimates of nutrient intake by a group (or groups) of apparently healthy people that are assumed to be adequate – used when an EAR (and thus an RDA) cannot be determined" (Food and Nutrition Board (Institute of Medicine), 2002). **Source: adapted from (Trumbo et al., 2002)**

EPA and DHA intakes during the first two years should be the same as the breast milk content of these fatty acids. Based on analysis of breast milk samples from nine countries, the level of AA in breast milk is nearly constant (~0.35-0.50% of FA), but EPA (~0.07-0.26% of total FA) and DHA (~0.15-1.0% of total FA) levels vary significantly depending on the maternal diet(Yuhas et al., 2006).

1.12.4. Essential fatty acids and child growth

Essential fatty acids are crucial as structural components of the cell membrane and affect growth, brain development, and vision and serve as signaling molecules and synthesis of different bioactive substances(Huffman et al., 2011, Innis, 2009, Simopoulos, 2002, German, 2011). Several studies conducted in the West assessed growth parameters in relation to supplementation with LC-PUFAs (DHA, DHA/ARA) obtained from different sources (algae, egg triglycerides or fungal oils), and none of them showed benefit on physical growth of term infants who were followed up to three years of age(Makrides et al., 2005, Simmer, 2001, Simmer et al., 2008). In a relatively recent Cochrane review of interventions studies for which physical growth was measured, there was no beneficial effect on physical growth upon supplementation with the LC-PUFA from 3 months to 9 years (Simmer et al., 2011). However, further research with larger sample size, a higher dose of DHA, control for other factors and standardization of growth measurements wererecommended (Simmer et al., 2011). Another Cochrane review looked into the effect of maternal LC-PUFA supplementation on child growth and development. In the latter review, LC-PUFAs supplemented to breastfeeding mothers improved the LC-PUFA content thereby increasing intake by the infant. The review, however, did not find a difference in length, weight, and head circumference, fat mass, and fat distribution between the supplemented and placebo groups (Delgado-Noguera et al., 2015). Most of the reviewed studies are conducted in Europe and the United States and used infant formulas; hence, making the same conclusion about the situation in the low-income countries is not warranted (Makrides et al., 2011). Furthermore, the type of fatty acid and combinations(DHA or DHA and ARA) did not affect the outcome on weight, length, and head circumference (Makrides et al., 2005, Lauritzen et al., 2015, Wright et al., 2006).

The proposed beneficial effects of essential fatty acids supplementation in developing countries may be related to the already low level of fatty acid due to thecommon rate of undernutrition (Smit et al., 2004, Huffman et al., 2011) and additional supplementation could be beneficial in promoting child growth and development (Makrides et al., 2011). Moreover, the effects of n-3 LC-PUFAs have been found to be pronounced among malnourished children and children from low socioeconomic settings (Smit et al., 2004). A highly purified fish oil supplementation trial in The Gambia among children from 3-9 months found a significant difference in MUAC and a marginally significant difference in triceps-skin-fold thickness among the fish oil group (van der Merwe et al., 2013). However, there was no difference in linear growth between the two groups. A recent review also added that n-3 LC-PUFAs are involved in the metabolism of glucose, fats and amino acids for efficient use of proteins in early life (Innis et al., 2013) which could be an indication for supporting physical growth.

1.12.5. Essential fatty acids and immunity

There is a complex relationship between nutrition and infection in which undernutrition increases susceptibility to infection and the infection, in turn, reduces appetite, intestinal absorption, increased catabolism and nutrients are shunted towards improving immune response to the ongoing infection (Katona and Katona-Apte, 2008, Dewey and Mayers, 2011). Malnutrition also impairs the intestinal barrier and immune response leading to increased risk of infection, chronic inflammation (Dewey and Mayers, 2011). Children during the first two years are at special risk of diarrhea and respiratory infections (Dewey and Mayers, 2011) and the impact on nutritional status could be severe depending on the amount of exposure to diarrheal episodes in which the probability of stunting at 2 years of age increased by 2.5% per episode and 25% of all stunting was attributable to having five or more episodes in the first two years in a pooled analysis of nine studies (Checkley et al., 2008).

Despite some limitation in the assignment of study subjects and lack of control of some confounding factors, a study involving more than a thousand children in Spain demonstrated that a decrease in respiratory symptoms (either by decreased incidence of infection or inflammation) was observed by supplementing infants with DHA/ARA (Pastor et al., 2006). However, the later did not demonstrate a difference in anthropometric parameters. Reduced duration and episodes of illness were also reported among Thai school children with supplementation with fish oil (Thienprasert et al., 2009).

It has been long known that n-3 fatty acids are important in the amelioration of inflammatory diseases mediated by eicosanoids (Innis, 1991, Calder, 2006, Calder, 2008). In general, inflammation is characterized by the production of inflammatory cytokines, an arachidonicacid derivedeicosanoids, inflammatory mediators, and adhesion molecules. n-3 LC-PUFAs decrease production of inflammatory mediators and expression of adhesion molecules through either inhibition of arachidonic acid metabolism or altering expression of inflammatory genes(Teitelbaum and Allan Walker, 2001, Calder, 2002)**(Figure 1.11).** The ratio of n-6: n-3 LC-PUFA should be kept at 1:1 in ideal circumstance but due to increasing use of vegetable oils both in developed and developing countries, there is a tendency to have more pro-inflammatory metabolites (Teitelbaum and Allan Walker, 2001).

Some argue that the importance of the balance of n-6/n-3 LC-PUFA with regards to reducing the risk of cardiovascular diseases is not clear and making recommendations in the face of lack of sufficient evidence among humans is problematic (Poli and Visioli, 2015, Harris, 2007). The relationship between n-3 and n-6 LCPUFA is so complex that some AA derived eicosanoids have both pro- and anti-inflammatory roles or may be involved in resolving inflammation (Poli and Visioli, 2015, Ferrucci et al., 2006). All mediators of from AA are not pro-inflammatory and at the same time anti-inflammatory actions of n-3 LC PUFAs may not always relate to changes in synthesis of lipid mediators (Calder, 2009). However, the incorporation of EPA and DAH to the membrane cells could lead to anti-inflammatory action in the cells at the expense of AA which intern decreases pro-inflammatory substrate (Calder, 2013). Increased level of both fatty acids resulted in lowest level of plasma TNF receptors. Another observational study conducted in 1123 persons aged 20–98 year reported several significant associations between plasma concentrations of total n- 6 and n-3LC PUFAs and various pro anti-inflammatory markers. Higher level of n-3 LCPUFA was directly associated with anti-inflammatory markers or decreased level of pro-inflammatory markers (Ferrucci et al., 2006). Much of the studies in the relationship of fatty acids and inflammation are conducted in vitro and the extrapolation of in vitro studies to humans has been problematic.(Fritsche, 2015) and practical recommendations are difficult to make (Harris, 2007) There is no justification so far in reducing intake of n-6 LCPUFA at least with the aim of reducing the risk of cardiovascular diseases(Harris, 2007) despite the clear evidence that incorporation EPA and DHA into membrane cells reduces inflammation at the expense of AA. The balance of n-6 and n-3 LCPUFAs in relation to other inflammatory condition is not also known. Relatively recent the review by Simopoulos et al 2016, still reaffirms the previously reported reviews that increased level of n-6 PUFAs are not healthy. It was concluded that the balance of n-6 and n-3 fatty acids is important for optimal health (Simopoulos, 2016).

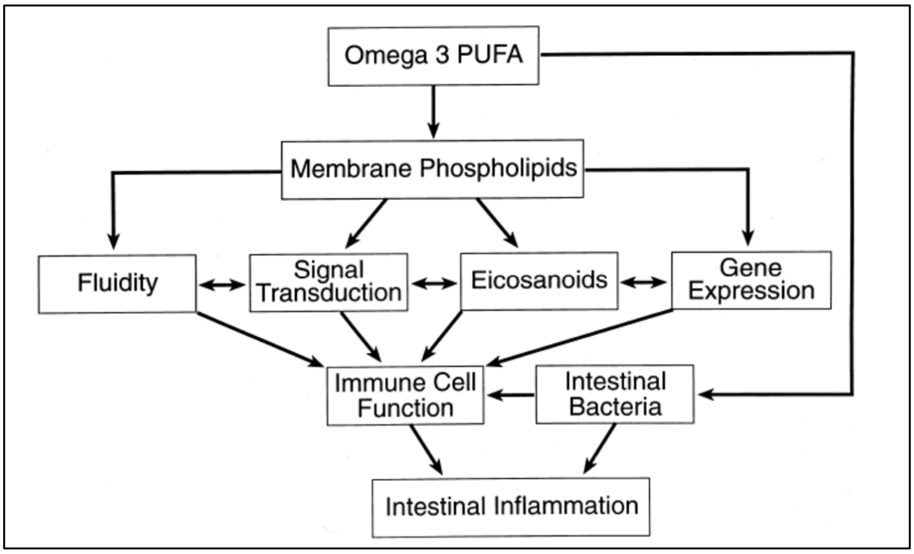


Figure 1.11: Mechanism by which v3 PUFAs might exert immunomodulatory and anti-inflammatory effects. Source (Teitelbaum and Allan Walker, 2001)

There has been an interest in the use of the anti-inflammatory effect of n-3 LC-PUFA to reduce gut inflammation (environmental enteropathy) and ameliorate the deleterious effect of EE. However, there have been very limited studies conducted using markers of intestinal inflammation as an outcome. The relatively recent trial in The Gambia demonstrated no effect of supplementation with highly purified fish oil on intestinal and systemic inflammation as measured lactulose: mannitol ratio and fecal calprotectin concentration respectively (van der Merwe et al., 2007).

1.13. Nutrition situation of children in Ethiopia

Despite recent advances in improving the health and nutritional status of children in Ethiopia, household food insecurity, hunger, and undernutrition remain critical issues (IFPRI/WHH/Concern Worldwide/UN, 2016). The country managed to decrease the prevalence of stunting by about 20% over the past 14 years; however, 40% and 19% of children under the age of five years were stunted and severely stunted respectively (CSA [Ethiopia] and ORC Macro, 2014). Stunting prevalence reduced by 1.4% points per year between 2001 and 2011 even it slowed to 1% point per year between 2011 and 2014(CSA [Ethiopia] and ORC Macro, 2011, CSA [Ethiopia] and ORC Macro, 2014). The rate of wasting showed a very small decrement in which it has fallen only by about 3 percentage points over the same period (**Figure 1.12**). Even if specific tests were not made by the EDHS surveys, micronutrient deficiencies among children are also widespread in which 44% of children age 6-59 months are anemic, with 21 percent mildly anemic, 20 percent moderately anemic, and 3 percent severely anemic (CSA [Ethiopia] and ORC Macro, 2011). The prevalence of anemia decreased by about 10% over the previous 6 years from 54% of children in 2005 to 44% of children in 2011.

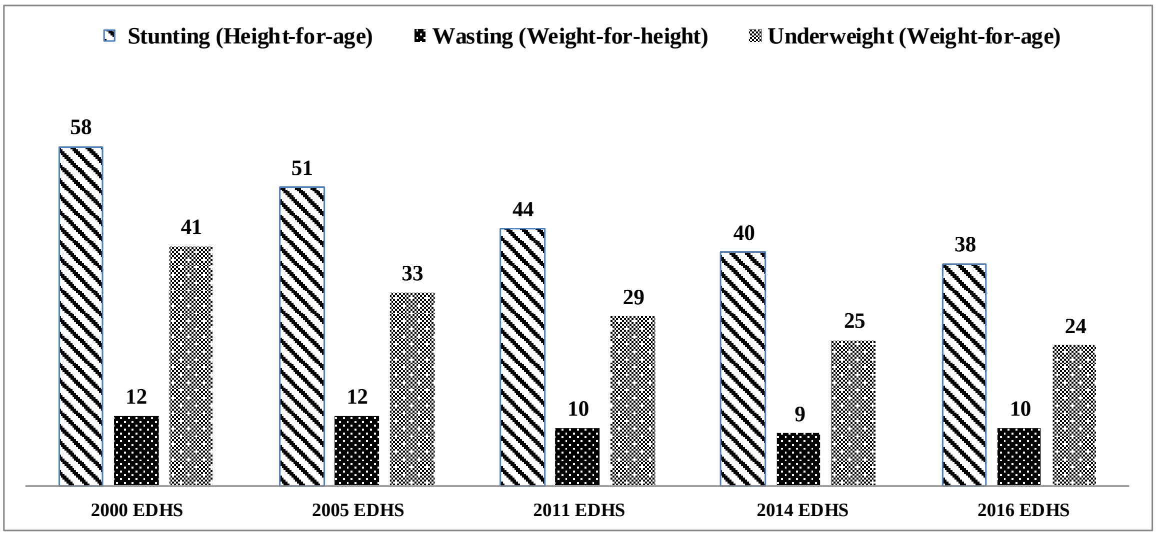


Figure 1.12 Trends in nutritional status of children in Ethiopia 2000-2016 (percent)

Adapted from: (CSA [Ethiopia] and ORC Macro, 2001); (CSA [Ethiopia] and ORC Macro, 2006); (CSA [Ethiopia] and ORC Macro, 2011); (CSA [Ethiopia] and ORC Macro, 2014); (CSA [Ethiopia] and ICF, 2016). EDHS= Ethiopia Demographic and Health Survey

Lack of dietary diversity and consumption of micronutrient dense food and suboptimal child feeding practices contribute to the high rates of child under-nutrition in Ethiopia. Only half of infants are exclusively breastfed and introduced complementary foods at the appropriate time, and only 4% of young children are receiving a minimum acceptable diet and just 5% of non-breastfed children are being fed in accordance with IYCF recommendations(CSA [Ethiopia] and ORC Macro, 2011). Like many developing countries, household food insecurity, inadequate health services, unsafe water supply and lack of sanitation facilities combined with deficiencies in the non-food factors of nutrition like maternal and child care practice are important determinants of undernutrition (IFPRI, 2005).

1.13.1. Practice of infant and young child feeding in Ethiopia

1.13.1.1. Initiation and maintenance of breast feeding

The practice of breastfeeding is universal in Ethiopia despite problems related to early initiation, exclusive feeding for the first six months and maintenance of breastfeeding for at least two years of age(CSA [Ethiopia] and ORC Macro, 2011, Disha et al., 2012, Disha et al., 2015). The rate of exclusive breastfeeding which was calculated based on feeding exclusively during the previous day, only 38% of Ethiopian children age 4-5 months(CSA [Ethiopia] and ORC Macro, 2001), 52% of children less than 6 months (CSA [Ethiopia] and ORC Macro, 2011) and ,58% during the first 6 months (CSA [Ethiopia] and ICF, 2016)were exclusively breastfed. The median duration of breastfeeding was 25.5, 25.8 and 25 months during the in 2000, 2005, and 2011 demographic and health surveys (CSA [Ethiopia] and ORC Macro, 2001, CSA [Ethiopia] and ORC Macro, 2006, CSA [Ethiopia] and ORC Macro, 2011).

1.13.1.2. Complementarity feeding practices

Despite the high rate of continued breastfeeding through the first year and beyond, complementary feeding practice has been problematic in Ethiopia (Headey, 2014). It was reported that only 4% of children age 6-23 months were fed according to the recommendations of infant and young child feeding (CSA [Ethiopia] and ORC Macro, 2011). About one in three children consumed some type of solid or semisolid food by 6-7 months in the 2000 EDHS report (CSA [Ethiopia] and ORC Macro, 2001), only one in two of children at 6-8 months of age were consuming solid or semisolid food during the 2005 national survey (CSA [Ethiopia] and ORC Macro, 2006)whereas about half of children received complementary foods by the age of 6-9 months in the 2011 national survey (CSA [Ethiopia] and ORC Macro, 2011, Disha et al., 2012).

The recommendations for minimum diet diversity, minimum acceptable diet, and consumption of iron-rich foods were observed in less than 10% of children aged 6-23 months (Disha et al., 2012). According to the 2016 EDHS report, only 7% of children age 6-23 months met the criteria for a minimum acceptable diet (CSA [Ethiopia] and ICF, 2016).

2

Chapter 2: Aims of the Researchand study setting

2.1. Overall aim of the research

The PhD research aimed at assessing the caregiver and child interaction during feeding, the role of infant and young child feeding indicators as measurement tools to assess the quality of complementary foods and growth of infants. It further necessitates to investigate the role of essential fatty acids provided through breast feeding and in complementary foods in the face of high rate of undernutrition (linear growth retardation) and low intake of n-3 long-chain poly unsaturated fatty acids in Ethiopia

Specific objectives of the research

1. To assess the type of caregiver-child interaction among children 6-23 months in southern region of Ethiopia
2. To determine the validity and seasonal stability of dietary diversity score as indicator of micronutrient density adequacy and its seasonal stability in two seasons among young infants in rural Ethiopia
3. To determine the association of child feeding practice indicators and nutritional status (growth) of infants and young children in two distinct seasons in rural Ethiopia
4. To evaluate the effect of n-3 LC-PUFA supplementation provided through breastmilk and supplementary food for 12 months on growth and morbidity of children 6-24 months in Rural Ethiopia.

The following research hypotheses were the basis for undertaking this Ph.D. study in Ethiopia

1. In the context of high rate of undernutrition during early ages, maternal or caregiver-child interactions during feeding are not proactive or responsive.
2. Information obtained from consumption of food groups over a period of time could be used as a tool to predict the density adequacy of micronutrient intake by infants
3. The summary indices of child feeding practices are associated with child growth or nutritional status and are stable over two distinct agricultural seasons
4. Providing children with preformed omega 3 fatty acids in the form of purified fish oil through maternal supplementation and incorporation into a processed supplementary food supports growth ,decreases morbidity and reduces systemic inflammation

2.2. Presentation of the study setting

2.2.1. Country profile

2.2.1.1. Geography

Ethiopia covers a total area of 1,104, 300 km2 and is located in the north-eastern part of Africa. It is a landlocked mountainous country bounded on the north-east by Eritrea and Djibouti, on the east and south-east by Somalia, on the south-west by Kenya and South Sudan and on the west and north-west by Sudan (FDRE (MOH) [Ethiopia], 2010). Ethiopia has great geographical diversity; its topographic features range from the highest peak at Ras Dashen, 4,550 meters above sea level, down to theAffarDepression, 110 meters below sea level (CSA [Ethiopia] and ORC Macro, 2011). The climate varies with the topography, from as high as 47 degrees Celsius in theAffarDepression to as low as 10 degrees Celsius in the highlands (CSA [Ethiopia] and ORC Macro, 2011).The rainy season occurs between mid-June and September, followed by a dry season occasionally interrupted in February or March by another short rainy season

2.2.1.2. Administrative structure

Ethiopia has nine Regional States: Tigray, Afar, Amhara, Benishangul-Gumuz, Gambella, Harari, Oromia, Somali, and Southern Nation Nationalities and Peoples Region (SNNPR); and the City Administration Councils of two cities: Dire Dawa and Addis Ababa (the capital). The regional states and city administrations are subdivided into 817 administrative Woredas (districts). The 817 Woredas are further divided into about 16,253 Kebeles (the smallest administrative unit). There are also two zones and seven Woredas designated as "special". These are medium sized towns or traditional sites of various population groups (CSA [Ethiopia] and ORC Macro, 2006). Jimma zone is one of the 12 administrative zones of Oromia Regional State where our study took place (**Figure 2.3**).

2.2.1.3. Demography

Ethiopia, which is the second most populated country in Sub-Saharan Africa, is experiencing rapid population growth. In 2016 the population has been estimated at 101.7 million (Population Reference Bureau, 2016). As of 2007, Ethiopia's population has been growing at a rate of 2.6 percent per annum (CSA [Ethiopia], 2008). The population is very young and is one of the least urbanized in the world. The majority of the population (84%) live in rural areas. The pyramidal age structure of the population has remained predominately young with 45% under the age of 15 years, and over half (52%) of the population in the age group of 15 and 65 years (CSA [Ethiopia] and ORC Macro, 2014) (**Figure 2.1**).

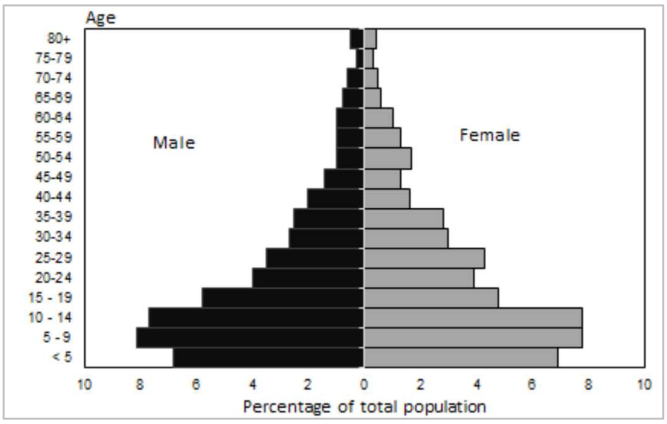


Figure 2.1. Population pyramid for Ethiopia (source:(CSA [Ethiopia] and ORC Macro, 2014))

2.2.1.4. Economy

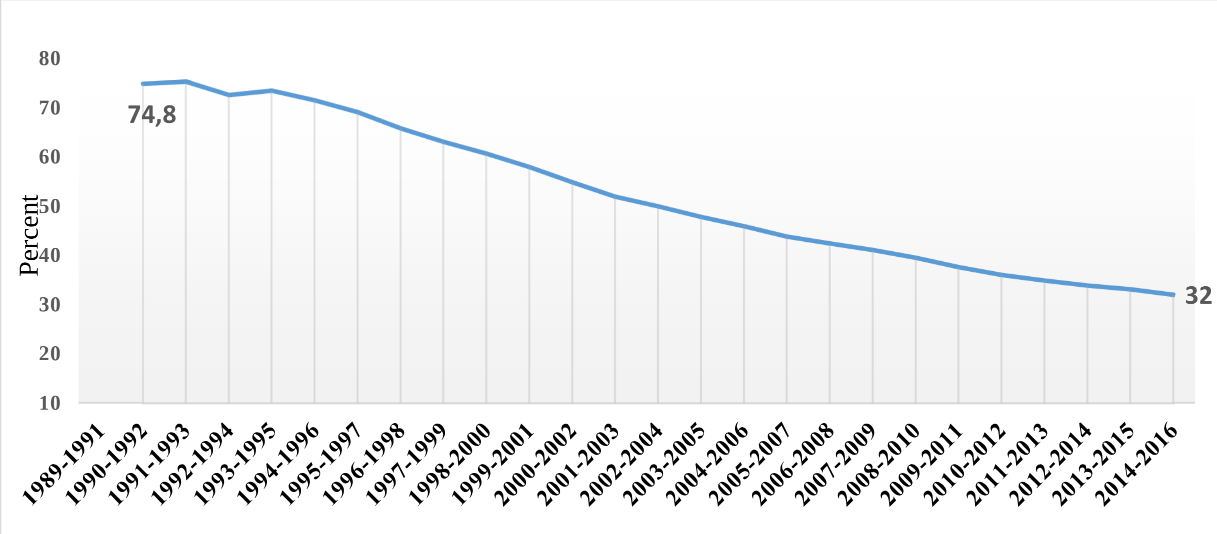
Ethiopia is an agrarian country and agriculture accounts for 42% of the gross domestic product or GDP (CSA [Ethiopia], 2016) and contributes to about 85%of the labor force and 90% of the total export earnings of the country. Coffee has long been one of the main export items of the country; however, other agricultural products are currently being introduced on the international market (CSA [Ethiopia] and ORC Macro, 2011). The sector is dominated by over 15 million smallholders producing about 95 percent of the national agricultural production. This shows that the overall economy of the country and the food security of the majority of the population depend on smallholder agriculture. Rain-fed agriculture remains the key source of livelihood for the majority of the population (CSA [Ethiopia], 2016). Regular droughts combined with poor cultivation practices, make Ethiopia's economy vulnerable to climatic changes. Fifty five percent of Ethiopians lived in extreme poverty in the year 2000, by 2011, 33.5% lived on less than $1.90 per day (FAO, 2016). The country's per capita income as measured by the Atlas Method was $590 which is substantially lower than the regional average in 2015(The World Bank, 2015)

2.2.1.5. Staple foods and consumption

The Ethiopian diet is mainly composed of cereals (maize, sorghum, and teff), tubers and root crops (ensete, potatoes, and sweet potatoes), pulses and oil seeds (FAO, 2008). Cereals take almost the majority of the total grain crops cultivated land and production for smallholder farmers in which it accounts for 87% of the production followed by pulses (10%) and oil seeds (3%) for the year 2015/16 (CSA [Ethiopia], 2016). Even if Ethiopia has one of the largest numbers of livestock, the productivity is known to be limited (FAO, 2008). Hence, consumption of meat and milk and dairy products are very low. As a result, undernourishment (it is a situation when a person is not able to acquire enough food to meet the daily minimum dietary energy requirements, over a period of one year) is very common even if showing a decline in the past decades(FAO, 2016) (**Figure 2.2**). Environmental and man-made factors cause widespread and severe food insecurity. The dietary energy supply is not sufficient to meet population energy requirements and almost half of the population is undernourished. Besides being quantitatively insufficient, food supplies also lack diversity (FAO, 2016).

When we look at consumption patterns of agricultural products, Ethiopian diet mainlycomposed of cereals, starchy roots and tubers, livestock products and oilseeds. However, cereals particular *teff* (Eragrostisabyssinica), maize, barley and sorghum, wheat, and millet are the main staples in most part of the country. Injera, the main traditional staple dish, is a large pancake made from fermented *teff* that is accompanied by a legume or meat based sauce or thin bread made of maize accompanied by vegetables, other sauces (FAO, 2008).

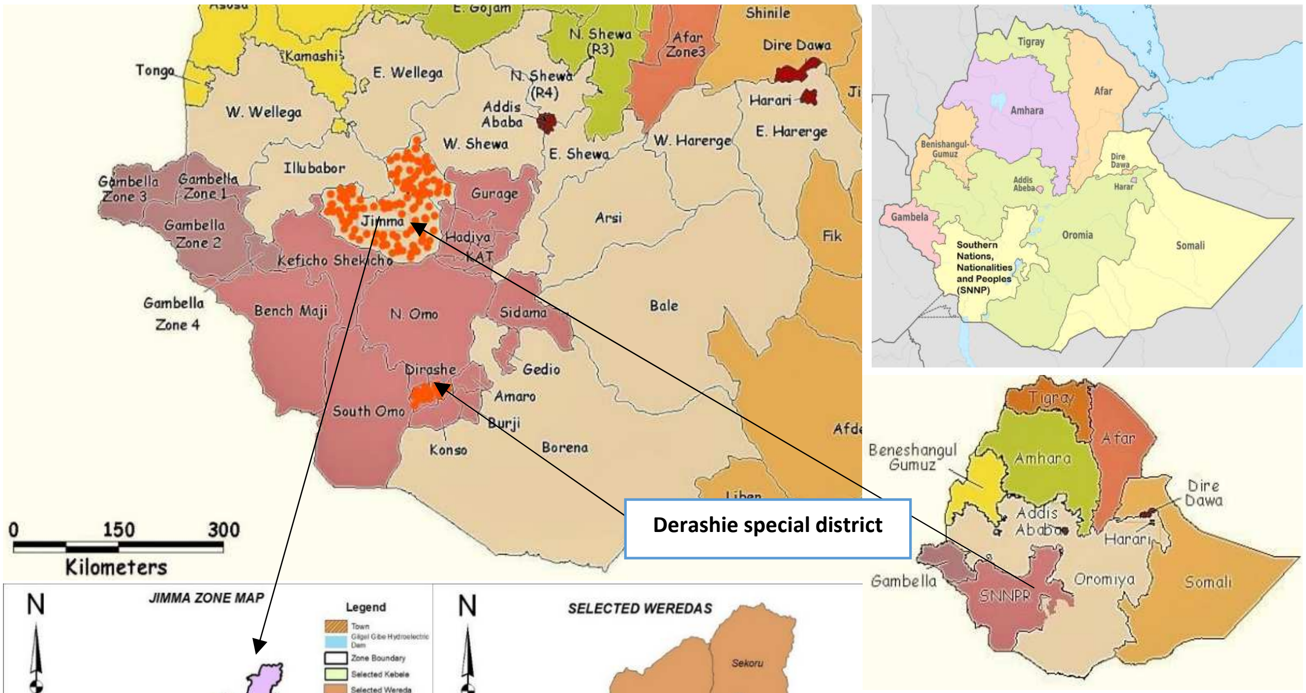
Figure 2.2. Prevalence of undernourishment (percent) (three years average)



Adapted from: FAO, 2016 (available at: http://www.fao.org/faostat/en/#country/238).
Undernourishment= refers to the proportion of the population whose dietary energy consumption is less than a pre-determined threshold. The calculation considers mean dietary energy supply as a proxy for food energy consumption

2.2.1.6. Child health and nutrition

Ethiopia continues to have one of the highest infant and young child mortality rates in the world despite significant improvements in this regard. Based on the Global Burden of Disease 2013 data, there has been a 64% reduction in the under-five mortality rate (from 204 deaths/1000 live births to 74 deaths/1000 live births) between 1990 and 2013(Deribew et al., 2016).In this analysis, lower respiratory tract infection, diarrheal diseases, and neonatal syndromes (preterm birth complications, neonatal encephalopathy, neonatal sepsis, and other neonatal disorders) accounted for 54% of the total under-5 deaths in 2013 followed by measles, whooping cough and malaria. The most important risk factors for diarrheal diseases, lower respiratory tract infection, and other common infections identified were childhood wasting, underweight, stunting, non-exclusive breastfeeding, and vitamin A deficiency(Deribew et al., 2016). The infant mortality rate has decreased in the last decade from 77 per 1000 live births in 2006 to 41 in 2015 and the under-five mortality rate also showed a decline from 123 per 1000 live births in 2006 to 59 in 2015 (UNICEF/WHO/The World Bank /UNDP, 2015).



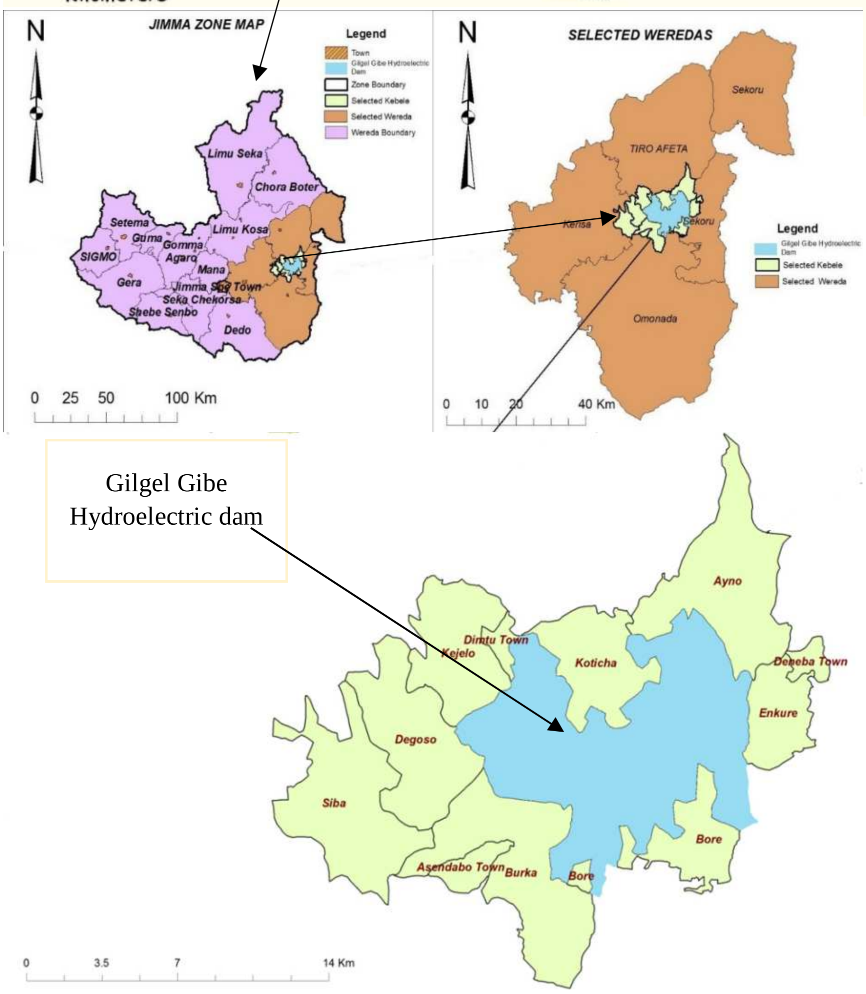


Figure 2.3: Map of the study sites.

Sources: Institute of Health Science Research (GGFRC), Jimma University, Ethiopia**Available at: http://www.indepth-network.org /Profiles/Gilgel%20HDSS.pdf**

2.3. Outline of the dissertation

The dissertation has four parts (**Figure 2.4**). The first part (**Chapter 1**) presents the overview of the global, regional and local situations regarding malnutrition, factors influencing nutritional outcomes such as caregiver-child interactions, infant and child feeding practices and the role of essential fatty acids in child growth, morbidity and inflammation. This is followed by brief description of the objectives and study settings. The second part include studies relating to pattern of caregiver-child interactions (**Chapter 3**), the role of simple indices of infant and young child feeding as indicators of complementary food quality **(Chapter 4**) and growth or nutritional status (**Chapter 5**) in pre-harvest and harvest seasons in rural Ethiopia. The third part investigates the role of n-3 LC-PUFAs provided through lactation and a processed supplementary food (corn soy blend plus one RDA of multiple micronutrients) in promoting growth and prevention of morbidity and systemic inflammation of children 6-24 months old **(Chapter 6).** Finally, a general discussion, conclusion and policy and research recommendations are provided (**Chapter 7**).

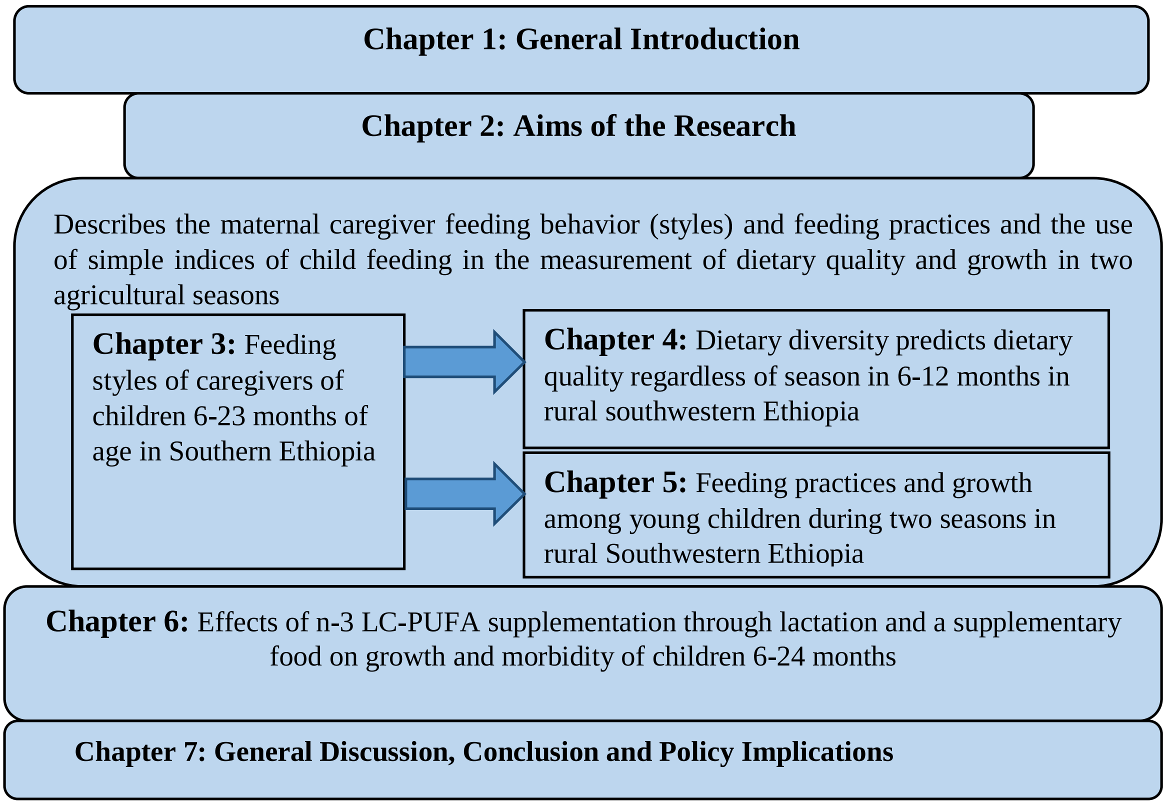


Figure 2.4. Schematic presentation of the Ph.D.dissertation

3

Chapter 3: Feeding styles of caregivers of children 6-23 months of age

Redrafted from: Wondafrash, Mekitie, Amsalu, Tseganeh, & Woldie, Mirkuzie. (2012). Feeding styles of caregivers of children 6-23 months of age in Derashe special district, Southern Ethiopia. *BMC Public Health, 12*(1), 1-8. doi: 10.1186/1471-2458-12-235

3.1. Abstract

Background: Apart from basic determinants, appropriate child care practices are important in prevention of growth faltering and undernutrition. Providing safe and appropriate quality complementary foods is crucial to child growth and development. However, some children in low-income communities grow normally mainly due to proper caregiver feeding behaviors. Hence, the objective of this study was to determine caregivers' feeding styles and toidentify predictors in Derashe special district, Southern Ethiopia.

Methods:A community based cross-sectional study design was employed in the seven randomly selected Kebeles (smallest administrative unit) of Derashe special district. A total of 826 caregivers provided data pertaining to socio-demographic variables. However, 764 caregivers had complete data for the outcome variable (caregiver feeding style). A multistage stratified sampling technique was used to identify study subjects. A Caregiver's Feeding Styles Questionnaire (CFSQ) was used to develop a behavior and belief item questionnaire to gather information pertaining to each feeding style adopted by caregivers. A multivariate multinomial logistic regression was employed to identify predictors of caregivers' feeding styles.

Results: The majority (80.6%) of caregivers were biological mothers. Seventy-six percent of the caregivers practiced a responsive feeding style. Caregivers other than the biological mother favoured a laissez-faire feeding style, while caregivers residing in rural Kebeles were more responsive. Caregivers with a breastfeeding frequency of more than eight times predicted both laissez-faire (RRR= 1.88; 95% CI= 1.03-3.41) and controlling (RRR= 1.70; 95% CI= 1.02-2.85) feeding styles as compared to responsive feeding.

Conclusion: Responsive feeding was the commonest feeding style adopted by the caregivers. Many of the caregivers who were rural residents and birth parents were responsive in child feeding. Instruments needed to be validated in the Ethiopian context and an additional prospective study based on direct observation of caregiver-child interactions is recommended.

3.2. Introduction

Malnutrition is the largest risk factor in the world for disability and premature mortality among young children, especially in developing countries (Muller and Krawinkel, 2005, FAO/WHO, 1992). Although the condition is entirely preventable, malnutrition is a significant underlying factor in more than half of the deaths of young children in these countries (Muller and Krawinkel, 2005, Pelletier, 1994). Malnutrition is a background factor for deaths from diarrhea, measles, acute respiratory infection, meningitis and malaria (Pelletier et al., 1995). In Ethiopia, 47%, 11% and 38% of children under five years of age are stunted, wasted and underweight, respectively (CSA [Ethiopia] and ORC Macro, 2006). An Ethiopian child is 30 times more likely to die by his or her fifth birthday than a child in Western Europe and the most common cause of child death is the interacting combination of malnutrition and infection (Joan and Mesfin, 2008).

Malnutrition is a complex phenomenon that stems from various underlying determinants, including a lack of optimal feeding practices for infants and young children. In UNICEF's conceptual framework for determinants of nutritional status, maternal and child care practices have been given due attention in addition to sufficient food supply at the household level, access to health services and a clean environment (UNICEF, 1997). Some children in low-income countries with high rates of malnutrition grow normally due to better educationand household management, or coping skills of their mothers (Pelto, 2000). Moreover, proper feeding practices, which ensure intake, are as important as the provision of complementary foods that meet nutritional requirements.

The caregiver-child interaction (feeding style) critically influences dietary intake on top of the dietary aspect of child feeding (Ruel et al., 2003). However, comparing feeding styles between studies prior to 1995 was difficult due to a lack of consistency in terminology and unclear definitions of feeding styles (Dettwyler and Fishman, 1992). Later on, feeding styles were conceptualized and defined by Birch and Fisher as controlling, laissez-faire, and responsive feeding styles (Birch and Fisher, 1995), and have been used in different researches (Ha et al., 2002, Ruel et al., 2003, Eshel et al., 2006).

The “controlling” feeding style occurs when a caregiver has complete control of when, what and how much the child eats, and includes restriction and force-feeding the child. In the “laissez-faire” feeding style the caregiver makes little effort to encourage eating; feeding is not encouraged even when the child may be marginally nourished (Dettwyler, 1989). Birch and Fisher defined the term “responsive” feeding as a condition in which “the caregiver is in close proximity to the young child during the meal and responds to the child’s hunger cues in a reasonable time” (Birch and Fisher, 1995). Responsiveness is now viewed by different authors as a three-step process, namely observation of child cues by the caregiver, interpretation of the signs and taking action as quickly as possible in response to those cues (Eshel et al., 2006, Bentley et al., 2011, Pelto, 2000).

Appropriate child feeding practices and behaviors of parents have a positive effect on growth of infants and young children (Saha et al., 2008). For instance, an analysis of data sets from several Latin American countries demonstrated that appropriate breastfeeding and complementary feeding practices were positively associated with child height-for-age in most of the countries studied (Ruel and Menon, 2002). However, the majority of the literature on child feeding and parental practices are based on women in affluent societies and their emphases have been on child overweight or obesity (Crouch et al., 2007, May et al., 2007 Jul;, Hurley et al., 2011). Control over feeding appears to vary among cultures, socioeconomic status and child’s gender (Hupkens et al., 1998). Hence, results of the effect of different parental or caregiver feeding behaviors among various socio-cultural settings should be used cautiously.

Most studies carried out in Ethiopia have focused mainly on identifying factors associated with undernutrition instead of overweight or obesity (CSA [Ethiopia] and ORC Macro, 2006, Girma and Timotiows, 2002). To the best of our knowledge, no studies have been conducted to assess child feeding behavior of caregivers and the association between these behaviors and child growth and development, dietary intake, eating behavior or morbidity in Ethiopia. Current interventions mainly address the issue of what to feed young children instead of how to feed them (Pelto, 2000). The aim of this study was to identify the different types of child feeding behaviors (styles) of caregivers and the factors that influence these behaviors.

3.3. Methods and Subjects

Study area and setting

A community based cross-sectional study was conducted in January 2009 in Derashe special district, Southern Ethiopia. Based on the 2007 census, the projected total population of the district in 2009 was 149,901 and the total household count was 29,980. Children 6-23 months of age constituted 6.7% of the total population. The livelihood of more than 92% of the district population is based on farming. The main crops grown in the area are maize, sorghum and teff (a species of*Eragrostis*native to Ethiopia).

Sampling

The source population of this study included all residents of Derashe district who were caregivers of children aged 6-23 months and resided in the district for more than 6 months.The sample size was calculated by using the single population proportion formula. Assumptions for the calculation were that 50% of caregivers practiced a "*responsive*" feeding style (P) with a 95% confidence level and 5% tolerable error. The calculated sample size was 384. As a multistage sampling technique was employed to identify study subjects, a design effect of 2 was used to reach a sample size of 768 to which 10% was added for non-responses. Thus, the final sample size was 845.

A multistage stratified sampling technique was used to identify study subjects after the Kebeles (smallest administrative units) were stratified into urban and rural. Based on the proportion of population by urban-rural residence (FDRE-PCC, 2008)seven rural and one urban Kebele were randomly selected in the district. A census was conducted in the selected Kebeles to identify households with children aged 6-23 months and to generate a sampling frame. A total of 3021 children in this age group who were started with complementary food were identified and listed. The calculated sample (845) was proportionally allocated to the selected Kebeles based on the total number of caregivers of 6-23 months children in each Kebele and study subjects were identified by simple random sampling technique.

Method of data collection

Data was collected by a face-to-face interview technique using a questionnaire adaptedfrom the Caregiver's Feeding Styles Questionnaire (Hughes et al., 2005, Patrick et al., 2005). The CFSQ was not as such directly taken and adapted through a standard cross cultural adaptation of an instruement. Rather we developed the questionnaire by taking behavior and belief items pertaining to the feeding styles (responsive, controlling, and laissez-faire)which might be adopted by the caregivers. The questionnaire was first prepared in the original language (English) and later translated into Amharic (the language used by all ethnic groups in the area) and back translated into English to check for its conceptual equivalence. Prior to the actual data collection, our instrument was pretested on caregivers from a similar population but outside the study area (5% of the sample size calculated) to check for the understandability and cultural sensitivity of the questionsadapted from the original instrument. Adjustments were made on the questionnaire based on the results of the pretest**.** Additional caregiver sociodemographic, child related andbreast feeding and complementary feeding practice questions were administered with the feeding style questions.

Twelve high school completedata collectors and two health officers, with previous experience in nutrition surveys, were recruited and trained by the principal investigators on the study instrument and data collection procedures. Reliability of the measurements was assured by training two health officers from the district for close follow-up. The principal investigators constantly checked strict adherence to the standards agreed upon by all field workers.

Measurement of caregivers' feeding styles and feeding practices

Data collectors asked the caregivers to respond to behavioral and belief items pertaining to each feeding style. However, many caregivers displayed behaviors and beliefs of more than one feeding style which made strict categorization into one of the three styles (controlling, laissez- faire or responsive) according to Birch and Fisher (Birch and Fisher, 1995) difficult. Hence, those caregivers adopting a particular style "most of the time" for the behavioral items, or "agreeing or highly agreeing"for the belief items, were considered as practicing responsive, controlling or laissez-faire feeding styles depending on the response to each behavior and belief items. The caregivers were asked to respond to all the questions regarding the three feeding styles and those who responded all the questions pertaining to one of the feeding styles according to the above operational definition were categorized as such. It is assumed that a caregiver might adopt more than one feeding style in different circumstances.

Data analysis

The data was analyzed using Stata version 11 (StataCorp, College Station, TX, USA). Nineteen caregivers were not found despite repeated visits. Sixty two questionnaires with inadequate information to measure caregiver feeding style were excluded making the total observation with complete information 764. Bivariate and multivariate multinomial logistic regression was employed to assess statistical associations between caregivers' feeding style and caregiver and child characteristics. Residence, age of caregiver and the study child, relation of the study child with the caregiver, educational status of the caregiver and the father of the study child, parity of the biological mother, birth interval between the study child and older sibling, frequency of breastfeeding and advice on child feeding by a health professional were entered as independent variables in the model. Responsive feeding style was used as a reference category in the model.

Variables which showed a significant association with caregivers' feeding style in the bivariate analysis, were entered into the multivariate multinomial logistic regression. Relative Risk Ratios (RRR), the relative risk ratios for the multinomial logistic model, were determined to assess whether the risk of the outcome falling in the comparison group (either controlling or laissez- faire feeding style) compared to the risk of the outcome falling in the reference group (responsive feeding style) increases or decreases with the independent variable. The p-value (P>|z|), is the probability that the z test statistic would be as extreme as, or more so, than what has been observed under the null hypothesis. If P>|z| is less than 0.05, the null hypothesis can be rejected and the RRR is considered significant at the 5% level.

Ethical considerations

Ethical approval was obtained from the Ethical Review Committee of Jimma University. Informed consent was secured from all caregivers of study children before commencing the interview.

3.4. Results

General characteristics of caregivers and study children

Of the 845 sampled caregivers, 826 were successfully included in the study making the response rate 97.7%. Biological mothers accounted for 666 (80.6%) of caregivers, while 160 (19.4%) were other caregivers such as grandmothers, sisters and other relatives. Most of the caregivers (87.5%) were rural residents. Fifty percent of the caregivers were 25-34 years of age. Seventy percent of the caregivers did not attend any formal education and 175 (71.7%) of those who attended school attended only elementary school. One hundred eighteen (17.7%) of the biological mothershad given birth to only one child. Five hundred and sixty five (78.9%) received advice from health professionals about child feeding (**Table 3.1**).

Table 3.1 Characteristics of caregivers of study children in Derashe special district, southern Ethiopia

Variables	Frequency	Percent
Residence of the caregiver (n=826)		
Urban	103	12.5
Rural	723	87.5
Age of the caregiver (years) (n=826)		
<20	87	10.5
20-24	145	17.6
25-29	260	31.5
30-34	156	18.9
>=35	178	21.5
Relation of the study child with the caregiver (n=826)		
Mother	666	80.6
Others*	160	19.4
Educational status of the caregiver (n=826)		
No formal education	582	70.5
Elementary	175	21.2
Above elementary	69	8.4
Educational status of the father of study child (n=826)		
No formal education	376	45.5
Elementary	251	30.4
Above elementary	199	24.1
Parity of the biological mother (n=666)		
Primiparous	118	17.7
Multiparous	341	51.2
Grand multiparous	207	31.1
Advice on young child feeding by a health professional (n= 716)		
Advised	565	78.9
Not advised	151	21.1

*Grandmothers, sisters, other relatives

The majority of interviewed caregivers (61.6%) had 12-23 month old children (**Table 3.2**). Biological mothers were the main caregivers of most of the 6-11 month old children (87%), while grandmothers were caregivers of 20 (6.4%) of these children. Mothers and grandmothers were the main caregivers of 396 (76.5%) and 64 (12.5%) of the 12-23 month old children, respectively (*data not shown*).

Table 3.2 Characteristics of the study children in Derashe special district, southern Ethiopia

Variables	Frequency	Percent
Age of the study child (n=826)		
6-8 months	188	22.8
9-11 months	129	15.6
12-23 months	509	61.6
Sex (n=826)		
Male	417.0	50.5
Female	409.0	49.5
Birth interval between the study and older sibling (n=637)[a]		
Short birth interval	422	66.2
Optimal birth interval	215	33.8
Frequency breast feeding in the last 24 hours (n=666)		
Less than recommended	432	64.9
Recommended	234	35.1

[a]*Optimal birth interval defined in this study as an interval between consecutive births of ≥ 36 months(WHO, 2007a).*

Breastfeeding and complementary feeding practices of caregivers

Six hundred sixty three (80.2%) of the caregivers were breastfeeding at the time of the study. Among the caregivers who specified breastfeeding frequency, 64.9% were feeding their children less than eight times per day. Two hundred ninety three (38.7%) of the interviewed caregivers had started giving complementary foods or drinks other than breast milk (mostly water, gruel and other semisolid cereal-based preparations) to their infants before they reached 6 months of age. For half of the caregivers, the reason for introducing foods/drinks too early was the belief that breast milk alone is not sufficient at that age. The mean (±SD) complementary feeding frequency was 4.6±2.0, 5.2±1.8 and5.2±2.0 times per day for study children 6-8, 9-11 and 12-24 months of age, respectively. Thirty eight percent of the study children ate from a separate bowl (data not shown).

Feeding styles of caregivers

The majority (75.7%) of caregivers fed their children mostly with a “responsive” feeding style, followed by “controlling” and “laissez-faire” in 98 (12.8%) and 88 (11.5%) of the caregivers, respectively(**Table 3.3**). On the other hand, 660 (80.0%) and 670 (81.2%) of the caregivers replied that the study child refused to eat if he/she was not hungry and when he/she was ill, respectively. Seventy seven (9.3%), 52 (6.4%) and 7 (0.9%) of the caregivers replied that the study child refused to eat if he/she was forced to eat, if someone other than the caregiver was feeding him/her and if he/she was not encouraged to eat, respectively (data not shown).

In response to food refusal by the child, 512 (66.9%) of the caregivers encouraged their children to eat either verbally or physically. According to the responses of 405 (53.0%) of the caregivers, the children ate more when encouraged verbally, while 292 (38.2%) of the respondents replied that the children ate more when food was provided on a separate plate (**Table3.3**). Five hundred eighty (70.3%) of the caregivers fed their sick child by verbal or physical encouragement. However, 182 (22.0%) of the caregivers force-fed their children during illness while 71 (8.7%) used other options to help their children eat under similar circumstances.

Table 3.3: Caregivers' feeding styles, responses to child's refusal to eat and conditions in which child eats more, Derashe special district, southern Ethiopia

Variables	Frequency	Percent
Caregivers' feeding style (n=764)		
Laissez-faire	88	11.5
Controlling	98	12.8
Responsive	578	75.7
Active responses for food refusal*		
Encourage him/her to eat verbally or physically	512	66.9
Offers the same food later	239	31.2
Offers alternative foods (favorite foods)	141	18.4
Forces the child to feed	84	11.1
Do not try any solutions	41	5.4
Condition in which the child eats more*		
When encouraged to eat	405	53.0
When food is presented in a separate plate	292	38.2
When the caregiver sits with the child while the child is eating	221	27.0
Forcing the child to eat	47	4.6

***** *There were multiple responses by a caregiver*

Predictors of caregiver feeding styles

Laissez-faire relative to responsive feeding style: the laissez-faire style is characterized by the lack of extra effort by the caregiver in making the child eat even if the child is at risk of undernutrition. Caregivers from rural villages were less likely (RRR= 0.16; 95% CI= 0.08-0.33) to practice a laissez-faire feeding style compared to a responsive feeding style given that the other variables in the model are kept constant. Caregivers other than biological mothers (RRR= 2.5; 95% CI= 1.43-4.38), who breastfed more than eight times during the previous day (RRR= 1.88; 95 % CI= 1.03-3.41) and biological mothers with an optimal birth interval between study child and older sibling (RRR= 2.46; 95 % CI= 1.38-4.40) were more likelyto practice a laissez-faire feeding style compared to a responsive feeding style (**Table 3.4**).

Table 3.4: Multinomial logistic regression of predictors of feeding style of caregivers, Derashe special district, Southern Ethiopia

Caregivers' feeding style	RRR *	P>|z|	[95% Conf. Interval]	
Laissez-faire				
Rural residence by the caregiver	0.16	0.000	0.08	0.33
Caregiver other than a biological mother	2.50	0.001	1.43	4.38
Frequency of BF more than eight times in the previous 24 hours	1.88	0.038	1.03	3.41
Optimal birth interval between the study child and older sibling (36-60 months)	2.46	0.002	1.38	4.40
Controlling				
Caregiver other than a biological mother	1.86	0.02	1.10	3.15
Frequency of BF more than eight times in the previous 24 hours	1.70	0.043	1.02	2.85
Optimal birth interval between the study child and older sibling (36-60 months)	0.54	0.038	0.30	0.97

***RRR=** Relative Risk Ratio; n= 764; The reference category is **"Responsive feeding style"**

Controlling relative to responsive feeding style: caregivers with a controlling feeding style try to control the time, type and the amount of food the child eats. They may also practice force-feeding when the child refuses to eat. Caregivers who were not biological mothers, compared to biological mothers, were more likely (RRR=1.86; 95% CI=1.1-3.15) to practice a controlling feeding style compared to responsive feeding. Caregivers who breastfed more than eight times during the previous day compared to those feeding less frequently were more likely (RRR= 1.7; 95% CI= 1.02-2.85) to practice a controlling feeding style. Moreover, it was also found that biological mothers with an optimal birth interval between study child and older sibling were less likely (RRR= 0.55; 95% CI= 0.32-0.95) to practice a controlling feeding style compared to a responsive feeding style **(Table 3.4)**.

3.5. Discussion

This study examined three types of caregivers' feeding styles (laissez-fair, controlling, responsive) from a socio-demographic, child feeding and caregivers' behavior perspectives. The majority of the studies investigating these three child feeding styles were carried out in developed countries and used a variety of techniques and instruments. This made direct comparisons of the findings from this study with other findings difficult.

In this study, 51% of the children were exclusively breastfed at 6 months of age. This is comparable to what was reported in the EDHS 2005 report (CSA [Ethiopia] and ORC Macro, 2006). The latter report showed that only one in three children of 4-5months of age were exclusively breastfed and the rate of exclusive breastfeeding for those less than six months was 49% (CSA [Ethiopia] and ORC Macro, 2006). In this study, caregivers were asked if they exclusively breastfed in the first six months whereas, the previous day breast feeding status wasassessed in the case of the EDHS report. Long term recalls tend to overestimate exclusive breast feeding rate; while, assessment of current status of breast feeding may not accurately represent the feeding pattern since birth (Bland et al., 2003).

The WHO recommends children of 6-8, 9-11 and 12-23 months of age to be fed with complementary foods 2-3, 3-4 and 3-4 times per day, respectively, with the addition of 1-2 snacks (PAHO/WHO 2003). In this study, 95% of the children within each of these respective age groups received complementary foods as frequent or more than the WHO recommendation. The mean (±SD) complementary feeding frequency per day was 4.6±2.0, 5.2±1.8 and5.2±2.0 times for children in the respective age groups.Considering the high rate of breast feeding (80%) and more than a third of the index children are breastfed for more than eight times per day, the frequency of feeding of complementary food could be more than recommened for this age. However, the quality of breast feeding, and the diversity and amount of complementary food consumed at each episode was not determined. The majority of caregivers were classified as practicing a responsive child feeding style (75.7%) which is consistent with the findings from a study in Vietnam (Ha et al., 2002).

In this study, 80.6% of caregivers were biological mothers. A study conducted in Nicaragua showed that mothers, as caregivers, were more likely than other caregivers to offer encouragement for eating (Engle and Zeitlin, 1996). Most of the caregivers in this study were rural dwellers but practiced a responsive feeding style most of the time. Two thirds of rural mothers in Bangladesh practiced active feeding ("behavior that encourages the child to eat or the mother to feed, either indirectly through words or directly through forcing food into the child's mouth") but only a third of them were responsive (Moore et al., 2006).

Caregivers' behavior in child feeding are part of the broader socio-cultural context of societies in different geographical locations (Engle et al., 2000). Several studies suggested that appropriate child feeding practices have an impact on child nutritional status (Galler et al., 1998, Saha et al., 2008, Ruel and Menon, 2002, Bentley et al., 2011, Hurley et al., 2011, Dettwyler and Fishman, 1992). Responsive or active feeding was found to be more beneficial to the child in some low- or middle-income countries (Engle and Zeitlin, 1996, Aboud et al., 2008, Bentley et al., 2011, Nti and Lartey, 2007) and non-responsive feeding was associated with child BMI and overweight or obesity in high-income countries (Hurley et al., 2011) though, cross-study definitions of responsive feeding were inconsistent. Responsiveness was also observed in reaction to a child's decreased appetite during diarrheal episodes in Guatemala (Bentley et al., 1992) and Peru (Bentley et al., 1991); and low interest in food in Nicaragua (Engle and Zeitlin, 1996). However, recent reviews concluded that the effect of responsive feeding on child weight or growth was mixed and failed to isolate the effect of responsive feeding, as most of these studies were done together with broader health or nutrition education and food supplementation programs (Bentley et al., 2011) and used cross-sectional designs (Hurley et al., 2011). Moreover, the mechanism of linking feeding behavior and child weight or growth differs between low- and middle-income and high-income countries (Bentley et al., 2011, Hurley et al., 2011).

In developing countries, there is very little information on the practice of controlling or laissez-faire feeding and implications for child nutrition. In Ghana, it was found that using force during feeding was common among caregivers with negative deviance in child feeding and had a direct effect on child's nutrient intake (Nti and Lartey, 2007). In

Nigeria, force-feeding and pressurizing was followed by food refusal by the child (Brown et al., 1988). In Vietnam, non-responsive feeding, characterized by pressure or force-feeding, was associated with the child's rejection of the food (Ha et al., 2002). A recent study in Malawi revealed that controlling feeding behavior was negatively associated with the acceptance of food by the child (Flax et al., 2011).

Unlike the present study, a laissez-faire feeding style was the most frequently observed behavior among caregivers in Peru and Guatemala (Bentley et al., 1991, Bentley et al., 1992). Encouraging feeding in these circumstances was a response only to a decrease in child food intake because of illness and hence was not pro-active.

This study did not assess the outcome of a particular feeding style practiced by caregivers, though the available literature on feeding styles underlined the association of feeding style with child growth and development, acceptance of food or child eating behavior and overweight or obesity (Sacco et al., 2007, Hoerr et al., 2009, Birch and Fisher, 2000, Aboud and Alemu, 1995, Dettwyler, 1989, Galler et al., 1998). As a limitation of the cross-sectional study design, temporality could not be established. Hence, it is difficult to determine whether a positive child response to the provided food leads to responsive feeding by caregivers or vice versa (Bentley et al., 2011). Nevertheless, behavior at any point in time could reflect prior caregiver-child interactions. The results of this study are also dependent on the reporting of the caregiver. Most studies used additional observations through videotaping of caregivers feeding behaviors which can increase the validity of the finding from interview-based studies(Sacco et al., 2007).

Conclusions and recommendations

Responsive feeding was the commonest style adopted by the caregivers. In this study, rural residence and being a biological mother were found to be independent predictors of responsive feeding after controlling for potential confounders. As this study was the first of its kind in the country, more research is needed to check the validity and reliability of the Caregiver's Feeding Styles Questionnaire in Ethiopia and to evaluate the effect of caregivers' feeding style on child dietary intake and nutritional status in different socio-cultural settings within the country. Moreover, longitudinal research starting from early infancy is recommended to see the factors affecting caregiver's child feeding behavior and other care practices.

4

Chapter 4: Dietary diversity predicts dietary quality regardless of season in 6-12 months old infants in South-west Ethiopia

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4.1. Abstract

Objective: Simple, cost-effective and convenient instruments like food group based scores are proposed to assess micronutrient adequacy of children in developing countries. We assessed the predictive ability and seasonal stability of dietary diversity score (DDS) to indicate dietary quality of infants.

Design: A 24-hr dietary recall assessment was carried out on a sample of 320 and 312 breastfed infants 6-12 months of age during harvest (HS) and pre-harvest(PHS) seasons respectively in Ethiopia. DDS was calculated based on seven food groups, while mean of micronutrient density adequacy(MMDA) was calculated for eight micronutrients. Multiple linear regression models were used to assess the relationship between DDS and MMDA, and differences in nutrient intake between the two seasons. A receiver operating characteristic analysis was performed to derive DDS cut-offs that maximize sensitivity and specificity of assessing dietary quality.

Setting: The study was conducted in the catchment of the Gilgel Gibe Field Research Centre of Jimma University, southwest Ethiopia.

Results: The mean (±SD) DDS for HS and PHS was 2.1(±0.94) and 2.3(±1.1) respectively. The DDS was associated with MMDA(β= 0.045, P<0.0001 in HS; β=0.044, P<0001 in PHS). A DDS of ≤2 food groups best predicted "low" MMDA(<50%) with 84% and 92% sensitivity, 36% and 43% specificity and 47% and 51% correct classification for the HS and PHS respectively.

Conclusions: DDS is predictive of dietary quality of breastfed infants. This study supports the use of DDS to indicate inadequate intake of micronutrients by breastfed infants in different seasons.

Keywords: Ethiopia, dietary diversity, micronutrient density adequacy, breastfed infants

4.2. Introduction

Ethiopia has one of the highest rates of child malnutrition (CSA [Ethiopia] and ORC Macro, 2011). The nutritional status is influenced by many interrelated factors (Black et al., 2008). Among others, calcium, zinc, iron, vitamin A and iodine deficiencies are common in Ethiopia, and have been attributed to low intake and low bioavailability (Andrew Sunil Rajkumar et al., 2012, Hotz and Brown, 2004, CSA [Ethiopia] and ORC Macro, 2011, Gibson et al., 2009). Moreover, the monotonous consumption of staple foods in complementary feeding fails to meet the daily requirements of infants and young children (Abebe et al., 2007, Gibson et al., 1998).

The assessment of nutritional status has been limited to anthropometry due to the time and personnel consuming nature of dietary assessments (Gibson, 1990). As a result, an intake assessment of nutrients, single food items, or a combination of foods to predict the risk of one or more health related problems has emerged as an alternative approach to actual quantification of nutrient intakes(Kourlaba and Panagiotakos, 2009). However, the problem of diagnostic ability of several dietary indices to predict health outcomes, dietary quality, or nutritional status has not been resolved for some reasons(Kourlaba and Panagiotakos, 2009).

The dietary diversity score (DDS) is a simple indicator of dietary diversity and is based on measuring the number of food groups consumed over a given reference period. A higher dietary diversity is associated with a higher likelihood of an adequate intake of essential micronutrients. The score has been shown to be associated with nutrient density (Moursi et al., 2008a, Dewey et al., 2006) and nutrient adequacy (Kennedy et al., 2007, Steyn et al., 2006) in breastfed and non-breastfed children. To date, there is only one study that looked into the stability of a summary child feeding index (of which DDS is a component) and its association with child growth over different time points (Moursi et al., 2008b). As the seasonal influence on dietary intake and nutrient density of local foods was demonstrated to be substantial, seasonal differences can be expected to affect the validity of simple dietary indices to predict dietary quality (Winichagoon, 2008).

To our knowledge, there is no study that assessed the validity of DDS using quantitative dietary intake data collected from breastfed children 6-12 months of age in two distinct seasons. The aim of this study was therefore to determine how seasonality affects the validity of a dietary diversity score in predicting nutrient adequacy of complementary foods for breastfed infants in southwest Ethiopia. We also identified an appropriate cut-off to predict "low" or "higher" dietary quality, and assessed differences between seasons.

4.3. Methods and Subjects

Sampling and design

A census of all infants aged 6-12months was carried out within the Gilgel Gibe Field Research Center (GGFRC) of Jimma University (southwest Ethiopia). Infants aged 6-12 months and born in the GGFRC, who have a biological mother or female caregiver, presenting no serious illness or malformation affecting anthropometric measurement, and no intention of leaving the area in the coming 6 months were listed. This procedure was applied in two seasons: during harvest season (HS) from October-December 2009, and pre-harvest season (PHS) from June-August of 2010. A total of 320 and 312 eligible infants were included in the HS and PHS respectively.

Study setting

The study was conducted in nine *kebeles* (smaller government unit) of the catchment area of GGFRC of Jimma University, southwest Ethiopia. The total population within the area in 2011 was 54,528 of which 15,719 (28.8%) were located in urban and 38,809 (71.2%) in rural communities. The main source of income for the community is farming and raising livestock. Maize, sorghum and teff (a species of *Eragrostis* native to Ethiopia) are commonly grown food crops in the area. Food consumption patterns of the population are expected to be different based on the differences in food availability in the two seasons.

Data collection

We used pre-tested interviewer administered questionnaires to collect data on socioeconomics and demographics, and infant feeding practices. Questions were adapted from Demographic and Health Survey (DHS) questionnaires (CSA [Ethiopia] and ORC Macro, 2006) and the World Health Organization (WHO) guideline for the assessment of infant and young child feeding (WHO, 2010b). We collected dietary intake data using a multiple pass 24-hr recall questionnaire adapted for commonly consumed foods in the study area (Gibson and Ferguson, 2008). Data collectors were fluent in local languages (Amharic and Afan Oromo), and received a five-day intensive training from the principal investigators.

Dietary intake assessment

Two 24-hr recalls of the infants' diet were conducted in each season. Compared to a weighed record method, the interactive 24-hour recall method is less expensive and burdensome for the respondents.

Spoons and graduated bowls were provided to mothers/caregivers weeks before the interview. They were told to use them during the coming weeks. Instructions were provided on how to estimate the portion sizes consumed by their infant using the utensils provided and pre-standardized household measures. Two sources of recipe data were used to convert composite dishes to ingredients. First, we collected recipe data of common composite dishes by asking five mothers to prepare them. For this purpose, ingredients were weighed using scales during the preparation. Secondly, we used averaged recipe data that was available from the Ethiopian traditional recipe table (ENI (MOH), 1980). During the 24-hr recall we assessed the recipe qualitatively, and matched it with the best available recipe. Foods that come in discrete units were recalled and converted into weights by using conversion lists that were compiled from weighing all such foods. A 1g precision laboratory scale (Ohaus Corp., USA) was used to record weights for the different sizes of household measures, bowls, and spoons provided to the mothers or caregivers.

During the interview, mothers were requested to show the type and amount of foods the infant had consumed in the last 24 hours. A pre-recorded conversion table was used that converts volumetric estimations of gruels and porridges to weight equivalents according to three reported consistencies (thin, medium, and stiff). All days of the week were represented in the final sample in order to offset the day-of-the-week effects on food or nutrient intake. The two 24 hour dietary recalls were separated by ±15days in both seasons. Moreover, the days of the recall were not announced after the preparatory visits to avoid systematic error or bias.

Food composition data were obtained from various sources. Values for energy and water soluble vitamins were compiled from the Ethiopian food composition table, and FAO's food composition table for Africa (EHNRI, 1998a, EHNRI, 1998b). Some missing foods and nutrient values were obtained from an analysis paper (Abebe et al., 2007), the food tables for East Africa (C E West et al., 1988) and the USDA (Gebhardt et al., 2010). Missing nutrient values were imputed using the formula by Gibson and Ferguson (Gibson and Ferguson, 2008). We were only able to use retinol, and trans-β-carotene concentrations to calculate the vitamin A activity due to the lack of data on other carotenoids (Dewey and Brown, 2003). We used the Lucille food intake program (DFSFQ, n.d)to enter consumed foods, and calculate corresponding nutrient values.

Dietary diversity score and micronutrient density adequacy

Food group consumption was recalled separately during the second day of the 24-hr dietary recall using the food group classification as suggested by Dewey et al.(Dewey et al., 2006). Dietary diversity score was obtained by summing the number of food groups consumed in the previous 24 hours by the infant. The following seven food groups were included: a) Grains, roots and tubers; b) Legumes and nuts; c) Dairy products; d) Flesh foods; e) Eggs; f) Vitamin A-rich fruits and vegetables (>130 retinol equivalents/100g); and g) Other fruits and vegetables. As recommended in the WHO infant and young child feeding (IYCF) practice measurement guide(WHO, 2008), fats and oils were not used in the analyses.

To validate the DDS, we used the average of two day micronutrient intakes fromcomplementary foods to calculate the micronutrient density adequacy (MDA) scores as proposed by Dewey et al. (Dewey et al., 2006). Micronutrient density adequacy of complementary foods was calculated based on nutrient density (per 100 kcal) rather than absolute nutrient intake, in order to account for the unknown variability in breast milk intake. Micronutrient densities per 100 kcal of complementary foods were calculated for two age categories (6-8 months and 9-12 months), and were compared with estimated desired nutrient densities. The desired nutrient densities were calculated for each nutrient using the formula as suggested by Dewey et al. (Dewey et al., 2006) (Formula 1).

Desired nutrient density of complementary foods for nutrient “x” =

$$\frac{\text{RNI of nutient "x"} - (\text{Nutrient "x" concentration in breast milk} \times \text{"average" breast milk intake volume})}{\text{Energy needed from complementary foods when breast milk intake is "average"}} \times 100 \ldots (\textbf{Formula 1})$$

Eight micronutrients (vitamin A and C, thiamine, riboflavin, niacin, iron, calcium, and zinc) were used for the calculation of MDA. The contribution to the overall nutrient intake by breast milk was subtracted from the Recommended Nutrient Intakes (RNI) of each micronutrient to obtain the required amount from complementary foods for all breastfed children. We assumed an “average” breast milk intake of 674 and 616g/d for 6-8 and 9-12 month old infants respectively; and we used the breast milk concentration of each micronutrient as documented by WHO (WHO, 1998). The RNI values of vitamins were taken from WHO/FAO (FAO/WHO, 2004). Bioavailability assumptions were made for calcium, iron, and zinc in order to compare nutrient intake and requirements. For calcium, iron, and zinc, ‘absorbed’ recommended amount was the basis for the calculation. For our analysis, 30% and 10% of the recommended intake of calcium and iron in complementary foods were assumed absorbed respectively (Dewey et al., 2006). Following international recommendations, 15% bioavailability was assumed for zinc from low bioavailability foods (WHO, 1996). The percentage of calcium, and iron absorbed from breast milk was estimated at 30% (Institute of Medicine, 1997) and 20% (Magnus Domellöf et al., 2002), respectively, while the percentage of zinc absorbed from breast milk was estimated at 50% (Institute of Medicine, 2001). The estimated energy requirements from complementary foods, assuming “average” breast milk intake, were 202 kcal at 6-8 months and 307 kcal at 9-11 months (Dewey and Brown, 2003).

After determining the desired nutrient density for each of the eight nutrients, MDA score per 100 kcal of complementary foods was calculated as the percentage of the desired nutrient density fulfilled by the complementary foods for breastfed 6-8 and 9-12 month old infants **(Formula 2).**

MDA for nutrient “x” =

[((nutrient "x" intake / complementary food energy intake) x 100) / desired nutrient density] x100…**(Formula 2)**

The mean of the individual MDAs was calculated to give mean micronutrient density adequacy (MMDA), which was then capped at 100%. We evaluated the sample's MMDA using two arbitrary cut-offs, used previously (Dewey et al., 2006, Moursi et al., 2008a)to assess dietary quality: MMDA <50% and MMDA ≥75%, representing "low" and "higher" dietary quality respectively.

We compared the observed nutrient intakes to the desired nutrient intake from complementary foods assuming every child had an "average" breast milk intake (g/day) as proposed by WHO for each age group(WHO, 1998). This time the desired nutrient density per 100kcal of complementary food was not calculated.

We assessed the contribution of the seven complementary food groups to the mean (SD) of the two days daily intakes of energy, protein, fat and micronutrients, and disaggregated by season to understand which groups are responsible for the intake of energy and micronutrients by the target infants. It provides additional information on the pattern of food group contribution to nutrient intakes.

Anthropometry

Anthropometric measurements were standardized through repetitive exercises. Recumbent length was measured on an infant measuring board and recorded to the nearest 0.1cm (SECA 210, Hamburg, Germany). Weight of the infant was measured naked or with very light clothes using mother-child digital scales, and recorded to the nearest 0.1kg (SECA Uniscale, Hamburg, Germany). Ages were recorded from birth certificates or immunization cards as often as possible. If the ages were absent, local events calendars were used to help the mother or caregiver estimate approximate age of the child. Most of the anthropometric data collectors used in HS were available for anthropometric measurement in the PHS after 5-6 months.

Data analysis

The anthropometric measures were converted into Z-scores of weight-for-age (WAZ), length-for-age (LAZ), and weight-for-length (WLZ) based on the WHO 2006 Growth Reference (WHO, 2010c). Underweight, wasting, and stunting were defined as <-2 WAZ, WLZ and LAZ respectively. Severe underweight, wasting, and stunting were defined as <-3 WAZ, WLZ and LAZ respectively.

We constructed an asset-based proxy to estimate the socioeconomic status (SES) of the included households. Items included were ownership of cattle, house construction materials, land (homestead, land under cultivation, fallow land) and ownership of other household assets. For household assets, categories for each item were 'owned' or 'not owned' by the household. The first principal component (56% of the variance) from a principal components analysis was divided in tertiles and used as a proxy for SES (Vyas and Kumaranayake, 2006).

Descriptive analyses were conducted for socio-demographic data, nutritional status, and percentage of the study population consuming each food group in relation to dietary diversity stratified by season. Means (±SD) were reported for continuous data that were normally distributed. Differences in means were statistically tested using a student *t*-test, while a χ^2-square test was used to compare categorical variables. As most distributions of nutrient intake data were skewed, we first presented median intakes (25-75 percentiles) by season for descriptive purposes. Secondly, we used multiple linear regression models to compare the mean energy and nutrient intakes by season, adjusting for significant differences in characteristics between the two seasons (HS and PHS).

We looked at bivariate associations between child, caregiver, household characteristic, and MMDA using simple linear regression models. Every variable with a modest association ($P<0.20$) was a candidate to be included in the multivariate analysis. A full model assessing the relation between DDS and MMDA was compiled after which non-significant variables were removed by a backward procedure using a likelihood ratio test ($P<0.05$).

We determined cut-offs in terms of number of food groups, at which we could classify infants at a reasonable MMDA with sufficient sensitivity and acceptable specificity. Hence, performance of DDS to indicate dietary quality was evaluated by receiver operating characteristics (ROC) analysis. The area under the curve (AUC) represents the probability of DDS as an indicator of dietary quality. An AUC above 0.50 indicates prediction of dietary quality by DDS; the higher the AUC value of dietary diversity indicators, the better the prediction.

Based on previous analysis(Moursi et al., 2008a, Dewey et al., 2006), sensitivity, and specificity stratified by season were determined to compare the performance of the DDS in correctly differentiating cases with MMDA <50% ("low" MMDA) or ≥75% ("higher" MMDA) for infants 6-8 and 9-12 months of age. When using DDS to predict "low" MMDA, the sensitivity indicates the fraction of infants who are correctly classified as having "low" MMDA; whereas the specificity indicates the fraction of infants who are above the cut-off for "low" MMDA and are correctly classified as such by DDS. For "higher" MMDA, the sensitivity indicates the fraction of infants who are correctly classified as having "higher" MMDA, whereas the specificity refers to the fraction of infants who are below the cut-off for "higher MMDA" and are correctly classified as such by DDS. Data was analyzed with Stata 12.0 (StataCorp, College Station, TX, USA). Statistical significance was set at 5% and all tests were two-sided.

Ethical considerations

The study was approved by the Ethics Review Board of Jimma University, Ethiopia (RPGC/104/2002) and the Ethics Committee of Ghent University Hospital, Belgium (B67020109188). Informed consent was secured from the mothers/caretakers before conducting the interviews and assessment

4.4. Results

Data was collected from 320 and 312 infants during HS and PHS respectively. A total of 45 infants (24 in HS and 21 in PHS), for whom MMDA could not be calculated, were excluded from analyses. Of these 45 there were 15 infants (n=10 in HS and n=5 in PHS) from whom incomplete dietary intake data was collected by the interviewers, 21 were not breastfeeding their children (n=11 in HS and n=10 in PHS), and 9 caregivers were unable to provide a reliable child age estimate (n=3 in HS and n=6 in PHS). The LAZ, WAZ, and WLZ of excluded observations were not statistically different from the rest of the observations in both seasons. Almost all primary caregivers (≥96%) were biological mothers.

Pre-harvest season mothers/caregivers were slightly older (26.6±4.4 vs. 26.0±5.7) compared to the HS sample, while there were more male children (57% vs. 48%) in the PHS sample. Infants from the PHS were on average older (9.7±1.6 vs. 8.6±1.8 months), had a lower mean LAZ and reported more diarrheal episodes compared to the HS sample. The majority of the mothers/caregivers in both seasons never attended formal education **(Table4.1).** Continued breastfeeding was reported in all infants included in this analysis, with no difference between seasons. However, most of them were started with additional foods or drinks earlier than the recommended time (6 months of age). The median duration of exclusive breastfeeding was 4.0 (IQR=3.0, 4.0) and 4.0 months (IQR=3.0, 6.0) in HS and PHS respectively. Eighty-eight and sixty-seven percent (P<0.05) of the infants were introduced to complementary foods before 6 months during HS and PHS respectively.

The mean DDS was 2.1 (SD=0.94) and 2.3 (SD=1.1) in HS and PHS respectively. Only 8% and 13% of the infants were taking four or more food groups; while 69% and 63% of infants were taking two or less food groups in the previous day of the interview in HS and PHS respectively. There was no difference on the recorded DDS between both seasons (P=0.09). The mean MMDA was 0.59 (SD=0.15) and 0.61 (SD=0.13) during HS and PHS respectively. The relatively low mean MMDA in both seasons is indicative of low micronutrient nutrient density of the complementary foods given to infants. The overall MMDA was significantly lower during the HS (P=0.02). However, after adjusting for infant age this difference was not significant **(Table 4.1).**

We found important differences in energy and micronutrient intakes between seasons. After adjusting for some sample characteristics which are statistically different between the two seasons, energy, protein, riboflavin, vitamin C, and absorbed calcium intakes were larger during the PHS than HS (**Table 4.2**). In contrast, we found that thiamine intake was lower during the PHS compared to HS. The median intake of vitamin A, iron, and zinc were lower than what is recommended from complementary foods, but the energy and protein contribution from complementary foods was higher than the desired values (**Table 4.3**).

Table 4.1: Characteristics of the study population by season

Variables	HS	PHS	*P*-value
Sex of the child , n and %			
Female	154 (52.0)	125 (43.0)	0.03
Age of the child, n and %			
6-8 months	152 (51.4)	71 (24.4)	<0.001
9-12 months	144 (48.6)	220 (75.6)	
Age of the mother , n and %			
<20 years	60 (20.6)	32(11.0)	0.004
20-34 years	202 (69.4)	233(80.1)	
>=35 years	29 (10.0)	26 (8.9)	
Socio-economic index , n and %			
Low	79 (27.3)	115 (39.9)	0.006
Medium	106(36.7)	89 (30.9)	
High	104 (36.0)	84 (29.2)	
Under five children in the HH , n and %			
0-1 child	79 (26.7)	66 (22.7)	0.26
>=2 children	217 (73.3)	225 (77.3)	
Cough* , n and %			
Yes	44 (14.9)	57 (19.6)	0.13
Fever * , n and %			
Yes	57 (19.3)	67 (23.0)	0.27
Diarrhoea * , n and %			
Yes	49 (16.7)	70 (24.1)	0.03
Source of drinking water for HH			
Unsafe water source	252(85.1)	229 (78.7)	0.04
Safe water source	44 (14.9)	62 (21.3)	
Human waste disposal by the HH , n and %			
Improper disposal	117 (39.7)	97 (33.3)	0.11
Proper disposal	178 (60.3)	194 (66.7)	
Maternal schooling , n and %			
Not attended	243 (82.1)	246 (84.5)	0.43
Attended formal school	53 (17.9)	45 (15.5)	
LAZ , mean and SD	-0.77 (1.4)	-1.0 (1.3)	0.02
Stunting , n and %			
LAZ <-2	55 (18.6)	61 (21.1)	0.44
LAZ<-3	18 (6.1)	16 (5.5)	0.78
WLZ , mean and SD	-0.63 (1.3)	-0.75 (1.3)	0.28
Wasting , n and %			
WLZ <-2	35 (11.8)	45 (15.5)	0.19
WLZ<-3	12 (4.1)	12 (4.1)	0.96
WAZ , mean and SD	-0.96 (1.2)	-1.1 (1.4)	0.08
Underweight , n and %			
WAZ <-2	52 (17.6)	75 (25.8)	
WAZ<-3	18 (6.1)	20 (6.9)	
Duration of EBF, months , median and	4.0 (3,4)	4.0 (3,6)	<0.001
Meal frequency , median and IQR†‡	2.0 (0,3)	2.0 (1,3)	0.43
DDS7 , mean and SD	2.1 (0.94)	2.3 (1.1)	0.09
MMDA , mean and SD	0.59 (0.15)	0.61(0.13)	0.02

HS, harvest season; PHS, pre-harvest season; DDS, dietary diversity score; MMDA , mean micronutrient density adequacy ; EBF, exclusive breast feeding ; IQR , Interquartile range ;
*Diarrhoea, cough, fever in the previous 2 weeks of the surveys
†Frequency of consumption of solid/semisolid or soft food;‡Two-sample Wilcoxon rank-sum (Mann-Whitney) test was used to calculate the p-value

Table 4.2: Infant energy and nutrient intakes during harvest and pre-harvest season (HS, n=296; PHS, n=291)

Nutrient intake from complementary food/day	Harvest Season[1]	IQR	Pre-harvest Season[1]	IQR	Mean Difference[2]	95% CI	*P*-value
Energy, kcal	233	159-305	280	197-347	40.0	22.0, 58.0	<0.001
Protein, g	6.7	4.5-9.2	8.0	5.1-10.6	1.10	0.48, 1.7	0.001
Vitamin A, RE	9.0	1.2-18.0	1.9	0.6-11.2	-0.34	-4.2., 3.48	0.86
Thiamine, mg	0.16	0.10-0.22	0.13	0.08-0.18	-0.03	-0.04, -0.14	<0.001
Riboflavin, mg	0.21	0.11-0.34	0.29	0.13-0.46	0.08	0.05, 0.11	<0.001
Vitamin C, mg	1.5	0.52-3.8	4.1	2.1-7.7	2.20	0.65, 3.70	0.005
Niacin, mg	0.67	0.35-1.01	0.70	0.45-1.2	0.08	-0.03, 0.18	0.11
Absorbed Calcium, mg	28	15-45	34	13-58	8.20	4.0,12.0	<0.001
Absorbed Iron, mg	0.69	0.24-1.44	1.01	0.32-1.65	0.13	-0.02, 0.29	0.10
Absorbed Zinc, mg	0.16	0.11-0.21	0.17	0.11-0.23	0.01	0.001,0 .03	0.04

RE, retinol equivalent; CI, confidence interval; IQR, Interquartile range

[1]Actual intakes are median values and interquartile range

[2] Difference of means and p-values are obtained from multiple linear regression model adjusting for child age, maternal age, and child sex, dairrhoea

Table 4.3: Desired and actual nutrient intake of infants from complementary foods in harvest and preharvest seasons

Nutrient intake from complementary food /day	Harvest Season						Pre-harvest Season					
	6-8 months (n=152)			9-12 months[3] (n=144)			6-8 months (n=71)			9-12 months[3] (n=220)		
	Desired[1]	Actual[2]	IQR	Desired	Actual	IQR	Desired	Actual[2]	IQR	Desired	Actual	IQR
Energy, kcal	202.0	232.0	159-309	307.0	277.0	158-301	202.0	275.0	188-352	307.0	282.0	202-343
Protein, g	2.0	6.6	4.8-9.5	3.1	6.7	4.0-9.0	2.0	8.3	4.9-11.5	3.1	7.1	0.60-15
Vitamin A, RE	63.0	12.0	7.0-31	92.0	8.0	0.60-15	63.0	1.4	0.30-5.5	92.0	2.3	0.6-15
Thiamin, mg	0.2	0.2	0.1-0.2	0.2	0.2	0.10-0.20	0.2	0.1	0.1-0.18	0.2	0.1	0.1-0.18
Riboflavin, mg	0.2	0.2	0.1-0.4	0.2	0.2	0.1-0.3	0.2	0.4	0.20-0.50	0.2	0.3	0.13-0.5
Vitamin C, mg	3.0	1.5	0.5-3.7	5.0	1.3	0.3-4.0	3.0	3.8	2.0-6.2	5.0	4.2	2.0-7.9
Niacin, mg	3.0	0.7	0.3-0.9	3.0	0. 7	0.4-1.1	3.0	0.7	0.4-1.1	3.0	0.7	0.5-1.2
Absorbed Calcium, mg	24.0	25.0	15-39	29.0	20.0	10-35	24.0	31.0	14-50	29.0	28.0	5.0-88
Absorbed Iron, mg	0.9	0.6	0.2-1.4	0.9	0.9	0.3-1.6	0.9	1.0	0.1-1.9	0.9	1.0	0.2-1.9
Absorbed Zinc, mg	0.7	0.2	0.1-0.2	0.7	0.2	0.16-0.31	0.7	0.2	0.1-0.3	0.7	0.3	0.16-0.34

RE, Retinol Equivalent; IQR, Interquartile range

[1]Desired nutrient intakes from complementary foods are calculated based on the assumption of “average” breast milk intake. Values are obtained after subtracting the estimated amount contributed by breast milk from the RNI. The RNI absorbed was the basis for the calculation. Absorption of zinc, calcium and iron was assumed to be 15 %, 30%, and 10% respectively.

[2]Actual intakes are the median values and interquartile rang

[3] Relatively few infants were 12 months old and most of the recommended intakes for a 12 month old overlap with that of 11 month old infant. They were categorized as 9-12 months and desired intake calculations were based on the recommendation of 9-11month old infants.

Consumption of different food groups varied with DDS. Staples like cereals, tubers, and roots were consumed most frequently in both seasons without a lot of discriminatory value between children. Beyond this food group, the DDS primarily increased through the consumption of legumes, and dairy (**Table 4.4**). The majority of the infants consumed vitamin A-rich fruits and vegetables, and other fruits and vegetables if their DDS was 3 or higher. Flesh foods including fish were the least consumed by the study children. Further analysis was performed to explain as to which food groups are responsible for the difference observed in individual nutrient intake between the two seasons. Dairy was the main source of calcium (39.1mg in HS and 50.3 mg in the PHS) in both seasons followed by eggs, other fruits and vegetables, and legumes; while vitamin A-rich fruits and vegetables, and other fruits and vegetables are important contributors of vitamin C intake in both seasons (**Table 4.5, 4.6**). The percentage contribution for energy and each micronutrient intake amoung cereal consumers was relatively high relative to other food groups which highlights the importance of cereals in the diet of the study children **(Table 4.6**). The later table was constructed from the food groups derived from quantitative food intake data, however, both tables demonstrate a similar pattern of contribution of the food groups to nutrient intake.

In multiple linear regression analysis, we found a significant and positive association between DDS and MMDA irrespective of season after adjusting for important confounders. For any additional increase in DDS by one unit there was an increase in MMDA by 4.5% and 4.4% in HS and PHS respectively (**Table 4.7**).

Receiver operating characteristic analysis was performed using the MMDA<50% and MMDA ≥75% cut-offs in both seasons. We found AUC values of 0.64 (95%CI: 0.60, 0.67) and 0.76 (95%CI: 0.80, 0.75) for MMDA <50% in HS and PHS respectively. The AUC values for MMDA ≥75% cut-off were 0.67 (95%CI: 0.53, 0.81) and 0.58 (95%CI: 0.47, 0.69) in HS and PHS respectively. All AUC values were significantly higher from a null value of 0.50 for MMDA<50% but not for MMDA ≥75% in PHS.

After performing a sensitivity and specificity analysis for the two MMDA cut-offs, we found that a DDS of ≤2 food groups performed best in predicting "low" MMDA with 84% and 92% sensitivity, 36% and 43% specificity, and 47% and 51% correct classification in HS and PHS respectively. The prediction of DDS for “low” MMDA remained valid when the analysis was disaggregated by age groups in both seasons (Figure 1). A DDS of ≥3 produced the trade-off between sensitivity and specificity values to predict “higher” MMDA with relatively low misclassification; but the sensitivity values were rather small (32% in HS and 24% in PHS). Age adjusted ROC analysis was performed to predict “higher” MMDA, but we could not find an acceptable trade-off between sensitivity and specificity in both seasons as very small proportion of the infants (6.4 in HS and 8.6% in PHS) achieved MMDA ≥75%.

Table 4.4: Distribution food groups consumed by infants by dietary diversity score and stratified by season

DDS*	Cereals, tubers and roots		Legumes and nuts		Vitamin A rich fruits and vegetables		Other fruits and vegetables		Flesh foods		Eggs		Dairy	
Harvest season (n=296)														
	n†	%‡	n	%	n	%	n	%	n	%	n	%	n	%
1	57	74	1	1	0	0	0	0	0	0	1	1	18	23
2	121	98	27	22	8	7	6	5	1	1	3	2	80	65
3	67	99	46	68	9	13	18	27	4	6	7	10	53	78
4	19	100	13	68	9	47	15	79	0	0	4	21	16	84
5	5	100	5	100	5	100	4	89	1	20	0	0	5	100
Pre-Harvest Season (n=291)														
1	55	75	1	1	2	3	0	0	0	0	1	1	14	19
2	104	95	32	29	11	10	17	16	0	0	4	4	52	47
3	68	99	46	67	25	36	12	17	1	1	12	16	43	62
4	29	97	22	73	19	63	18	60	0	0	10	30	18	60
5	8	100	8	100	6	75	7	88	1	13	3	42	7	88
6	1	100	1	100	1	100	1	100	0	0	1	100	1	100

*DDS, Dietary Diversity Score

†Total number of subjects who consumed each food group; ‡Percentage of subjects who consumed each food group. Note that the numbers and percentages are not mutually exclusive; as an infant can consume more than one food group at a time.

Table 4.5: The contribution of food groups to nutrient intake in harvest and preharvest seasons among consumers*

Food groups (n) [a]	**Energy (kcal)**		**Protein (g)**		**Fat (g)**		**Calcium (mg)**		**Zinc (mg)**		**Iron (mg)**		**Retinol (micg)**		**Thiamine (mg)**		**Riboflavin (mg)**		**Ascorbic acid (mg)**		**Niacin (mg)**	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Harvest season																						
Cereals (269)	238.6	93.6	7.0	3.2	8.5	4.7	29.4	19.4	0. 2	0.1	1.0	0.8	11.7	13.2	0.2	0.1	0.2	0.1	4.6	8.4	0.8	0.5
Legumes and nuts (92)	240.8	98.9	7.0	3.5	8.2	4.6	27.6	18.1	0.2	0.1	1.2	0.8	10.1	10.9	0.2	0.1	0.2	0.1	6.0	10.4	1.0	0.5
Vitamin A rich fruits and vegetables (31)	243.4	102.5	6.9	2.8	8.4	4.3	30.5	16.7	0.2	0.1	0.2	0.1	17.5	24.3	0.2	0.1	0.2	0.1	12.4	16.0	0.8	0.5
Other fruits and Vegetables (43)	261.4	86.6	7.5	3.0	9.6	4.4	33.2	17.6	0.2	0.1	1.0	0.8	20.3	21.0	0.2	0.1	0.3	0.1	9.6	12.1	0.9	0.5
Flesh foods (6)	250.3	123.0	6.5	3.7	8.5	7.4	20.7	19.4	0.2	0.1	1.1	0.3	7.0	9.2	0.2	0.1	0.2	0.1	6.3	8.5	1.2	0.9
Eggs (15)	310.2	103.9	8.6	3.5	13.2	5.5	35.1	16.8	0.2	0.1	0.9	0.8	12.8	16.9	0.2	0.1	0.2	0.1	3.9	6.7	1.0	0.7
Dairy (172)	261.8	85.4	8.3	3.0	10.0	4.3	39.1	18.6	0.2	0.1	0.2	0.1	16.7	14.4	0.2	0.1	0.3	0.1	5.4	9.1	0.7	0.5
Pre-harvest season																						
Cereals (265)	279.5	113.9	8.1	4.0	10.9	6.4	37.1	27.1	0.2	0.1	1.2	1.0	12.4	27.1	0.1	0.1	0.3	0.2	7.0	9.2	0.9	0.6
Legumes (110)	298.1	113.9	8.8	3.9	12.1	6.7	40.9	26.9	0.2	0.1	1.4	1.0	12.2	34.9	0.1	0.1	0.3	0.2	8.2	10.1	1.2	0.7
Vitamin a rich fruits and vegetables (64)	288.3	140.4	8.1	4.6	10.8	7.4	38.0	28.0	0.2	0.1	1.3	1.0	14.3	22.1	0.1	0.1	0.3	0.2	13.4	12.6	1.0	0.7
Other fruits and vegetables(55)	313.0	114.9	8.6	3.7	11.9	6.0	36.4	23.7	0.2	0.1	1.3	1.2	18.3	46.1	0.1	0.1	0.3	0.2	10.7	8.4	1.3	0.7
Flesh foods (2)	304.4	46.4	8.3	1.1	12.2	0.4	27.8	21.2	0.1	0.1	1.6	1.2	3.8	2.4	0.1	0.1	0.2	0.2	2.5	4.8	1.4	1.2
Eggs (31)	373.7	112.4	11.4	3.6	17.7	6.5	51.6	25.4	0.3	0.1	1.1	0.8	21.0	59.7	0.2	0.1	0.4	0.2	9.7	10.6	1.0	0.8
Dairy (135)	301.1	96.3	9.4	3.1	13.1	4.9	50.3	21.0	0.2	0.1	1.2	1.0	12.7	32.9	0.2	0.1	0.4	0.2	7.1	8.5	0.8	0.6

SD, Standard Deviation

*All values are means and SD of energy, protein, fat and micronutrient intakes from each complementary food group as consumed by infants in harvest and preharvest seasons. For calcium, iron and zinc the absorbed values are used in the calculation

n, the number of infants consuming the food group (the values are not mutually exclusive)

aThe data is obtained from the recall of food groups consumed in one of the days of the 24hours recalls to measure detail quantitative intake

Table 4.6: The perentage contribution of food groups to energy and nutrient intake in harvest and preharvest from those who consumed the food group[a]

Food groups[a]	**n**	**Energy**	**Protein**	**Fat**	**Calcium**	**Iron**	**Zinc**	**Retinol**	**Thiamine**	**Riboflavin**	**Ascorbic acid**	**Niacin**
Harvest season												
Cereals	305	52.1	45.7	21.4	29.2	62.9	59.4	20.1	49.9	21.4	69.3	63.7
Legumes and nuts	142	20.8	24.6	31.5	21.9	31.5	13.6	0.2	13.8	29.8	38.2	58.3
Vitamin A rich fruits and vegetables	43	8.4	5.5	1.0	12.2	5.3	5.4	11.7	5.2	24.9	0.1	21.5
Other fruits and Vegetables	79	12.3	6.4	47.5	9.8	4.9	7.4	44.2	7.8	16.7	0.2	21.3
Eggs	20	31.0	41.0	22.5	31.5	20.6	37.7	14.3	27.2	37.7	13.5	21.4
Flesh foods	8	16.0	37.2	69.3	19.3	12.8	12.4	0.1	8.3	5.5	1.3	18.3
Dairy	233	39.7	51.6	40.3	73.5	26.4	40.4	79.7	52.6	52.7	5.8	0.6
Fats and oils	65	14.9	0.1	40.3	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.2
Pre harvest season												
Cereals	297	47.5	42.8	22.8	28.9	59.8	52.5	49.6	50.5	20.1	58.5	51.6
Legumes and nuts	154	17.7	21.8	27.6	21.2	32.2	12.3	0.1	14.8	24.6	17.4	53.5
Vitamin A rich fruits and vegetables	89	7.2	3.4	1.6	8.0	1.8	1.9	53.6	16.0	17.1	0.0	16.6
Other fruits and Vegetables	101	18.0	9.8	44.0	6.3	5.6	13.0	49.0	4.7	23.3	0.3	35.0
Eggs	38	28.4	34.5	29.0	19.2	18.4	41.4	3.7	28.3	23.1	3.1	13.4
Flesh foods	13	19.3	39.0	69.5	17.2	22.7	31.4	5.8	32.0	14.5	9.8	18.0
Dairy	217	41.6	51.6	32.2	73.3	25.9	44.0	15.5	44.3	55.4	3.1	0.7
Fats and oils	49	12.2	0.4	32.2	0.7	0.1	0.2	2.2	0.1	0.1	0.0	0.3

n, the number of infants consuming the food group (the values are not mutually exclusive)

[a]The data is obtained from the quaqntitative 24 hours recall

Table 4.7: The association between dietary diversity score and mean micronutrient density adequacy stratified by season†

Variables	**MMDA (HS)**				**MMDA (PHS)**			
	Crude beta	**95% CI**	**Adjusted beta**	**95% CI**	**Crude beta**	**95% CI**	**Adjusted beta**	**95% CI**
DDS	0.047	0.03,0.06 ***	0.045	0.028,0.062***	0.043	0.029,0.057***	0.044	0.031,0.058***
Age of the child								
6-8 months	Ref		Ref		Ref		Ref	
9-12 months	0.061	0.029,0.094**	0.050	0.018,0.082**	0.021	0.01, 0.03**	0.081	0.047, 0.110***
Age of the mother								
<20 years	Ref		Ref					
20-34 years			0.061	0.016,0.10**				
>=35 years	0.067	0.001,0.063*	0.080	0.016,0.14*				

DDS, Dietary Diversity Score; MMDA, Mean Micronutrient Density Adequacy**;** HS, Harvest Season; PHS, Preharvest Season

**P*<0.05; ***P*<0.01 ****P*<0.001

†Adjustments were made for socio-economic index, child age, maternal age, maternal schooling, diarrhoea, cough, fever, gender, and height-for-age z-score. Variables for which an association was found for at least one seasons are tabulated.

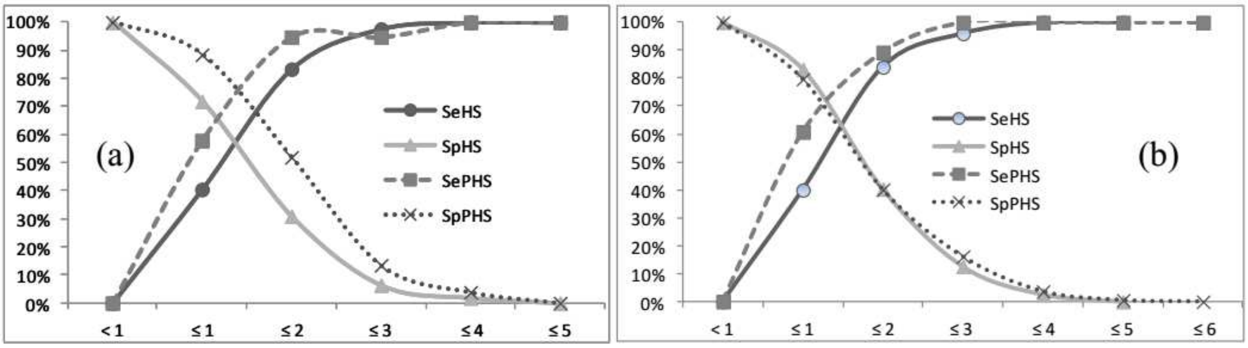

Figure 4.1: Sensitivity and specificity of dietary diversity score (DDS) to predict "low" nutrient intake, MMDA<50%, for breastfed infants 6-8 (a) and 9-12 (b) months of age. SeHS, sensitivity in Harvest Season; SpHS, specificity in Harvest Season; SePHS, sensitivity in Pre-harvest Season; SpPHS, specificity in Pre-harvest Season. Sensitivity indicates the percentage of children having “low” MMDA who are identified as such by DDS. Specificity indicates the percentage of children who are above the cut-off for “low” MMDA and are correctly identified as such by DDS.

4.5. Discussion

The present study showed that a DDS calculated from seven food groups was able to predict diet quality expressed in term of micronutrient density adequacy in breastfed infants, and that seasonality did not alter this relationship, notwithstanding the significant difference in intakes of most of the nutrients between seasons.

The regression analysis showed significant association between DDS and MMDA in both seasons. After adjusting for maternal, socioeconomic, and child factors, there was no major attenuation of the regression coefficients. A positive association between DDS and dietary quality, albeit expressed by different indicators or scores, among breastfed and non-breastfed children was demonstrated previously by other authors with different correlation coefficients (Moursi et al., 2008a, Steyn et al., 2006, Kennedy et al., 2007 , Hatloy et al., 1998). However, most of such studies were conducted during a specific season which hampers external validity. Other studies pooled data over different seasons but did not present data on seasonal confounding (Working Group on IYCF Indicators, 2006, Dewey et al., 2006, Moursi et al., 2008a).

There was a relatively higher mean nutrient intake per food group consumed for most of the nutrients in the PHS compared to HS. However no difference in DDS and age-adjusted MMDA was noted between the two seasons. On the other hand some food groups like flesh foods and dairy were consumed less frequently during the PHS. These two observations suggest that the relatively higher overall nutrient intake during the PHS was mainly the consequence of larger average portion sizes, and/or a higher intra-day frequency of food group consumption. Cereals, tubers, and roots were the most commonly consumed foods in both seasons in this study. Similar observations were made in other developing countries (Moursi et al., 2008a, Kennedy et al., 2007). However, there was inconsistency regarding the consumption pattern of other food groups in relation to DDS in the present study in which dairy and flesh foods were relatively more consumed in the HS.

The mean MMDA for this study sample was suboptimal indicating inadequate intakes of one or more micronutrients. In a similar analysis in the same age group in Madagascar (Moursi et al., 2008a), most of the nutrients were consumed in amounts less than desired. Another study conducted during the pre-harvest season in southern Ethiopia demonstrated that all the nutrients measured were significantly lower than the “estimated need” calculated (Gibson et al., 2009) even if comparable reference values were used to calculate the “estimated need” adjusted for age and body weight. The dietary reference intake value for calcium used in our study was based on an adequate intake (AI) (Institute of Medicine, 1997) which is lower than the UNICEF/WHO (WHO, 1998) and FAO/WHO/UNU (FAO/WHO/UNU, 2004) reference values used in the study in southern Ethiopia. Furthermore, the significant gap between actual and desired/estimated need could be due to a relatively high prevalence of chronic food insecurity in that part of South Ethiopia compared to our study area (Regassa and Stoecker, 2012). Overall, the density approach of MMDA allows to adjust for the unknown breast milk intake assuming a balance between energy intake from both complementary foods and breast milk, and assuming average breast milk intake levels as proposed by WHO(WHO, 1998). Consequently, if actual breast milk intake is less than the proposed average, calculations show a higher mean desired nutrient density and desired intake, which implies lower MMDA and higher deficit between desired and actual intakes of energy and nutrients.

We did not find important differences in MMDA and DDS, between PHS and HS. Conversely, with the exception of vitamin A and thiamine, energy and nutrient intake were generally higher during the PHS compared to the HS. This finding was rather unexpected as previous studies suggest that the PHS is characterized by a relatively higher level of household food insecurity (Hirvonen et al., 2015), the latter being expected to be associated with a more suboptimal energy and nutrient intake. However, we found that the number of meals given to the average child was similar during PHS and HS. As we did not collect household food insecurity indicators, it remains unclear if either our study area is not suffering from previously reported fluctuations in food insecurity, or if household food insecurity during the PHS does not trickle down to the child’s energy and nutrient intakes.

In the present study, a DDS ≤2 food groups was a good proxy for “low" micronutrient density adequacy. This finding is in good agreement with a previous analysis from a ten-country study (Working Group on IYCF Indicators, 2006) and was subsequently proposed by others (Moursi et al., 2008a). Increasing the cut-off of DDS increased sensitivity at the expense of specificity, and higher percent misclassification. In a country like Ethiopia with high rates of micronutrient malnutrition, the trade-off for higher sensitivity could be more important in order to screen children at risk of micronutrient deficiencies, and include them in nutrition intervention programmes, such as targeted supplementation or promotion of appropriate infant and young child feeding. However, with limited preventive nutrition programmes, one should not compromise specificity as more sensitivity implies more false positives. Hence, the trade-off between sensitivity and specificity depends on the desired purpose of the DDS. Based on a series of analyses of datasets from developing countries, WHO developed indicators for assessment of infant and young child feeding practices in which consumption of 4 or more food groups was considered to be associated with a good quality diet(WHO, 2008). Nevertheless, the utility of DDS to classify children with "higher" intake was not ascertained in our analysis. This was mainly due to the observation that only 2% and none of the infants 6-8 months age had achieved MMDA ≥75% in HS and PHS respectively. The overall proportions of study infants who were correctly classified with "low" or "higher" intake by DDS were also relatively low compared to previous analyses (Moursi et al., 2008a, Working Group on IYCF Indicators, 2006) in which larger samples were used to validate DDS with 1g and 10g minimum limits.

This study has a number of strengths and limitations to consider. In the valuation of the performance of DDS as an indicator of child dietary quality expressed in terms of MMDA, we ran regression analyses by taking into account all measured covariates. In contrast to previous studies, this study also used two 24 hour recalls to collect data on quantitative nutrient intake. This offers a better estimation of the usual intake distribution at the population level. We were limited to compile MMDA based on data availability for only eight micronutrients in Ethiopia. However, most of the micronutrients of public health importance were included. We did not estimate individual breast milk intake, but rather we used the age-adjusted mean intake as proposed by the WHO guideline (WHO, 1998). For that reason, we opted to convert the intakes into MDAs that are inversely associated with the caloric contribution from breast milk.

Conclusions

We found a positive association between dietary diversity and micronutrient density adequacy and energy intake from complementary foods and this association remained stable over seasons. This exercise once more corroborates previous findings that low dietary diversity scores indicate a qualitatively poor diet in young children. In areas where there are limited resources, the dietary diversity score can give a rough idea on micronutrient intake, irrespective of the agricultural seasons which influence food availability over the year. For more detailed information on prevalence and type of micronutrient deficiency, more rigorous approaches should be used.

5

Chapter 5: Feeding practices and growth among young children during two seasons in rural Ethiopia

Redrafted from: Wondafrash, M., L. Huybregts, C. Lachat, K. P. Bouckaert and P. Kolsteren (2017). "Feeding practices and growth among young children during two seasons in rural Ethiopia." BMC Nutrition **3**(1): 39.

5.1. Abstract

Background: The use of indices of infant and young child feeding practices to predict growth has generated inconsistent results, possibly through age and seasonal confounding. The aim of this study was to evaluate the association of a dietary diversity score (DDS) and infant and child feeding index (ICFI) with growth among young children in a repeated cross-sectional and a follow-up study in two distinct seasons in rural southwest Ethiopia.

Methods: We used a repeated cross-sectional design comparing child feeding practices to nutritional status in 6-12 month old childrenduring harvest (HS; n=320) and pre-harvest season (PHS; n=312). In addition, 6-12 month old children from the HS were reassessed 6 months later during PHS. In addition to child anthropometry, child feeding practices were collected using 24-hour and 7-day dietary recalls.

Result: The mean (±SD) length-for-age z-score (LAZ) of the 6-12 month old children was -0.77(±1.4) and -1.0 (±1.3) in HS and PHS, respectively, while the mean (±SD) of the follow-up children in PHS was -1.0 (±1.3). The median DDS (IQR) was 2.0 (1.0, 3.0.), 2.0 (2.0, 3.0) and 3.0 (2.0, 4.0) for the children in HS, PHS and the follow-up children in PHS, respectively. The DDS in HS was positively associated with LAZ at follow-up (β= 0.16; 95%CI: 0.01, 0.30; *P*=0.03) after controlling for confounding factors. ICFI and DDS were not associated with mean LAZ, weight-for-height z-score and weight-for-age z-score within season. However, the odds of being stunted when having a DDS≤2 was 2.3 times (95%CI: 1.10, 4.78; *P*=0.03) higher compared to a DDS>2 child in HS and 1.7 times (95%CI: 1.04, 2.71; *P*=0.04) higher for the pooled sample of 6-12 months old children in HS and PHS.

Conclusions:

The DDS was found to be an indicator for child stunting during the Ethiopian harvest season. The DDS can be an appropriate tool to evaluate the association of child feeding practices with child growth irrespective of season. Inclusion of other dimensions in the construction of ICFI should be considered in future analysis as we found no association with growth.

5.2. Introduction

Child undernutrition is persistent in low income countries (Bhutta et al., 2013, Ahmed et al., 2012, Lutter et al., 2011). Ethiopia has one of the highest rates of childhood stunting despite a reduction since 2000 (CSA [Ethiopia] and ORC Macro, 2011). Among others, suboptimal child feeding practices are important underlying determinants of global poor child growth (UNICEF, 2012). Of the children who are breastfed, only a third of children 0-6 and 6-23 months old are exclusively breastfed and receive complementary food respectively (CSA [Ethiopia] and ORC Macro, 2011). Moreover, only 4% of children 6-23 months old are fed as per the global recommendations.

The construction of summary child feeding indices to assess nutritional outcomes has gained momentum since the early 2000's. The infant and child feeding index (ICFI) was developed based on breast feeding, bottle feeding, feeding frequency and dietary diversity in the previous 24h and consumption of food groups in the previous 7 days. It has been suggested that infant and child feeding practices tend to cluster in which earlier good practices by a caregiver are more likely to continue later with more awareness on appropriate feeding behaviors (Arimond and Ruel, 2003b). Hence, data on child feeding practices collected over a short period of time were reported to indicate longer term health and nutritional outcomes. On the other hand, a previous review questioned the strength of several existing dietary quality indices in terms of their diagnostic capacity of various health outcomes (Kourlaba and Panagiotakos, 2009). Demographic and Health Survey (DHS) data have shown that dietary diversity alone was also associated with height -for-age z-score (HAZ) in 10 out of the 11 countries analyzed (Arimond and Ruel, 2004). Nevertheless, the associations of infant and child feeding practices with HAZ were not consistent across geographical locations and age categories. The majority of studies showed associations for children aged 12 months and older and not for children in their first year of life. The arbitrary selection of components and cutoffs, as well as determining the contribution of components to the overall index have been highlighted as limitations to predict specific health outcomes or nutritional status (Kant, 2004).

Summary indices that include more information on feeding practices as a whole, in contrast to the diet quality itself, were reported to predict nutritional status (Arimond and Ruel, 2003b). Several evaluations have shown a positive association of such summary indices of child feeding practices with linear growth, i.e. height/length-for-age z-score (HAZ/LAZ) or stunting (HAZ/LAZ <-2) (Ruel and Menon, 2002, Zhang et al., 2009, Bork et al., 2012, Moursi et al., 2009, Sawadogo et al., 2006). Conversely, the associations between child growth and individual components of the summary feeding index, i.e. breast feeding or bottle feeding alone, were found to be rather incoherent.

Adding to the complexity of associations between dietary quality or feeding practices indicators and child nutritional status are the profound seasonal effects in Sub-Saharan Africa on food availability and accessibility. To our knowledge, no study has examined the impact of seasonality on these associations. Previously, we found an association of DDS with mean micronutrient density adequacy (MMDA) and the association proved to be stable over two distinct seasons in southwest Ethiopia (Wondafrash et al., 2016). Moreover, having a DDS≤2 was associated with an inadequate intake of micronutrients. The aim of this study was to assess the association of ICFI and DDS with growth among infants and young children. As a first step, we analyzed the associations between ICFI and DDS with child growth using repeated cross-sectional data from two distinct seasons to assess the impact of seasonality on the strength of the association in children 6-12 months of age. Secondly, we assessed the association of ICFI and DDS with child growth longitudinally using follow-up data.

5.3. Methods and subjects

Study setting

The study was conducted in nine kebeles (*smaller government unit*) of the catchment area of Gilgel Gibe Field Research Center of Jimma University, southwest Ethiopia. The Research Center was established in 2005 to serve as one of the Demographic Surveillance Systems in Ethiopia and field epidemiology attachment site of Jimma University. According to the census conducted in 2008, the total population within the area was 48,316 of which 49.4% were males with an average household size of 4.7 (SD=2.2). One third of the households were urban (Institute of Health Science Research (Jimma University), October, 2011). The main source of income for the community is farming and raising livestock. Maize, sorghum and teff (a species of *Eragrostis* native to Ethiopia) are commonly grown food crops in the area. Food consumption patterns of the population are expected to be different based on the differences in food availability in the two seasons, i.e. harvest (HS) and pre-harvest (PHS) season.

Sampling and design

We combined two study designs. Firstly, a repeated cross-sectional survey was conducted using a census of all children aged 6-12 months within the study area during HS, from October to December 2009, and PHS, from June to August 2010(IHSR (Jimma University), 2011). Children aged 6-12 months old and born in the area, who have a biological mother or female caregiver, presenting no serious illnesses which warrant immediate medical attention or referral (e.g. rapid breathing, nasal flaring, reported high fever, abnormal body movement, persistent diarrhea (≥2 weeks) or vomiting, etc.) or malformations affecting anthropometric measurement, and no intention of leaving the area in the coming 6 months were invited to participate. A total of 320 and 312 children were eligible in the HS and PHS, respectively.

Secondly, we conducted a follow-up or cohort study of children who participated in the cross-sectional survey in HS. The follow-up was done 6 months later, i.e. in PHS from June to August 2010 **(Figure 5.1).**

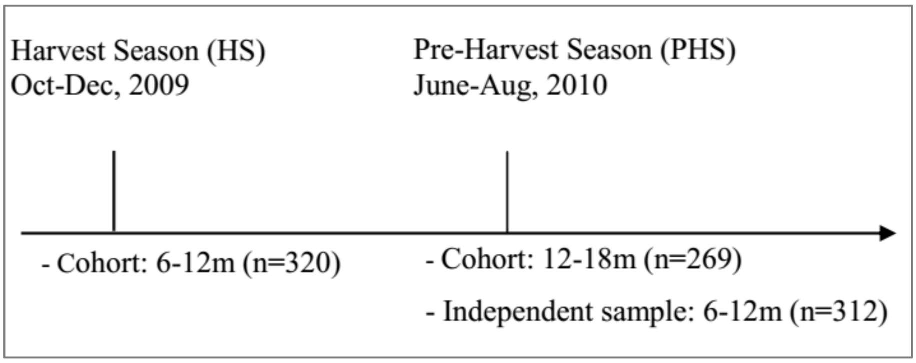

Figure 5.1: Study design and age groups of different samples

Data collection

We used pre-tested interviewer-administered questionnaires to collect data on socio-economic status, demographics, and infant and young child feeding practices. Questions were adapted from the Ethiopian Demographic and Health Survey (EDHS) questionnaires (CSA [Ethiopia] and ORC Macro, 2006) and the World Health Organization (WHO) guideline for the assessment of infant and young child feeding (WHO, 2007b). Data collectors with previous experience of data collection in the research center and able to fluently speak the local languages (Amharic and Afan Oromo) were recruited and received a five-day intensive training from the principal investigators.

Dietary diversity score

Dietary diversity was measured based on the method suggested by Dewey *et al* (Dewey et al., 2006). The DDS was obtained by summing the number of food groups consumed in the previous 24 hours by the infant. The following seven food groups were included: a) Grains, roots and tubers; b) legumes and nuts; c) dairy products; d) flesh foods; e) eggs; f) vitamin A-rich fruits and vegetables (>130 retinol equivalents/100g); g) Other fruits and vegetables. Dietary diversity score was used in our analyses as it is or dichotomized. Previous analyses showed that a DDS ≤2 is associated with a low micronutrient intake (Dewey et al., 2006, Moursi et al., 2008b, Wondafrash et al., 2016) which warrants the investigation of its association with growth of infants and young children.

Construction of the infant and child feeding index

The ICFI was calculated using core indicators as described previously (Arimond and Ruel, 2002b, Moursi et al., 2008b). Dietary diversity was assessed using the method described above and age-specific scores were assigned **(Table 5.1).** As there was no information on energy density of complementary foods or breast milk intake, the scoring given to feeding frequency is based on the previous recommendation (PAHO/WHO 2003) which assumes low energy density and average breast milk intake. A score of +1 was attributed for those who met the lower end of the recommendation and +2 for those who exceeded the recommendation. Food group frequency was assessed using a questionnaire developed specifically for this purpose in which we asked the number of days a specific food group was consumed. Frequency was then scored 0 if not consumed during the previous week, +1 if consumed on 1-3 days and +2 if consumed for ≥4 days. These scores were summed up yielding a possible range of 0-14. Subsequently, new scores were assigned depending on age category as shown in **Table 5.1.** Each component of ICFI was defined and scored according to the current age-specific infant and young child feeding recommendations (WHO, 2010b). The ICFI was calculated by adding up all the age-specific scores. The index was cut into three categories in which a score of 0-5 was considered low, 6-7 as medium and 8-9 as high, as described previously by

Moursi et al (Moursi et al., 2009). The index was also treated as a continuous variable in our analyses.

Table 5.1: Scoring system used to create the infant and child feeding index for children aged 6-12 months[a]

Components	Age-specific scores	
	6-8 months	**9-12 months**
Breast feeding	Yes =2 No=0	Yes =2 No=0
Bottle feeding	Yes =0 No=1	Yes =0 No=1
Dietary diversity (24 hours) [b]	0-1 = 0 2=1 3+ = 2	0-2 = 0 3=1 4+ = 2
Food group frequency (previous 7 days)[c]	0-2 = 0 3-4=1 5+ = 2	0-3 = 0 4-5=1 6+= 2
Feeding frequency[d]	0-1 = 0 2=1 3+ = 2	0-2 = 0 3=1 4+ = 2

[a]The the scoring for the construction of ICFI was adapted from (Moursi et al., 2009)

[b]Scores for dietary diversity was attributed in a manner that reflects the age-specific distributions. Dietary diversity was calculated by summing the number of food groups consumed in the 24-h period.

[c]For food group frequency, each food group was scored 0 if not consumed during the previous week, +1 if consumed on 1–3 days, and +2 if consumed on 4 days or more. These scores were summed to give a possible range of 0–14 and then new scores were assigned to reflect age-specific distribution

[d]A score 0, +1 or +2 was attributed for those who did not meet the lower end, met the lower end and those who exceeded the lower end of recommendation respectively.

Anthropometry

Anthropometric measurements were standardized through repetitive exercises. Recumbent length was measured on an infant measuring board and recorded to the nearest 0.1cm (SECA 210, Hamburg, Germany). Child's weight was measured naked or with very light clothes using mother-child digital scales and recorded to the nearest 0.1kg (SECA Uniscale, Hamburg, Germany). Age was recorded from birth certificates or immunization cards as much as possible. If reliable documents for age estimation were absent, local events calendars were used to help the mother or caregiver estimate the approximate age of the child.Accordingly, 70% of the ages were estimated by local event calander whereas the remainng 30% were obtained either from immunization cards or birth cirtificates. Most of the anthropometric data collectors in HS were also available in PHS.

Household socio-economic status

We constructed an asset-based proxy to estimate the socio-economic status (SES) of the included households. Items included were ownership of cattle, house construction materials, land (homestead, land under cultivation, fallow land) and ownership of other household assets. For household assets, categories for each item were 'owned' or 'not owned' by the household. The first principal component (56% of the variance) from a principal components analysis was divided in tertiles and used as a proxy for SES (Vyas and Kumaranayake, 2006).

Morbidity

Morbidity was assessed qualitatively by asking the mother about the presence of diarrhea (defined as ≥3 episodes of loose stool per day), fever and cough (yes/no) in the previous two weeks of the survey.

Data analysis

The anthropometric measures were converted into z-scores of weight-for-age (WAZ), length-for-age (LAZ), and weight-for-length (WLZ) based on the WHO 2006 Child Growth Standards (WHO, 2010c). Underweight, wasting and stunting were defined as WAZ, WLZ and LAZ <-2 z-scores from the median of the standard, respectively (WHO, 2006). Descriptive analyses were conducted for SES and demographic data, morbidity, maternal schooling, household characteristics, nutritional status, dietary diversity and feeding practices of children aged 6-12 months for the repeated cross-sectional study. Demographic data, morbidity, nutritional status and dietary diversity were described separately for the follow-up children aged 12-18 months.

Means (±SD) were reported for continuous data which were normally distributed, otherwise the median (IQR) was used. Differences in means were statistically tested using a student t-test for normally distributed outcomes or Wilcoxon-Mann-Whitney test for skewed outcomes, while a χ^2-square test was used to compare categorical variables. The DDS was used as a standalone outcome and as a component of ICFI in the analyses. We used multiple linear regression models to analyze the association between DDS and ICFI with LAZ, WLZ and WAZ adjusted for SES and demographic data, morbidity status, maternal schooling of the 6-12 month old children in the repeated cross-sectional studies and follow-up children. Multiple logistic regressions were used to model DDS and ICFI with binary categories of LAZ, WLZ and WAZ (cut-off value: -2 SD). Bivariate analyses were performed between all predictor variables and anthropometric outcomes and then every variable with a modest association (P<0.25) was a candidate to be included in the multivariate analyses. A full model assessing the relation between DDS, ICFI and components of ICFI was compiled after which non-significant variables were removed by a backward procedure using a likelihood ratio test (P<0.05*).*

The internal consistency of the ICFI was checked by using both the Fisher exact test of association of ICFI (categorized) with its components being bottle feeding, food group frequency and feeding frequency, and by the Cronbach α coefficient.

As there was no effect of season on the association of DDS and ICFI and child growth or nutritional status, the data of the two cross-sectional studies were pooled and the same analysis was performed to increase power adjusting for season and the abovementioned confounding variables. As the interaction of DDS or ICFI with season was not significant (P >0.1), the interaction term was not included in the models. Moreover, a DDS cutoff of ≤2 was used to dichotomize DDS based on a prior sensitivity and specificity analysis (Wondafrash et al., 2016) and its association with child anthropometry was tested.

ANCOVA was used to assess the longitudinal relationship of anthropometric indices with DDS and ICFI in the follow-up children (Twisk, 2003). For this purpose anthropometric indices at follow-up (PHS) were used as outcome variables and anthropometric indices at baseline (HS) as predictor variables. The DDS and ICFI at

HS were also entered separately in the models as predictor variables. Data was analyzed with Stata 13.0 (StataCorp, College Station, TX, USA). Statistical significance was set at 5% and all tests were two-sided.

Ethical considerations

The study was approved by the Ethics Review Board of Jimma University, Ethiopia (RPGC/104/2002) and the Ethics Committee of Ghent University Hospital, Belgium (B67020109188). Informed consent was secured from the mothers/caretakers before conducting the interviews and assessment.

5.4. Results

In the repeated cross-sectional surveys in HS and PHS, data was collected from 320 and 312 children aged 6-12 months respectively. In total, 24 children in HS and 21 children in PHS who had incomplete complementary food intake data and/or information pertaining to breast feeding (HS=21; PHS =15) or uncertain age (HS =3; PHS =6) were omitted from analysis. The mean LAZ, WAZ and WLZ of excluded observations did not differ statistically from the rest of the observations in both seasons.

Of the 296 children with complete data at HS, 27 (9%) children could not be traced after repeated visits at follow-up. Three observations from the follow-up children were excluded from analysis as they did not have complete dietary diversity data.

Nutritional status

In the repeated cross-sectional surveys in HS and PHS, 6-12 month old children had a stunting prevalence of 18.6% and 21.1% respectively, and a wasting prevalence of 11.8% and 15.5% respectively. The mean LAZ was -0.77±1.4 and -1.0 ±1.3 for the children in HS and PHS respectively**.** At follow-up, 21% of 12-18 month old children were stunted and 20% were wasted, whereas the mean LAZ was -1.0±1.3 (**Table 5.2**).

Child feeding practices

All children aged 6-12 months were partially breastfed at the time of the surveys. The median (IQR) recalled duration of exclusive breastfeeding was 4.0 (3.0, 4.0) and 4.0 (3.0, 6.0) months in HS and PHS respectively; while 19% and 11% of these children were bottle-fed during HS and PHS respectively. Eighty-eight and sixty seven percent (*P*<0.05) of the children were introduced to complementary foods before 6 months of age in HS and PHS respectively. There was no significant difference in DDS (*P*=0.17) and ICFI (*P*=0.09) between the two seasons (*P*=0.17) for children aged 6-12 months (**Table 5.2).**

The follow-up data showed that breastfeeding was continued well into the second year of life. Only 4.8% of the follow-up children were not breastfed. The median DDS (IQR) was 3.0 (2.0, 4.0) (**Table 5.2).**

Table 5.2: Characteristics of 6-12 month old children in the repeated cross-sectional and follow-up study in HS and PHS

Variables	HS (n=296)	PHS (n=291)	P-value ¥	Follow- up at PHS
Gender of child				
Female	154 (52.0)	125 (43.0)	0.03	137 (51.5)
Age of the child (6-12)				-
6-8 months	152 (51.4)	71 (24.4)	<0.001	
9-12 months	144 (48.6)	220 (75.6)		
Age of the child (12-18)				
12-15 months				183 (68.8)
16-18 months				83 (31.2)
Age of the mother				-
<20 years	60 (20.6)	32 (11.0)	<0.01	
20-34 years	202 (69.4)	233 (80.1)		
≥35 years	29 (10.0)	26 (8.9)		
Socioeconomic status, n(%)				-
Low	79 (27.3)	115 (39.9)	0.01	
Medium	106 (36.7)	89 (30.9)		
High	104 (36.0)	84 (29.2)		
Under-five children in the HH, n(%)				-
0-1	79 (26.7)	66 (22.7)	0.26	
≥2	217 (73.3)	225 (77.3)		
Morbidity, n(%)				
Cough*	44 (14.9)	57 (19.6)	0.13	36 (13.5)
Fever*	57 (19.3)	67 (23.0)	0.27	53 (19.9)
Diarrhea*	49 (16.7)	70 (24.1)	0.03	67 (25.2)
Source of drinking water for HH, n(%)				-
Unsafe water source	252 (85.1)	229 (78.7)	0.04	
Human waste disposal by the HH, n(%)				
Improper disposal	117 (39.7)	97 (33.3)	0.11	
Maternal schooling, n(%)				-
No formal schooling	243 (82.1)	246 (84.5)	0.43	
LAZ, mean ± SD	-0.77 ± 1.4	-1.0 ± 1.3	0.02	-1.0 ± 1.3
Stunting, n(%)				
LAZ <-2	55 (18.6)	61 (21.1)	0.44	55 (20.7)
LAZ<-3	18 (6.1)	16 (5.5)	0.78	16 (6.0)
WLZ, mean ± SD	-0.63 ± 1.3	-0.75 ± 1.3	0.28	-0.81 ± 1.2
Wasting, n(%)				
WLZ <-2	35 (11.8)	45 (15.5)	0.19	51 (19.2)
WLZ<-3	12 (4.1)	12 (4.1)	0.96	11 (4.1)
WAZ, mean ± SD	-0.96 ± 1.2	-1.1 ± 1.4	0.08	-1.1 ± 1.1
Underweight, n(%)				
WAZ <-2	52 (17.6)	75 (25.8)	0.02	49 (18.4)
WAZ<-3	18 (6.1)	20 (6.9)		13 (4.9)
Current breast feeding				
Yes	296(100)	291(100)	-	253 (95.1)
DDS, median (IQR)	2.0 (1.0, 3.0)	2.0 (2.0, 3.0)	0.17	3.0 (2.0, 4.0)
ICFI, median (IQR)	5.0 (5.0, 7.0)	6.0 (5.0, 7.0)	0.09	-

*HS= harvest season; PHS = pre-harvest season; HH= household; LAZ = length-for-age z-score; WLZ= weight-for-length z-score; WAZ= weight-for-age z-score; DDS= Dietary diversity score; ICFI= Infant and Child Feeding Index; *Diarrhea, cough, fever in the previous two weeks of the studies; ¥ P-values are calculated for the difference of the two independent samples at HS and PHS.*

Internal consistency of the components of ICFI for the cross-sectional study

The ICFI was positively associated with dietary diversity, food group frequency and feeding frequency and bottle feeding (*P*<0.001) in the repeated cross-sectional study but not with breastfeeding as all were breastfed. However, the overall internal consistency estimated by the Cronbach-α coefficient was low (α=0.45 in HS and α=0.47 in PHS), but slightly better in the age group of 6-8 months in PHS (α=0.58).

Association between feeding practices and child nutrition

Repeated cross-sectional study

The ICFI and DDS were not associated with LAZ and WHZ among 6-12 month old children in both HS and PHS.After each component of the ICFI was assessed separately for its association with LAZ and WHZ, only bottle-feeding was associated with LAZ in PHS (β= -0.74, 95%CI: -1.20, -0.28; *P*=0.002) in favor of those who were bottle fed **(Table 5.3)**. Children with a DDS≤2 account for 68% and 63% of all children in HS and PHS respectively. The odds of being stunted for children with a DDS≤2 was 2.3 (95%CI: 1.1, 4.8; *P*=0.03) times higher compared to their peers with DDS>2 during the HS.

In view of the absence of an important seasonal impact on the association between DDS, ICFI and child nutrition, we pooled the data of 6-12 month old children from the two seasons to increase power when testing the association of DDS and ICFI with nutritional status of children. We observed that the odds of being stunted were 1.70 (95%CI: 1.04, 2.71; *P*=0.04) times higher among children with a DDS≤2 compared to those with a DDS>2. Moreover, being 9-12 months old [OR=2.0; 95%CI: 1.24, 3.18; *P*<0.01], having diarrhea [OR=1.70; 95%CI: 1.05, 2.78; *P*=0.04] and the presence of ≥2 under five children in the same household [OR =2.3; 95%CI: 1.24, 4.46; *P*=0.01] were significantly associated with stunting **(Table 5.4)**.

Follow-up study

The DDS of the 6-12 months old children during HS was positively associated with LAZ at follow-up period during PHS (β= 0.16; 95%CI: 0.01, 0.30; *P*=0.03) **(Table 5.5).** The occurrence of diarrhea in the same season was negatively associated with LAZ (β= -0.35; 95%CI: -0.67, -0.03; *P*=0.03). However, the DDS was not associated with LAZ and WHZ within seasons for the follow-up children.

Table 5.3: Associations of ICFI and its components with LAZ and WLZ of children in the repeated cross-sectional study§

Variables	Harvest season (n=296)					Preharvest season (n=291)				
	n	Mean LAZ (SD)	P-value¥	Mean WLZ (SD)	P-value¥	n	Mean LAZ (SD)	P-value¥	Mean WLZ (SD)	P-value¥
Tertile of ICFI										
Low (reference)	152	-0.86 (1.41)		-0.66(1.26)		132	-1.04(1.23)		-0.78(1.42)	
Medium	97	-0.66 (1.43)	0.63	-0.59(1.30)	0.39	119	-1.20(1.48)	0.41	-0.70(1.36)	0.21
High	47	-0.74 (1.35)	0.78	-0.63(1.26)	0.50	40	-0.60(1.25)	0.40	-0.80(0.99)	0.96
Dietary diversity (24h)*										
Low (reference)	140	-0.77(1.47)		-0.65(1.25)		157	-1.13(1.28)		-0.75(1.34)	
Medium	99	-0.81(1.38)	0.17	-0.62(1.37)	0.50	75	-1.18(1.51)	0.72	-0.87(1.36)	0.74
High	57	-0.66 (1.31)	0.60	-0.63(1.13)	0.42	59	-0.64(1.18)	0.27	-0.61(1.33)	0.43
Food group frequency										
Low (reference)	62	-0.95(1.51)		-0.67(1.11)		34	-1.22(1.31)		-0.79(1.55)	
Medium	75	-0.74(1.52)	0.46	-0.84(1.39)	0.66	52	-1.09(1.41)	0.78	-0.74(1.30)	0.93
High	159	-0.72(1.31)	0.28	-0.52(1.26)	0.52	205	-1.01(1.31)	0.24	-0.74(1.32)	0.96
Bottle feeding*†										
Yes (reference)	56	-0.75(1.43)		-0.68(1.29)		32	-0.21(1.22)		-0.25(1.15)	
No	240	-0.78(1.41)	0.54	-0.62(1.26)	0.39	259	-1.15(1.31)	0.002	-0.80(1.35)	0.15
Feeding frequency*										
Low (reference)	147	-0.88(1.40)		-0.67(1.30)		158	-1.09(1.21)		-0.85(1.41)	
Medium	63	-0.55(1.24)	0.18	-0.36(1.19)	0.05	54	-1.12(1.47)	0.44	-0.48(1.51)	0.07
High	86	-0.76(1.53)	0.92	-0.78(1.25)	0.87	79	-0.91(1.50)	0.79	-0.74(1.03)	0.61

ICFI= Infant and child feeding index;LAZ = length-for-age z-score; WLZ= weight-for-length z-score;

The low, medium and high categories are based on the scoring given for the each component of infant and child feeding index as 0, +1 and +2 respectively as described in* ***Table 5.1*

¥P-values are from multiple linear regression models controlling for confounding variables using backward stepwise selection. The association of individual components and LAZ and WLZ were modeled separately.

§All analyses are adjusted for child and maternal characteristics and household socioeconomic status in the multivariate regression model.

Table 5.4: Predictors of stunting in the pooled sample of all 6-12 months old children participating in the repeated cross-sectional study*

Variables	n†(%)	Odds Ratio	95% CI	P-value
DDS				
DDS>2	204 (34.8)	Reference		
DDS≤2	383 (65.2)	1.7	1.04,	0.04
Season				
HS	296 (50.4)	Reference		
PHS	291 (49.6)	1.0	0.64,	0.91
Human waste disposal				
Improper disposal	214 (36.5)	Reference		
Proper disposal	373 (63.5)	0.7	0.42,	0.05
Maternal schooling				
No formal schooling	489 (83.3)	Reference		
Attended formal school	98 (16.7)	0.5	0.25,	0.07
Age of the child				
6-8 months	223 (38.0)	Reference		
9-12 months	364 (62.0)	2.0	1.24,	<0.01
Age of the mother				
<20 years	92 (15.8)	Reference		
20-34 years	435 (74.7)	0.6	0.36,	0.03
≥35 years	55 (9.5)	0.7	0.27,	0.35
Diarrhea[¥]				
No diarrhea	466 (79.7)	Reference		
Diarrhea present	119 (20.3)	1.7	1.05,	0.04
Under-five children in the				
0-1	145 (24.7)	Reference		
≥ 2	442 (75.3)	2.3	1.24,	0.01

*DDS= dietary diversity score; HS = harvest season; PHS= pre-harvest season; HH= household. *Results from multiple binary logistic regression models using a backward stepwise selection of predictors. [¥]Diarrhea in the previous two weeks of the studies. † Total numbers included in the analysis. Due to missing observations, the numbers for predicator variables “Age of the mother” and “Diarrhea” do not add up to 587.*

Table 5.5: Multiple regression assessing the association between DDS and Length-for-Age Z-score adjusting for relevant confounding factors among the follow-up children in PHS*****

Variables	n†(%)	β	95% CI	P-value
DDS during HS	266	0.16	0.01, 0.30	0.03
HAZ during HS	266	0.33	0.23, 0.43	<0.001
Age of the mother				
< 20 years	54 (21.1)	Reference		
20-34 years	181 (68.3)	0.43	0.01, 0.80	0.02
≥35 years	27 (10.6)	0.66	0.04, 0.13	0.01
Diarrhea¥				
No diarrhea	199 (74.8)	Reference		
Diarrhea Present	67(25.2)	-0.35	-0.67, -0.03	0.03
Maternal schooling				
No formal schooling	217 (81.6)	Reference		
Attended formal school	49 (18.4)	0.33	-0.05, 0.71	0.05
Socio-economic status				
Low	71 (26.9)	Reference		
Medium	94 (35.6)	0.41	0.06, 0.77	0.04
High	99 (37.5)	0.25	-0.10, 0.60	0.24

DDS =Dietary diversity score; HAZ: height-for-age z-score; HS =harvest season;
**Analysis of covariance was performed to determine predictors of growth at follow-up. ¥Diarrhea in the previous two weeks of the follow up study. †Total numbers included in the analysis. Due to missing observations, the numbers for predicator variables "Age of the mother" and "Socioeconomic status" do not add up to 266.*

5.5. Discussion

The dietary diversity score during the HS was found to be an independent predictor of the subsequent 6 months of linear growth. Stunting was found to be associated with low DDS (≤2) only during HS. Moreover, we did not find such an association for ICFI. Within seasons, we did not find cross-sectional associations between DDS and ICFI and mean LAZ, WHZ and WAZ.

We previously found that children who consumed ≤2 food groups were likely to have inadequate micronutrient intake (Wondafrash et al., 2016). In the present analysis, the odds of being stunted was significantly higher for those who consumed ≤2 food groups in the previous 24 hours. This was observed both among children in the HS and in the pooled data. Previous analyses also showed that DDS of ≤2 was an indicator of a low nutrient dense diet (Dewey et al., 2006, Moursi et al., 2008a). An evaluation of the association of DDS with child anthropometry was not performed taking a DDS of ≤2 as a cutoff in those previous analyses. In one study in Mali, children in urban households with lowest DDS had higher risk of stunting and underweight (Hatløy et al., 2000). Ruel and Arimond used tertiles of dietary diversity and the association with HAZ was observed in many of the countries for which the analyses was performed using DHS data (Arimond and Ruel, 2004). Furthermore, it was observed that significant proportions of children from the African countries included in the analysis were consuming ≤2 food groups. A Kenyan study found an association between dietary diversity, calculated from three days average consumption of unique food groups, and five anthropometric indices. However, contrary to the present study, the samples of these previous studies were heterogeneous in terms of age groups and breast feeding status, which might explain the inconsistent results of associations between DDS and growth(Onyango, 2003). Younger children are more dependent on breast milk and their complementary food lack diversity compared to older children (Onyango et al., 2002).

We did not observe a significant association between ICFI and LAZ after adjusting for confounders. This was also reported previously by studies conducted in rural West Africa (Ntab et al., 2005) and China (Zhang et al., 2009). This finding, however, is in contrast to many other studies which showed significant associations of ICFI with LAZ (Ruel and Menon, 2002, Moursi et al., 2009, Sawadogo et al., 2006, Bork et al., 2012, Ma et al., 2012). Prior to the adoption of the WHO IYCF measurement indicators, the grouping of food items was done differently apart from differences in the number of food groups used to construct the DDS (Marriott et al., 2012, Jones et al., 2014), a component of ICFI. The numbers of components used to construct ICFI were also not uniform over different studies. For instance, we used five components whereas Sawadogo et al (Sawadogo et al., 2006), Ntab et al (Ntab et al., 2005) and Zhang et al (Zhang et al., 2009) used six, seven and eight components to construct the feeding index, respectively. Ntab et al. used an age group of 12-42 months for the analysis of the association between the ICFI and LAZ (Ntab et al., 2005). Moreover, dietary diversity is an important component in determining the relationship with child anthropometry when summary feeding measures are used (Ruel and Menon, 2002, Moursi et al., 2008b, Sawadogo et al., 2006). The majority of the children (67%) in the present study consumed few food groups which in turn can affect ICFI's discriminatory power and hence its association with LAZ (Disha et al., 2012). All children in this study were breastfed and were given a high score for the breast feeding component in the construction of the ICFI. The latter scenario might have diluted the association of the ICFI with LAZ in comparison to a setting with a mixture of breastfed and non-breastfed children. Moreover, other studies have shown that breast feeding continues for an extended period of time in sick or malnourished children (Marquis et al., 1997a, Simondon and Simondon, 1998, Simondon et al., 2001a, Simondon et al., 2001b) who in turn were given a high score for the summary index.

There was no significant association between DDS and ICFI during the HS and PHS in 6-12 months old children participating in the repeated cross-sectional study. However, we found that DDS in HS was an independent predictor of linear growth during the 6 months of follow-up. This has also been observed by other researchers using a longitudinal ICFI (a summary indicator from repeated ICFIs over time) rather than an ICFI measured at the time of child anthropometry assessment. Such a composite ICFI was found to be associated with LAZ in children who were followed up for seven months (Moursi et al., 2008b). Another study in urban China showed that a longitudinal ICFI, but not dietary diversity, was associated with LAZ measured one year later (Ma et al., 2012). This might indicate that of the feeding practice indices constructed from a brief recall might provide useful information on feeding in a long term. DDS, rather than ICFI, was a predictor of growth for the follow-up children in the present study, but it is known that dietary diversity is an important factor in defining the relationship of ICFI with child anthropometry. Nevertheless, optimal growth is more of a cumulative effect of feeding practices involving several nutrients on top of other factors such as household food security, environmental hygiene and healthcare (Ruel and Menon, 2002, Arimond and Ruel, 2003b, Lutter et al., 2011, Black et al., 2008, Onyango, 2003, Jones et al., 2014).

Overall, child feeding practices in the present study are suboptimal compared to international recommendations with 88% and 67% of study children being introduced to complementary foods before the recommended age of 6 months during the HS and PHS respectively. Bottle feeding was relatively common and associated with LAZ in favor of bottle fed. On the contrary, premature introduction of complementary foods negatively affected linear growth in a longitudinal study among Vietnamese children (Hop et al., 2000). According to an Ethiopian national survey, non-breastfed children were more likely to be fed with solid and semi-solid foods compared with breastfed children (CSA [Ethiopia] and ORC Macro, 2011) but its association with growth was not elucidated. Breastfeeding for an extended period time was proposed to be associated with low HAZ in rural Senegal after mothers delayed weaning for stunted children (Simondon et al., 2001a, Simondon et al., 2001b). A stronger association was also found between dietary diversity and HAZ in non-breastfed children compared to breastfed peers (Marquis et al., 1997a, Arimond and Ruel, 2004). However, we were not able to demonstrate such associations as all of our children were breastfed.

This study has a number of strengths, but also several limitations that need to be addressed. First, the study was conducted in two distinct seasons in which intake of nutrients and LAZ scores significantly differed, which adds to the external validity of the overall analysis. Secondly, we followed children from the HS to another season to assess clustering of child feeding practices and its effect on growth in the long term. However, the internal validity of the components of the ICFI was found to be lower than what has been arbitrarily accepted. It is also impossible to rule out the day-to-day variability, random or reporting error in the assessment of child feeding practices which can result in misclassification and a reduced precision to detect an association (Arimond and Ruel, 2003b). Over-reporting of desirable feeding behaviors is a common occurrence in the measurement of feeding practices using recall techniques (Arimond and Ruel, 2003b). As it has been demonstrated previously (Moursi et al., 2008b, Ma et al., 2012), it could have been more appropriate to construct the ICFI from feeding practice data collected from the same subjects at different time points to evaluate the occurrence of clustering of feeding behaviors and their impact on growth or nutritional status in the long term.

In conclusion, DDS and ICFI are not unequivocally related to child growth. Whereas DDS during the HS was able to predict longer term growth and only very poor dietary diversity was found to be associated with child stunting during HS and in the pooled data. Our data suggests the use of a minimal DDS (≤2) rather than a mean DDS as an indicator for suboptimal child growth. We did not find any association between ICFI and child growth, which might suggest that there is a room to add other dimensions of appropriate practices of complementary feeding, such as responsiveness and hygienic preparation and storage of complementary foods.

6

Chapter 6: Effects of n-3 LC-PUFA supplementation through breast milk and processed supplementary food on growth and morbidity of children 6-24 monthsof age in rural Ethiopia

The chapter is based on an acceptedmanuscript as**:**Argaw A,**Wondafrash, M.,**Kolsteren, P., Bouckaert, P K., Lachat, L., Belachew, T., De Meulenaer, B., and Huybregts L; Effects of n-3 LC-PUFA supplementation through breast milk and supplementary food on growth and morbidity of children 6-24 months:a randomized controlled trial in rural Ethiopia (*accepted for publication at The American Journal of Clinical Nutrition*)

6.1. Abstract

Background: Recurrent infections and enteric enteropathy contribute to growth faltering in low-income countries. Long-chain omega-3 polyunsaturated fatty-acids (n-3 LC-PUFAs) in fish-oil may improve immune maturation, intestinal wall integrity and resistance to infections, and could therefore lead to growth benefits in young children who are at risk.

Objective: To evaluate whether n-3 LC-PUFAs supplementation through breast milk and/or supplementaryfood affects morbidity, systemic inflammation and growth of children 6-24 months in Southwest Ethiopia.

Methods: A 4-arm double-blind randomized controlled trial was conducted byincluding 360 mother-infant pairs. Infants were 6-12 months old at enrollment. Subjects were provided with n-3 LC-PUFAs rich fish-oil. Study arms were both the lactating mother and child receiving the fish oil intervention (MCI); only the lactating mother receiving intervention (MI); only the child receiving intervention (CI) and both mother and child receiving a placebo supplement or control (C). Either member of the mother-child dyad not receiving the fish oil intervention received a placebo supplement. The primary study outcome was linear growth assessed by monthly changes in length-for-age z-score. Anthropometric measurements were taken monthly, and hemoglobin, C-reactive protein, and blood LC-PUFAs were measured at baseline and after 6 and 12 months of follow-up. Weekly morbidity surveillance was conducted throughout the study.

Results: Fish-oil supplementation resulted in significant increases in blood n-3 LC-PUFA concentration (P<0.01) and decreases in AA/DHA+EPA ratio (P<0.001) in all intervention arms. No significant intervention effect was found on linear growth, morbidity or systemic inflammation. Compared to the control group, significantly larger monthly changes in weight-for-length z-scores was found in the CI arm [effect size (95% CI): 0.026 (0.004, 0.048) per month; P=0.016)] and the MCI arm [(effect size (95% CI): 0.022 (0.000, 0.043) per month; P=0.051)].

Conclusions: N-3 LC-PUFA supplementation of lactating women and children did not affect linear growth and morbidity of children in a low-income setting. N-3 LC-PUFA supplementation given directly to children increased relative weight gain. The trial was registered at ClinicalTrials.gov as NCT01817634.

6.2. Introduction

Stunting occurs in more than a third of children in sub-Saharan Africa and accounts for 21% of disability adjusted life years of the region (Black et al., 2013, Black et al., 2008). Recurrent infections affect energy and nutrient intake and expenditure compromising child growth (Scrimshaw, 1989, Assis et al., 2005). Stunting is associated with environmental enteric dysfunction (EED), a chronic inflammation of the small intestine that impairs absorptive capacity and barrier functions with subsequent immunostimulation (Goto et al., 2009, Panter-Brick et al., 2008). The high prevalence of EED in children in low income countries may explain the lack of important impacts of various nutrition-specific interventions to prevent child stunting (McKay et al., 2010).

Long-chain omega 3 polyunsaturated fatty-acids (n-3 LC-PUFA) are known to modulate immune cell functions and inflammatory processes (Gottrand, 2008, Damsgaard et al., 2007, Anderson and Fritsche, 2002, Field et al., 2008). The modulation may lead to a more rapid maturation of the immune system, an improved integrity of the intestinal wall and an improved resistance to infectious diseases in young children. *In vitro* experiment has shown that n-3 LC-PUFAs increase intestinal barrier function by stimulating transepithelialresistance and reducing Interleukin-4 (IL-4)mediated permeability (Willemsen et al., 2008). In animal experiments, n-3 LC-PUFAsenhanced intestinal repair after severe malnutrition (López-Pedrosa et al., 1999) and chemically induced damage (Horie et al., 1998). Dietary LC-PUFA supplementation of formula-fed infants(Pastor et al., 2006, Lapillonne et al., 2014, Birch et al., 2010), older children(Minns et al., 2010, Thienprasert et al., 2009) and infants with prenatal maternal supplementation(Imhoff-Kunsch et al., 2011) led to clinical benefits in both inflammatory and infectious conditions. However, there is limited evidence on the impact of n-3 LC-PUFA supplementation on breastfed children in low-income settings characterized by very poor hygienic conditions and a high burden of infectious diseases.

The evidence on the impact of n-3 LC-PUFA supplementation on child growth is mainly based on studies in high-income populations (Makrides et al., 2005, Simmer et al., 2008, Delgado-Noguera et al., 2010). Only one study evaluated the efficacy of fish-oil supplementation to infants in a low-income country and found no notable effect on growth and morbidity (van der Merwe et al., 2013). However, the trial was conducted in infants with a relatively high intake of n-3 LC-PUFAs from breastmilk. Yet, a trial in

Ghana found that a lipid-based nutrient supplement (LNS) containing alpha-linolenic acid (ALA), the precursor of n-3 LC-PUFAs, had an effect on child linear growth which was attributed to the higher plasma n-3 PUFA level in the LNS group (Adu-Afarwuah et al., 2007). Observational studies in Congo and Brazil also showed that infant weight gain and linear growth was positively associated with breastmilk n-3 PUFA concentration (Rocquelin et al., 2003, Tinoco et al., 2009).In settings where the intake of n-3 LC-PUFA is low and chronic immunostimulation contributes to poor growth, the effects of an increased n-3 LC-PUFA intake on child growth could be warranted.

Breastmilk is the predominant source of n-3 LC-PUFAs for infants in developing countries (Michaelsen et al., 2011, Prentice and Paul, 2000). However, n-3 LC-PUFAs concentration is significantly influenced by maternal intake of marine foods which are unavailable to rural populations living far from coastal areas like in Ethiopia(Michaelsen et al., 2011, Brenna et al., 2007, Yuhas et al., 2006, Innis, 2007, Nishimura et al., 2013). In Ethiopia, the total dietary n-3 PUFAs supply is below the recommended level for infants and young children which poses further risk as children gradually transfer to complementary diet (Michaelsen et al., 2011).

This study answers to the call of the Lancet Series on maternal and child nutrition from 2013 (Bhutta et al., 2013) to provide more evidence on the health impacts of n-3 LC-PUFA in low-income countries. The study aims to test the efficacy of n-3 LC-PUFA supplementation on child physical growth and morbidity. We assessed two delivery strategies to supplement the child with additional n-3 LC-PUFA. First, by incorporating n-3 LC-PUFA rich fish-oil in a food supplement given directly to target children. Second, by supplementing the lactating mothers of the target children with fish-oil capsules. We hypothesize that n-3 LC-PUFA delivered through maternal and /or direct child supplementation improves child growth and reduces morbidity and systemic inflammation in a low-income setting in Ethiopia.

6.3. Methods and subjects

Participants

The trial was carried out from November 2013 to February 2015 in 3 districts of Jimma zone in Southwest Ethiopia located at ±300 km from the capital Addis Ababa. The area lies in a midland agro-climatic zone where subsistence farming is the predominant form of livelihood for the population. The main crops produced in the area are maize, sorghum and teff (a species of *Eragrostis* native to Ethiopia), and staple foods are very low in n-3 LC-PUFAs.

Study participants were identified from the Gilgel Gibe Health and Demographic Surveillance System (GGHDSS) that registers all births in the study area, and through an additional census conducted by the study team. Mothers with infants were invited to a nearby school in each of the 3 study districts to be screened for study eligibility. Mothers with a singleton infant of age 6-12 mowere eligible if the infant was not wasted (weight-for-length z-score (WLZ) ≥-2 SD and presented no bilateral pitting edema), was being breastfed, and the mother declared not having the intention to leave the study area for more than one month over the coming year. Exclusion criteria included infants or mothers with known chronic illness or medical treatment for this purpose, infants with a congenital abnormality that could affect feeding or growth, or with severe anemia (hemoglobin<7.0 g/dL), and children or mothers taking a nutritional supplement other than the ones provided by the project at enrolment or during the follow-up period.

Study design

This study was a randomized, double-blind, placebo-controlled trial involving child n-3 LC-PUFA supplementation through lactation (maternal intervention) and/or through supplementary feeding (child intervention). Mothers were randomly allocated to receive either intervention capsules containing fish-oil or placebo capsules of identical appearance containing corn-oil. Children of same mothers were randomly allocated to receive either an intervention food supplement containing fish-oil or the same food supplement without fish-oil as control. Thus, there were 4 study arms: both mother and child received fish-oil (MCI), mother received fish-oil but child received placebo (MI), child received fish-oil but mother received placebo (CI), and both mother and child received placebo (C).

A statistician not part of the research team generated a randomization scheme for the allocation of the maternal and child interventions. Each unique randomization number was randomly allocated to one of 6 letter codes for the maternal intervention (A, E, I, J, L, T) - three letters each for the intervention and the control group - using a computer program in permuted blocks. The randomization procedure was repeated for the child intervention by randomly allocating each randomization number to one of 6 letter codes representing the child intervention (B, K, P, V, Y, Z). The maternal capsules and the child food supplements were prepared in packages containing monthly supplies, and labeled with one of their corresponding letter codes by a person not involved in the study. Information regarding the assignment of the letter codes to the intervention and control supplements was sealed in an opaque envelope and sent to the study director (PK), who locked the envelope away until analysis was completed. Before the study started, two researchers (AA, MW) sealed each of the randomization numbers with the corresponding letter codes of the maternal and child interventions into separate opaque envelopes that were marked with unique participant numbers to be attributed at study inclusion. At study inclusion, a researcher opened the next sealed envelope in the presence of an eligible mother-child pair (in order of arrival), provided them with the first monthly dose of the maternal and child supplements corresponding to the letter codes and filled out the study participation card.

Interventions

The maternal intervention consisted of identical air tight gel capsules containing either fish oil (intervention) or corn oil (placebo). Both the intervention and placebo maternal supplements were provided by Biover NV (Belgium). The daily dose for intervention and control mothers consisted of 2 capsules. The two intervention capsules contained 500 mg n-3 LC-PUFAs (215 mg docosahexaenoic acid-DHA + 285 mg eicosapentaenoic acid-EPA). Each capsule additionally contained 5 mg of d-alpha-tocopherol, an antioxidant. Supplements were packaged in small non-transparent polystyrene containers containing a monthly supply of 60 capsules.

The child food supplements were produced by Michiels Fabrieken NV (Zulte, Belgium) and consisted of an extruded corn-soy blend fortified with 19 micronutrients. Both the intervention and control food supplements were designed to provide an energy top-up equivalent to half of the estimated average requirement for energy and fat and micronutrients at a dose of the recommended nutrient intake from complementary foods for an 11mo old infant (**Table 6.1**).Iron was added at a lower dose of 12.5 mg/d. The intervention food supplement was additionally enriched with microencapsulated fish-oil providing a daily dose of 500 mg n-3 LC-PUFAs (169 mg DHA + 331 mg EPA). The microencapsulation of the fish-oil reduced the fishy smell and taste, and minimized oxidation of n-3 LC-PUFAs. Monthly supplies of the food supplements were packaged using non-transparent bags with a protective aluminum coat and with nitrogen injected into the internal atmosphere to prevent oxidation.

Table6.1. Nutritional composition of daily rations (40 g) of food supplements given to study children[1]

Nutrient	Intervention supplement	Control supplement
Energy (kcal)	165	160
Energy (kJ)	693	671
Protein (g)	5.9	5.1
Carbohydrates (g)	24.4	26.1
Fat (g)	4.7	3.6
DHA (mg)	169	0.0
EPA (mg)	331	0.0
Calcium (mg)	421.8	412.4
Selenium (µg)	12.3	12.4
Magnesium (mg)	82.6	79.6
Zinc (mg)	8.8	8.4
Iron (mg)	12.7	12.3
Iodine (µg)	94.4	94.3
Vitamin C (mg)	30.7	29.8
Thiamine (mg)	5.7	6.1
Riboflavin (mg)	0.4	0.5
Niacin (mg NE)	5.5	5.0
Vitamin B6 (mg)	0.4	0.3
Pantothenic acid (mg)	1.9	1.8
Biotin (µg)	6.7	6.3
Vitamin B12 (µg)	0.7	0.7
Folate (µg DFE)	109.5	112.4
Vitamin A (µg RE)	447.0	458.2
Vitamin D (IU)	208.5	216.9
Vitamin E (mg α-TE)	4.2	4.5
Vitamin K (µg)	9.1	9.9

[1]Intervention supplement, corn-soy blend fortified with 19 micronutrients and fish-oil; control supplement, corn-soy blend fortified with multiple micronutrients and without fish-oil.
DFE, dietary folate equivalents; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; NE, niacin equivalents; RE, retinol equivalents; TE, tocopherol equivalents.

Study supplements were transported in cold-chain (6°C) and stored in a cold room (<16°C) at Jimma University until distribution. Supplementation was provided for a total duration of 12 mo through monthly distribution schedules. Mothers were instructed to take a daily dose of two capsules and provide their child a daily dose of 2 spoons (± 40 g) of the food supplement mixed with preboiled water. They were advised to keep the supplement packages closed in a cool and dark place and avoid direct cooking of the food supplements.

Weekly home-visits were conducted by trained project community workers to provide counseling on breastfeeding and preparation of supplementary foods, and monitor compliance to supplementation. Compliance to the maternal intervention was measured by counting remaining capsules in the supplement container and asking mothers about their frequency of breastfeeding during the day prior to the home-visit. Child compliance was assessed by collecting weekly data on the amount of supplementary food remaining in the food bag, i.e. 'full', '¾ full', '½ full', or 'empty'. Leftover supplements from the previous month were collected during the monthly distributions of new supplies.

Children in all study arms received deworming medication (500 mg of mebendazole) at the age of 12 and 18 mo. Children and mothers with any illness were referred to the nearby health post with a referral slip for the local health extension workers, whereas the presence of any danger sign or suspected potential adverse event was immediately reported to the researchers.

The study protocol was approved by the Ethics Committee of Ghent University Hospital in Belgium (registration number: B670201214299), the Institutional Review Board of Jimma University in Ethiopia, and the National Health Research Ethics Committee of Ethiopia. The use of study supplements was approved by the Food, Medicine and Health Care Administration and Control Authority of Ethiopia. Participants were asked to sign a written consent after an individual information session in the presence of a community member. The trial was registered at *ClinicalTrials.gov* as NCT01817634.

Outcomes

The primary study outcome was linear growth as measured by the monthly change in length-for-age z-score (LAZ) over the 12 mo intervention follow-up. Secondary outcome measures included ponderal growth assessed by the monthly change in weight-for-length z-score (WLZ), mid-upper arm circumference (MUAC), and head circumference

(HC); the longitudinal prevalence of diarrhea, vomiting, fever, acute respiratory illnesses, and any illness that required medical attention during the intervention period; the longitudinal development of stunting (LAZ<-2), wasting (WLZ<-2), anemia, and systemic inflammation, and blood LC-PUFAs concentrations at midterm (after 6 mo of intervention) and at endline (after 12 mo of intervention).

Field measurements

Child anthropometry was recorded monthly at the 3 study sites by teams of trained data collectors using calibrated equipment and standardized techniques (Cogill, 2003).

All measurements were done in duplicate by two different teams and written down on separate forms so that the first measurement could not influence the second measurement. A study data quality manager then compared the duplicate measurements and both teams repeated the measurement when there was a large difference (≥0.5 kg for weight, ≥1.0 cm for length, and ≥0.5 cm for MUAC and HC). Length was measured in the recumbent position to the nearest 1mm using a portable foldable length board (SECA 417, Germany). Weight measurements were taken together with the mother using an electronic scale (SECA 876, Germany) to the nearest 50 g. Flexible, non-stretchable measuring tapes were used to measure HC (SECA 212, Germany) and MUAC (MUAC S0145610, UNICEF) to the nearest 1mm. Child age was estimated using date of birth data from the GGHDSSthat typically records the dates of birth during the neonatal phase. For study sites outside the catchment of the GGHDSS (16% of the participants), date of birth was either copied from birth certificate or recalled using a local events calendar.

Hemoglobin and C-reactive protein (CRP) concentrations were measured on site from a single finger prick blood sample by using the QuickRead go instrument (Orion Diagnostica, Espoo, Finland) that was calibrated using control kits at the start of every measurement day. Anemia was defined as having a hemoglobin concentration <11.0 g/dL and the presence of systemic inflammation by a CRP concentration ≥5 mg/L(WHO, 2014).

Structured questionnaires and observation forms were used to collect morbidity data through weekly home-visits. Trained project community workers interviewed mothers to estimate the number of days since the last visit, typically during the previous 7 days, that the child experienced diarrhea, cough and vomiting. Diarrhea was defined as the occurrence of ≥3 liquid/ semi-solid stools within a day. The occurrence of an illness that required medical attention (hospital or health center visits) was also recalled as a proxy for serious illness. Children were assessed for runny nose, fever, tachypnea and difficulty of breathing (grunting or chest in-drawing) during each home-visit. Fever was defined as having an armpit body temperature of ≥37.5˚C; tachypnea as having a respiratory rate of ≥50 per minute for infants and ≥40 per minute for older children (>12 mo). Pneumonia was operationalized as the concurrent presence of cough and tachypnea.

Household wealth status was assessed by structured questionnaire adapted from the DHS wealth index items (CSA [Ethiopia] and ORC Macro, 2006) and a principal component analysis (PCA) was applied to generate a latent factor that was used to categorize subjects into wealth tertiles. Households were observed on a five component hygiene behaviours using a checklist (spot test).Four components related to drinking water (three items), food (four items), personal hygiene (eight items), and domestic household hygiene (six items) were used (Webb et al., 2006).Each item was scored as 0 or 1 with one representing a favourable behaviour. A summary hygiene index or score was then generated from the sum of the four specific indices and had a total of 21 points possible (0–21 points). We added few items in relation to food and personal hygiene components of the index.

Community workers, all female high school graduates from the local communities, were recruited to conduct the weekly follow-up home-visits for compliance assessment and morbidity surveillance. They received intensive training at the start of the study and refresher trainings during the monthly distribution schedules. The research team supervised community workers through unannounced monthly home-visits applying the Lot Quality Assurance Sampling approach (Pezzoli et al., 2009). Every mother-child dyad had a study participation card that community workers would sign off after a home-visit. During the supervision home-visits, the number of capsules and the amount of supplementary food would be counted and estimated and compared to the weekly report of the community worker. The morbidity record of the community workers was also compared against morbidity data collected by study nurses during the monthly distribution schedules.

LC-PUFA analysis

Blood spot samples were collected in the field from a sub-sample of 164 children. Prior to sample collection, blood spot cards (TFN, Munktell) were impregnated with BHT (2,6-Di-tert-butyl-4-methylphenol) to minimize the oxidation of LC-PUFAs as previously described by Ichihara et al. (Ichihara et al., 2002). A large drop of blood from a finger prick was collected on preprinted circles on the spot cards. Blood spot cards were transported in cold chain; dried overnight at room temperature; and once dry, inserted into aluminum-coated airtight envelopes with desiccants before storage at -80^{0}C in a central lab. Samples were later shipped on a dry ice to a laboratory in Belgium and stored at -80°C upon arrival until analyses.

To measure whole blood DHA, EPA, and arachidonic acid (AA) concentrations, discs of 8mm diameter (corresponding to ±21.8 µl of blood) (41) were punched from the blood spot cards and lipid extraction was performed based on the method detailed by Bailey-Hall et al. (42). Fatty-acid methyl esters were separated by gas chromatography with cold-on column injection (0.1 µL; GC-FID 6890N, Agilent Technologies) and a fatty-acid methyl ester column (CP-Sil 88, 60 m length, 0.25 mm ID, 0.20 µm film thickness, Agilent Technologies). Retention times were compared to a standard (GLC-68 D, Nu-Chek-Prep), and fatty-acids were quantified relative to 19:0 internal standard.

Statistical analyses:

A sample size of 360 subjects (90 for each study arm) was required to detect an effect size of 0.027 on monthly LAZ changes over the 12 months of follow-up with a statistical power of 80%, a type I error of 5%, and an anticipated attrition of 20% (Diggle et al., 2004). Continuous double data entry was performed using EpiData v1.4.4.4 (EpiData Association, Odense, Denmark). Consistency checks were conducted using Stata 13.1. Average value of the duplicate anthropometry measurements was used for analysis and LAZ and WLZ scores were calculated based on the WHO 2006 Child Growth Standards(de Onis et al., 2004b).

Growth curves were fitted for the difference from baseline of the monthly anthropometric measurements using linear mixed-effects model that included child as random intercept and intervention exposure time as random slope. Fixed-effects included intervention group, intervention time, quadratic time, and the interaction between intervention group and time that compare each intervention arm with the control arm on monthly linear growth over time. For the outcomes assessed at baseline, midterm and endline, we fitted mixed-effects linear probability model (LPM) with child-level as a random intercept whereas fixed effects included intervention group, intervention time and the interactions between intervention group and time to compared study arms by the development of an outcome over the three measurement points. For LC-PUFAs measurements, mixed-effects linear model with random intercept child was used to estimate the main effect of intervention group on the midterm and endline measurements adjusted for the baseline measurement. The main effect of time and time by group interactions were not considered in this model because blood concentrations reach a level of saturation after only few weeks of supplementation. Percentage change from the baseline measurement was also calculated for LC-PUFAs measurements. We applied log transformations to normalize the heavily skewed LC-PUFA distributions before analysis.

Longitudinal prevalence of morbidity outcomes during follow-up were calculated using as numerator the total number of days the outcome was positive and as denominator the total days observed or assessed (Morris et al., 1996). We used a negative binomial regression model to compare the impact of the interventions between study arms adjusting for the total days observed or assessed. Compliance and breastfeeding frequency during the study follow-up were compared among study arms using Fisher's exact test for the categorical variables and Kruskal-Wallis test for the continuous variables.

In the presence of significant overall group difference, pairwise comparisons of each intervention arm against the control group were done using Bonferroni adjustment to control for overall type-I error. All models were adjusted for sex of the child due to the imbalance among study arms at baseline. We also assessed for interactions between child sex and intervention group on the study outcomes. All analyses were conducted using Stata 13.1 (StataCorp, Texas, USA) with statistical significance set at $\alpha<5\%$. All statistical tests were two-sided. Analyses were done based on the intention-to-treat principle.

6.4. Results

From a total of 413 mother-infant pairs screened, 360 were randomized into the C (n=90), MI (n=89), CI (n=90), and MCI (n=91) intervention arms **(Figure 6.1)**. Three-hundred and fourteen (87%) participants completed all the 12 months of intervention follow-up (C: 91%, MI: 90%, CI: 79%, and MCI: 89%). We analyzed all children with at least one follow-up measurement for an outcome; which included 352 (98%) of children initially randomized into the study for the monthly anthropometry outcomes and 329 (91%) children for the outcomes assessed at midterm and endline. Study arms were comparable on baseline characteristics of participants except that there was an imbalance in child sex (**Table6.2)**.

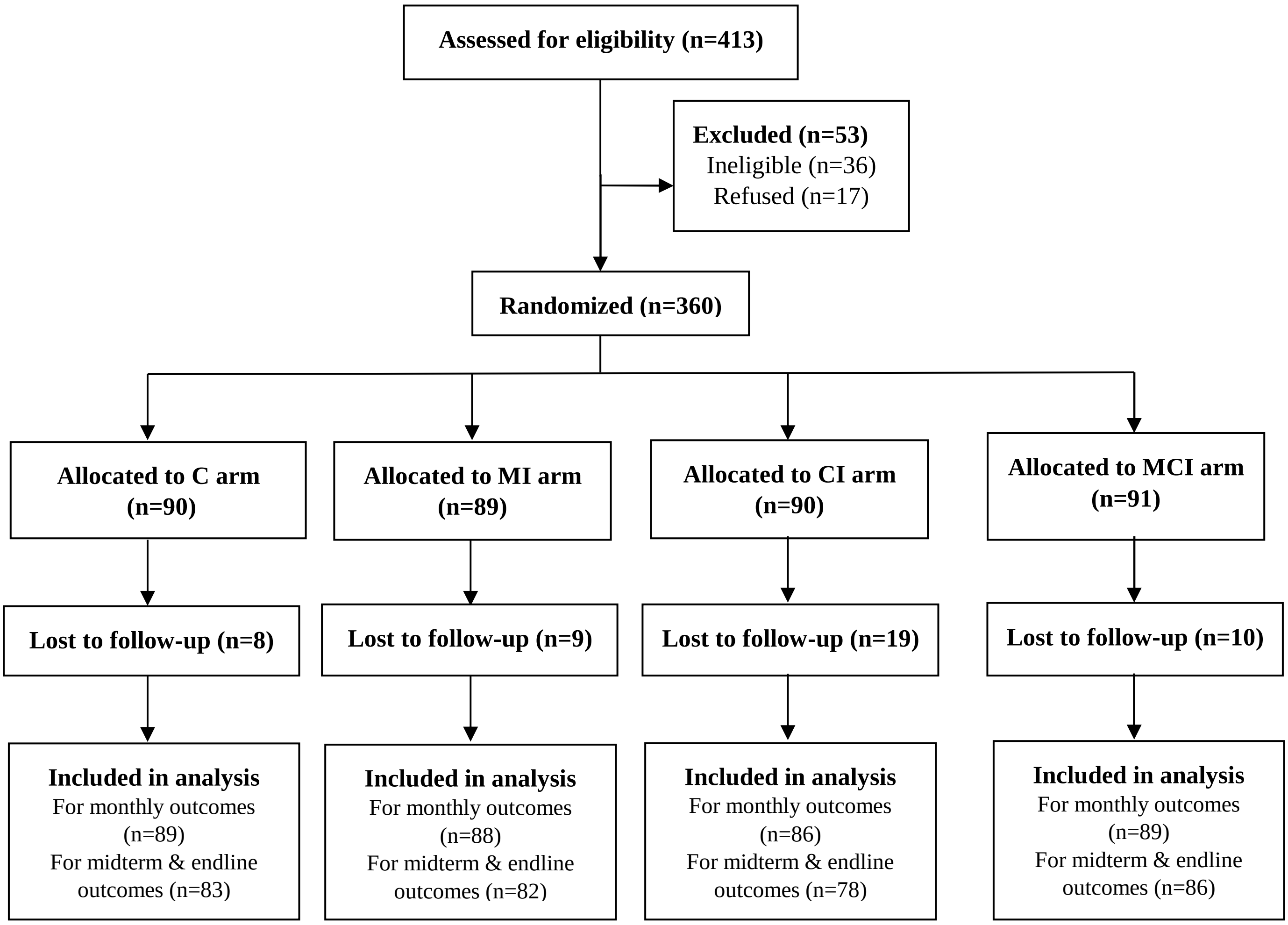

Figure 6.1 Trial flowchart summarized according to the consolidated standards of reporting trials. Analyses included all subjects with at least one follow-up measurement of the outcome variable. C, both mother & child received placebo; CI, child received fish-oil & mother received placebo; MCI, both mother & child received fish-oil; MI, mother received fish-oil & child received placebo.

Table 6.2: Baseline characteristics of study participants[1]

Characteristics	C (n=90)	MI (n=89)	CI (n=90)	MCI (n=91)
Sex, Females	42 (46.7)	49 (55.1)	36 (40.0)	54 (59.3)
Age (mo)	8.89 ± 2.16	9.18 ± 2.09	8.93 ± 2.10	8.68 ± 2.00
Maternal age (yrs)	26.0 ± 5.04	25.8 ± 4.82	26.1 ± 5.48	26.3 ± 5.28
Maternal education				
No formal education	42 (47.2)	44 (50.6)	45 (54.2)	47 (53.4)
Primary education	36 (40.5)	29 (33.3)	25 (30.1)	31 (35.2)
Secondary and above	11 (12.4)	14 (16.1)	13 (15.7)	10 (11.4)
Household wealth tertiles				
Lowest	26 (29.2)	33 (37.9)	27 (32.5)	30 (34.1)
Middle	30 (33.7)	25 (28.7)	30 (36.1)	31 (35.2)
Highest	33 (37.1)	29 (33.3)	26 (31.3)	27 (30.7)
Hygiene score tertiles				
Lowest	30 (34.1)	26 (30.2)	29 (34.5)	30 (34.5)
Middle	31 (35.2)	29 (33.7)	30 (35.7)	25 (28.7)
Highest	27 (30.7)	31 (36.1)	25 (29.8)	32 (36.8)
Maternal height (cm)	157 ± 4.67	157 ± 5.55	157 ± 5.95	157 ± 5.74
Maternal BMI (kg/m^2)	20.2 ± 2.41	20.3 ± 2.50	20.1 ± 2.57	21.0 ± 3.31
Breastfeeding during previous day				
4-6 times/day	10 (11.1)	6 (6.74)	11 (12.2)	8 (8.89)
7-9 times/day	21 (23.3)	28 (31.5)	26 (28.9)	23 (25.6)
> 10 times/day	59 (65.6)	55 (61.8)	53 (58.9)	59 (65.6)
Anthropometric indices				
Length-for-age z-score	-0.99 ± 1.07	-1.03 ± 1.10	-1.08 ± 1.15	-0.97 ± 1.11
Weight-for-length z-score	0.26 ± 0.94	0.18 ± 1.04	0.06 ± 0.99	0.09 ± 0.93
Head circumference (cm)	44.1 ± 1.79	43.9 ± 1.68	44.0 ± 1.70	43.7 ± 1.71
Mid-upper arm circumference (cm)	14.1 ± 1.06	14.0 ± 1.04	13.9 ± 1.15	13.9 ± 1.07
Hemoglobin (g/dL)	10.6 ± 1.49	10.9 ± 1.28	10.5 ± 1.37	10.9 ± 1.20
Inflammation, CRP >5 mg/L	22 (24.4)	22 (24.7)	21 (23.3)	22 (24.2)
Blood LC-PUFAs, mg/L (n=164)	n=45	n=33	n=40	n=46
DHA	46.2 ± 16.8	51.0 ± 16.6	49.0 ± 14.1	52.8 ± 17.2
EPA	4.85 (3.21, 5.73)	4.70 (3.79, 5.50)	4.68 (3.35, 5.48)	4.53 (3.75, 6.00)
AA	222 ± 80.2	246 ± 66.7	245 ± 66.5	258 ± 67.9
AA/DHA+ EPA ratio	4.36 ± 0.73	4.48 ± 0.62	4.57 ± 0.52	4.50 ± 0.84

[1]Values are expressed as n (%), mean ± SD, or median (25th, 75th percentiles). C, both mother & child received placebo; MI, mother received fish-oil capsules & child received placebo; CI, child received fish-oil fortified supplementary food & mother received placebo; MCI, mother received fish-oil capsules and child received fish-oil fortified supplementary food. AA, arachidonic acid; BMI, body mass index; C, both mother and child received placebo; CI, child received fish-oil and mother received placebo; CRP, C-reactive protein; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; MCI, both mother and child received fish-oil; MI, mother received fish-oil and child received placebo.

Compliance and effects on LC-PUFA status

Overall mean (SD) supplementation duration was 11.0 (2.88) mo with 11.2 (2.68) in C, 11.2 (2.65) in MI, 10.4 (3.56) in CI, and 11.3 (2.45) in MCI intervention arms. Compliance, i.e. the ratio of actual supplement consumption over prescribed supplement consumption, was similar among study arms for both the child and the maternal supplements (**Table 6.3)**. Median (IQR) compliance was 79.7 (62.6, 91.4) percent for the child food supplements and 69.9 (52.2, 80.4) percent for the maternal capsules. The average frequency of breastfeeding during the study follow-up was ≥ 7 times per day in 84.2% of the study participants with no significant difference among intervention arms.

In the sub-group of 164 children with bloodspot samples collected for LC-PUFA analysis, compared to the control arm, fish-oil supplementation resulted in significantly higher blood DHA concentration in all the MI (P=0.003; increase from baseline=16.9%), CI (P=0.010; increase from baseline=17.0%), and MCI (P<0.001; increase from baseline=19.7%) arms (**Table 6.4**). Blood EPA concentration was also significantly higher in the CI (P<0.001; increase from baseline=126%) and the MCI arms (P<0.001; increase from baseline=137%), and marginally significantly higher in the MI arm (P=0.057; increase from baseline=56.5%). The ratio of AA/DHA+EPA showed significant decreases in all the intervention arms (P<0.001; decreases from baseline=17.8- 40.4%).

Table 6.3 Compliance to maternal and child interventions by study arms

Variable	C	MI	CI	MCI	*P*[1]
Breastfeeding frequency, n(%)					
1-3 times/day	0 (0.00)	2 (2.25)	0 (0.00)	5 (5.49)	
4-6 times/day	12 (13.3)	13 (14.6)	17 (18.9)	8 (8.79)	0.084
7-9 times/day	61 (67.8)	50 (56.2)	54 (60.0)	57 (62.6)	
≥ 10 times/day	17 (18.9)	24 (27.0)	19 (21.1)	21 (23.1)	
Capsules compliance[2], n(%)					
0-25	6 (6.67)	9 (10.1)	13 (14.4)	8 (8.79)	
26-50	9 (10.0)	10 (11.2)	14 (15.6)	15 (16.5)	0.440
51-75	41 (45.6)	40 (44.9)	29 (32.2)	31 (34.1)	
76-100	34 (37.8)	30 (33.7)	34 (37.8)	37 (40.7)	
Median (IQR)	70.5 (60.6, 79.6)	65.3 (54.7, 81.0)	67.4 (41.8, 81.0)	72.8 (51.4, 82.6)	0.340
Food supplement compliance[2], n(%)					
0-25	1 (1.11)	3 (3.37)	5 (5.56)	2 (2.20)	
26-50	8 (8.89)	9 (10.1)	9 (10.0)	12 (13.2)	0.700
51-75	22 (24.4)	24 (27.0)	25 (27.8)	29 (31.9)	
76-100	59 (65.6)	53 (59.6)	51 (56.7)	48 (52.8)	
Median (IQR)	84.1 (67.4, 91.8)	81.0 (62.3, 92.4)	79.0 (62.3, 89.9)	77.4 (59.6, 91.8)	0.450

[1]*P*-values for group difference were obtained from Fisher's exact test for the categorical variables and Kruskal-Wallis test for the continuous variables.
[2]Compliance is defined as the ratio of the total number of capsules or estimated amount of food supplement actually consumed over the total number of capsules or amount of food supplement prescribed.

Table6.4. Blood LC-PUFAs concentration (mg/L) at midterm and endline by study arms

Outcomes	Midterm		Endline		Effect Size (95%CI)[1]	*P*	% change[2]
	N	Mean ± SD	N	Mean ± SD			
DHA							
C	40	50.9 ± 18.7	31	41.8 ± 14.9	Reference group		-1.36
MI	31	59.3 ± 24.5	28	63.9 ± 24.9	13.5 (3.73, 23.3)	0.003	16.9
CI	32	66.7 ± 26.8	27	51.6 ± 26.2	11.9 (2.18, 21.7)	0.010	17.0
MCI	40	66.0 ± 24.3	35	63.6 ± 22.0	16.0 (6.51, 25.5)	<0.001	19.7
EPA[2]							
C	40	5.12 (4.07, 8.67)	31	5.04 (3.76, 5.76)	Reference group		14.5
MI	31	7.66 (4.76, 9.20)	28	6.59 (4.63, 8.95)	2.09 (0.99, 4.45)	0.057[3]	56.5
CI	32	11.5 (6.75, 21.3)	27	9.90 (4.30, 15.2)	4.39 (2.07, 9.33)	<0.001[3]	126
MCI	40	13.3 (6.54, 22.5)	35	10.9 (5.83, 17.8)	5.83 (2.81, 12.1)	<0.001[3]	137
N-3 LC-PUFA (DHA + EPA)							
C	40	58.7 ± 23.7	31	47.2 ± 16.1	Reference group		-0.30
MI	31	67.9 ± 28.1	28	72.0 ± 28.6	16.0 (4.04, 27.9)	0.004	21.7
CI	32	83.6 ± 37.2	27	62.3 ± 31.9	19.6 (7.69, 31.5)	<0.001	26.0
MCI	40	83.4 ± 34.6	35	76.9 ± 28.5	24.9 (13.3, 36.5)	<0.001	36.3
AA							
C	40	239 ± 73.9	31	215 ± 70.9	Reference group		-2.64
MI	31	212 ± 62.7	28	243 ± 87.7	-3.62 (-33.8, 26.6)	1.000	-0.68
CI	32	218 ± 65.5	27	183 ± 90.7	-28.2 (-58.5, 2.07)	0.077	-16.8
MCI	40	202 ± 63.2	35	214 ± 65.5	-27.1 (-56.6, 2.35)	0.083	-15.4
AA/DHA+ EPA ratio							
C	40	4.27 ± 0.85	31	4.65 ± 0.96	Reference group		4.53
MI	31	3.33 ± 0.86	28	3.47 ± 0.90	-1.07 (-1.51, -0.62)	<0.001	-17.8
CI	32	2.89 ± 0.93	27	3.26 ± 1.17	-1.44 (-1.89, -1.00)	<0.001	-35.7
MCI	40	2.68 ± 1.07	35	3.00 ± 1.08	-1.64 (-2.07, -1.21)	<0.001	-40.4

[1]Mixed-effects linear model with child-level as random intercept and fixed effects including child sex, baseline measurement, and intervention group; *P*-values are for overall group difference; Effect sizes comparing intervention arms against the control are estimated using Bonferroni adjustment for multiple comparisons.
[2]Percent change (median) from baseline at midterm and endline measurements.
[3]Values are expressed as median (IQR) and because analysis was based on log transformed values, effect sizes (95% CI) are expressed as the antilog of coefficients indicating the ratio of the geometric means in an intervention arm over the control arm.
AA, arachidonic acid; C, both mother and child received placebo; CI, child received fish-oil and mother received placebo; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; LC-PUFA, long-chained polyunsaturated fatty acids; MCI, both mother and child received fish-oil; MI, mother received fish-oil and child received placebo.

Effects on growth and nutritional status

We found no significant effect in any of the intervention arms on the primary study outcome linear growth (LAZ) (**Figure 6.2)**. However, a statistically significant group difference was detected on ponderal growth expressed as WLZ (*P*=0.013). Compared to the control arm, significantly larger monthly increases in WLZ were detected in the CI arm (effect size (95% CI): 0.026 (0.004, 0.048) z-scores per month; *P*=0.016) and the MCI arm (effect size (95% CI): 0.022 (0.000, 0.043) z-scores per month; *P*=0.051); whereas no significant effect was found in the MI arm. We also found a trend towards larger monthly MUAC increases in the CI and the MCI arms compared to the control arm, though the differences were not statistically significant. No significant effect of any of the interventions was found on the nutritional status of children using stunting, wasting, and anemia (**Table6.5**).

Effects on morbidity and inflammation

The occurrence of systemic inflammation, as determined by CRP concentration (**Table 6.5**), and common childhood illnesses (**Table 6.6**) were not statistically different between study arms.We did not find any adverse effect related to the study supplements in all participating mothers and children.

Table 6.5. Nutritional status and systemic inflammation by study arms

Outcomes	Baseline				Midterm				Endline				P[1]
	C (n=90)	MI (n=89)	CI (n=90)	MCI (n=91)	C (n=83)	MI (n=81)	CI (n=78)	MCI (n=84)	C (n=82)	MI (n=80)	CI (n=71)	MCI (n=81)	
Stunting, n(%)	15 (16.7)	11 (12.4)	23 (25.6)	21 (23.1)	20 (24.1)	23 (28.4)	25 (32.1)	24 (29.3)	26 (31.7)	26 (32.5)	35 (49.3)	24 (29.6)	0.202
Wasting, n(%)	6 (6.67)	6 (6.74)	11 (12.2)	7 (7.69)	7 (8.43)	2 (2.47)	8 (10.3)	5 (5.95)	7 (8.54)	6 (7.50)	9 (12.7)	5 (6.17)	0.934
Anemia[2], n(%)	49 (54.4)	39 (44.3)	55 (61.1)	47 (51.6)	20 (24.1)	24 (29.6)	28 (35.9)	26 (31.0)	15 (18.3)	17 (21.3)	18 (25.4)	13 (16.0)	0.378
Inflammation[2], n(%)	22 (24.4)	22 (24.7)	21 (23.3)	22 (24.2)	22 (26.5)	25 (30.9)	27 (34.6)	32 (38.1)	14 (17.1)	23 (28.7)	18 (25.4)	15 (18.5)	0.512

[1]Mixed-effects linear probability model with random intercept child and fixed effects including child sex, intervention group, intervention time, and groupXtime interaction; *P*-values are for groupXtime interaction indicating groups' difference on development of the outcome over time.

[2]Anemia, hemoglobin<11.0 g/dL; inflammation, CRP>5 mg/L.

C, both mother & child received placebo; MI, mother received fish-oil capsules & child received placebo; CI, child received fish-oil fortified supplementary food & mother received placebo; MCI, mother received fish-oil capsules and child received fish-oil fortified supplementary food.

Table 6.6. Morbidity outcomes during the intervention follow-up by study arms[1]

Outcomes	C	MI	CI	MCI	*P*
Diarrhea					
Child wks with event/total child wks followed	248/4195	222/4066	190/3858	189/4213	
Longitudinal prevalence (95% CI)	5.91 (5.64, 6.20)	5.45 (5.18, 5.73)	4.93 (4.67, 5.20)	4.49 (4.25, 4.73)	0.165
Cough					
Child wks with event/total child wks followed	258/4195	243/4066	212/3858	239/4213	
Longitudinal prevalence (95% CI)	6.16 (5.88, 6.45)	5.98 (5.70, 6.27)	5.50 (5.22, 5.79)	5.68 (5.41, 5.96)	0.948
Vomiting					
Child wks with event/total child wks followed	78/4195	74/4066	66/3858	71/4213	
Longitudinal prevalence (95% CI)	1.87 (1.72, 2.03)	1.82 (1.67, 1.99)	1.71 (1.56, 1.87)	1.69 (1.55, 1.85)	0.878
Runny nose					
No. of visits with diagnosis/total no. of visits	1468/4204	1494/4069	1389/3864	1508/4226	
Longitudinal prevalence (95% CI)	34.9 (33.2, 36.8)	36.7 (34.9, 38.6)	35.9 (34.1, 37.9)	35.7 (33.9, 37.5)	0.976
Tachypnea					
No. of visits with diagnosis/total no. of visits	208/4204	160/4069	183/3864	201/4226	
Longitudinal prevalence (95% CI)	4.95 (4.30, 5.67)	3.93 (3.35, 4.59)	4.74 (4.07, 5.47)	4.76 (4.12, 5.46)	0.720
Fever					
No. of visits with diagnosis/total no. of visits	11/4204	12/4069	9/3864	12/4226	
Longitudinal prevalence (95% CI)	0.26 (0.13, 0.47)	0.29 (0.15, 0.52)	0.23 (0.11, 0.44)	0.28 (0.15, 0.50)	0.988
Pneumonia					
No. of visits with diagnosis/total no. of visits	30/4204	17/4069	19/3864	18/4226	
Longitudinal prevalence (95% CI)	0.71 (0.48, 1.02)	0.42 (0.24, 0.67)	0.49 (0.30, 0.77)	0.43 (0.25, 0.67)	0.656
Illness requiring medical attention					
Child wks with event/total child wks followed	45/4195	39/4066	43/3858	45/4213	
Longitudinal prevalence (95% CI)	1.07 (0.95, 1.19)	0.97 (0.86, 1.09)	1.12 (1.00, 1.26)	1.06 (0.95, 1.19)	0.619

[1]Longitudinal prevalence was calculated using as numerator the total number of days that the child presented a positive outcome and as denominator the total number of days observed or assessed. *P*-values for group difference from a negative binomial regression model comparing intervention groups by number of days with morbidity adjusted for child sex and log number of days observed or assessed.

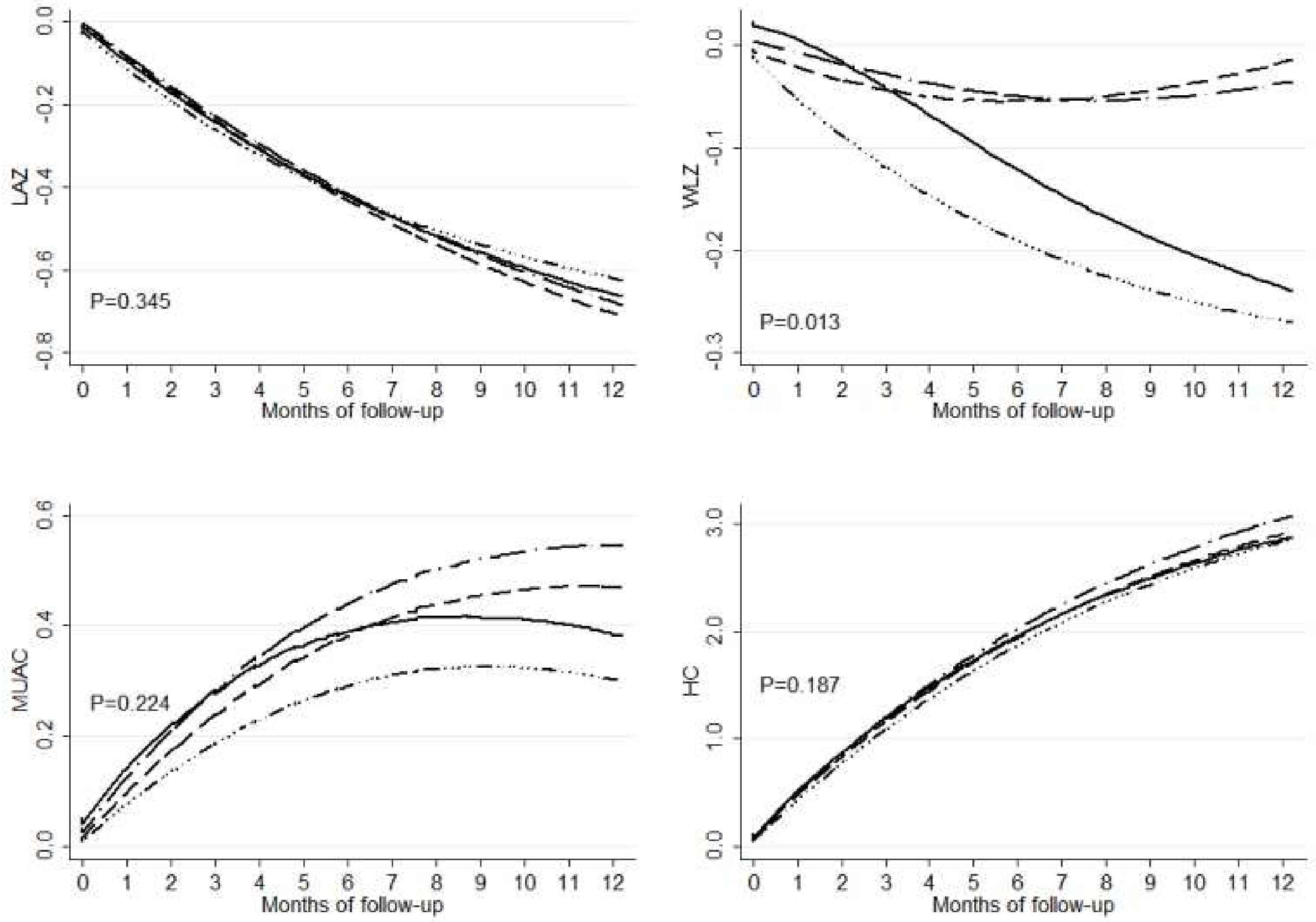

Figure 6.2. Monthly anthropometric growth over 12 mo in the C (——), MI (·······), CI (— — —) and MCI (— · — ·) arms. Plotted values for the difference from baseline of the monthly measurements are estimated from linear mixed-effects model using random intercept child and random slope intervention time with fixed effects including time, quadratic time, intervention group, and timeXgroup interactions adjusted for child sex. *P*-values are for group difference on monthly changes of the outcome over time. Effect-sizes (95% CI) on monthly changes in WLZ were 0.006 (-0.016, 0.028); *P*=1.000 in the MI, 0.026 (0.004, 0.048); *P*=0.016 in the CI, and 0.021 (0.000, 0.043); *P*=0.051 in the MCI, which were estimated using Bonferroni adjustment for multiple comparisons.
C, both mother and child received placebo; CI, child received fish-oil and mother received placebo; HC, head circumference (cm); LAZ, length-for-age z-score; MCI, both mother and child received fish-oil; MI, mother received fish-oil and child received placebo; MUAC, mid-upper arm circumference (cm); WLZ, weight-for-length z-score.

6.5. Discussion

This study evaluated the impact of n-3 LC-PUFA supplementation on child growth and morbidity in a low-income setting. N-3 LC-PUFA rich fish-oil was supplemented for 12 month through breastmilk (MI), supplementary feeding (CI) or a combination of both (MCI). In all the intervention arms, fish-oil supplementation increased blood n-3 LC-PUFA concentration and lowered the ratio of AA/DHA+EPA indicating tissue uptake of the study supplements. However, except for an increase in monthly relative weight gain and a trend towards larger MUAC increments in the CI and MCI arms compared to the control, we found no impact of fish-oil supplementation on child linear growth, systemic inflammation and morbidity.

The lack of impact of n-3 LC-PUFA supplementation on child linear growth in this study is consistent with previous studies in high income countries that evaluated the supplementation of lactating mothers or infants (Makrides et al., 2005, Simmer et al., 2008, Delgado-Noguera et al., 2010). In this setting, however, we hypothesized that a higher dietary n-3 LC-PUFA intake would modulate the child’s immune response to infectious challenges and inflammation which would subsequently lead to growth benefits. Nonetheless, we were unable to detect an effect on reported morbidity or systemic inflammation. This finding is in agreement with the Gambian study where fish-oil supplementation to breastfed infants had no effect on growth, morbidity, systemic and intestinal inflammation, or gut integrity (van der Merwe et al., 2013). The addition of fish-oil to multiple micronutrient supplementation in Malawian children 12-35 mo of age also did not improve EED or common illnesses (Smith et al., 2014). In contrast, supplementation of DHA and AA to infant formulas resulted in a delayed onset and lower incidence of allergic symptoms and upper respiratory infections (URI) in two randomized controlled trials in the US (Birch et al., 2010). In two non-randomized studies in Spain and France, infant formula containing DHA and AA was associated with a lower incidence of respiratory symptoms and diarrhea (Pastor et al., 2006, Lapillonne et al., 2014). A lower incidence and shorter duration of URI episodes was also reported among Thai schoolchildren(Thienprasert et al., 2009) and US toddlers(Minns et al., 2010) who received milk containing fish-oil or DHA.

The lack of effect on growth and morbidity upon supplementation with n-3 LC-PUFA has also been observed by other researchers and a number of hypothesis could be forwarded.First, it could be argued that n-3 LC-PUFA status in the study children was already satisfactory and hence the added effect of supplementation would be negligible. This hypothesis was proposed by The Gambian study where it was found that breastmilk substantially contributed to n-3 LC-PUFA intake. The lack of reference values for LC-PUFA status hampers a direct assessment of children's status. However, baseline n-3 LC-PUFA concentrations of the children in our setting were significantly lower compared to the children from The Gambian study. Despite a possible difference in the method of quantification of fatty acids, our interventions also increased baseline DHA status by 16.9-19.7% and EPA by 56.5- 137%, and decreased the AA/DHA+EPA ratio by 17.8-40.4%compared to the Gambian study. Thus, it is less likely that the children in our study had an optimal n-3 LC-PUFA status and it is less likelythat functional benefits associatedwith increased n-3 LC-PUFA could appear. Second, we anticipated an important impact of n-3 LC-PUFA supplementation on morbidity symptoms and systemic inflammation in a child population characterized high exposure to recurrent infections. However, the period prevalence of diarrhea in our children was rather low (5.2%) compared to previous studies from LIC (2.4 - 16.3%)(Checkley et al., 2008). The proportion of children with systemic inflammation was also much lower in our children (24.2%) compared to the n-3 LC-PUFA study in the Gambia (44.4%) (van der Merwe et al., 2013). Hence, we cannot exclude that the relatively lower prevalence of morbidity and systemic inflammation in the study population might have attenuated the impact of the intervention. Third, the impact on children's outcomes could be conditional on the intake of n-3 LC-PUFAs from sources other than fish-oil. In the control children, blood concentrations of n-3 LC-PUFAs remained rather stable during the 12 mo of follow-up. This was an unexpected finding given that breastmilk is the only important source of preformed n-3 LC-PUFAs and one would expect that breastmilk is gradually complemented or replaced by complementary foods which are typically very low in n-3 LC-PUFAs. A possible reason for the stable n-3 LC-PUFA concentrations in this group could be the high frequency of breastfeeding throughout the follow-up. Finally, it should be noted that the children in the n-3 LC-PUFA intervention arms were also given multiple micronutrients in the food supplement and deworming medication to avoid any hampering of the efficacy of the n-3 LC-PUFAs by micronutrient deficiencies and intestinal parasite infections. Children in the control group also received the multiple micronutrient fortified food supplement without fish-oil and deworming medication in order to evaluate the net impact of n-3 LC-PUFAs on child growth and morbidity. It can therefore not be excluded that both the micronutrients and deworming might have exerted a positive, albeit small, effect on child linear growth, thereby possibly diluting the intervention effect.

The heterogeneous findings on child morbidity between our and the Gambian studies and other studies conducted in affluent countries may relate to differences in clinical end points and background n-3 LC-PUFA status of the study populations. The clinical benefits reported in the later studies might have been due to n-3 LC-PUFAs' modulating immune response to inflammatory conditions rather than infectious diseases. Differential effects may also arise from differences in etiologies as animal experiments found that dietary n-3 fatty-acids can improve or impair the host response to infectious challenges depending on the type of pathogen (Anderson and Fritsche, 2002). It is also possible that breastfeeding might have provided some protection in our and the Gambian children compared to the formula-fed infants and older children in the other studies. Moreover, the effects of n-3 fatty-acids on the immune system may vary according to dose, age of exposure, polarization and profile of the involved immune system (T helper, Th1/Th2)(Gottrand, 2008).

Our study found that direct fish-oil supplementation to children or combined with maternal fish-oil supplementation resulted in higher ponderal growth, indicated by increased WLZ rates, and a trend to larger MUAC increments over time as compared to the control group. This finding has also been reported by other trials that showed positive impacts of n-3 LC-PUFA supplementation on body mass index, MUAC, skinfold-thickness and waist circumference suggesting changes in body composition(van der Merwe et al., 2013, Lauritzen et al., 2005b). The competitive roles of n-3 and n-6 fatty-acids on the regulation of genes encoding for fat oxidation, thermogenesis, and adipose differentiation, demonstrated in animal studies, have been suggested as plausible mechanisms for effects on the rate and/or composition of weight gain (Lapillonne et al., 2004, Lapillonne et al., 2003). On the other hand, the clinical significance of the observed intervention effect on ponderal growth of children in our study requires careful consideration because of the small effect size (Heird, 1997). Moreover, the difference in the rate of weight gain from the C arm was 0.5 and 0.6 g/d in the CI and MCI arms; which could be very small despite the lack of reference for this age group. The recommended nutritionally significant rate of weight gain between formula-fed and breast-fed infants in the first few months of life is >3 g/d (AAP (Committee on Nutrition), 1988).

The study has a few limitations that need to be considered. We did not include markers of immune cell function, intestinal inflammation and indicators of intestinal integrity which impedesa more mechanistic understanding of the effect of the intervention. It is also important to note that we had limited power to detect small differences in morbidity outcomes with low prevalence estimates. The study also did not consider possible effect modification by conditions at birth such as gestational age and birth weight. We also did not monitor intake of n-3 LC-PUFA from other sources other than the supplement. However, previous dietary surveys in the study area indicated that intake of fish and other sea foods, the only important sources of n-3 LC-PUFA, is rare.Only one out of around 300 infants consumed fish during the pre- and post-harvest seasons as measured by repeated 24-hr dietary recalls(Wondafrash et al., 2016).

In conclusion, this study adds to the growing body of evidence on the lack of growth benefits from n-3 LC-PUFA supplementation to infants and young children. Despite an improvement in n-3 LC-PUFA status, we did not find a benefit on linear growth or morbidity. The increased pondral growth in the intervention arms could indicate increased adiposity but this should be confirmed by body composition measurement. Nonetheless, the need for n-3 LC-PUFA supplementation remains a point of discussion due to the possibility that it may improve the development of the visual system and cognitive performance in early life.

7

Chapter 7: General Discussion

7.1. Introduction

World Health Organization has put a global target of reducing stunting by 40% of the 2010 level by the year 2025(WHO, 2012) at a rate by 3.9% annually(de Onis and Branca, 2016). The organization has aimed at alleviating the double burden of malnutrition in children, starting from the earliest ages. The plan includes six global nutrition targets: child stunting, wasting, and overweight; anemia in women of reproductive age; low birth weight; and exclusive breastfeeding. A later prediction, however, indicated that there will only be a 26% reduction from the level in 2010 (de Onis et al., 2013). Much of the African region remain behind the targeted level of stunting. Some of the strategies underlined to alleviate malnutrition include promotion of breastfeeding, food-based approach to improving micronutrient intake or supplementation, prevention of severe malnutrition, hygienic preparation and consumption of complementary foods. It is now generally agreed by economists all over that, the return on investment in early childhood is many times the size of the original investment (Irwin et al., 2007).

The WHO strategies mentioned above could prevent about a quarter of child deaths occurring in under 36-month-olds and reduce stunting by one-third in the same age group (Bhutta and Salam, 2012). Apart from the nutrition specific intervention, the recent emphasis is given to nutrition-sensitive interventions involving agriculture, maternal education and empowerment, improved hygiene and safe water supply, social protection programs which ensure better purchasing power, and access to essential health care services and goods (Ruel and Alderman, 2013). One of the initiatives toward this end has been the Scale Up Nutrition (SUN) Movement which utilizes both nutrition specific and nutrition sensitive approaches linking health, care, hygiene and sanitation, access to food and resources and women’s empowerment (http://scalingupnutrition.org/).

As a result of ever increasing size of the population of the world, meeting the demands in terms of adequate and quality food is a challenge. The problem of inadequate amount and poor quality diet is more pronounced in low income countries. Like many countries in the region, undernutrition remains one of the top public health problems in Ethiopia. Despite the country’s reduction in the level of undernutrition over the past decade and half, the prevalence of stunting, wasting, and underweight is one of highest in Africa (CSA [Ethiopia] and ICF, 2016). Much of the linear growth faltering occurs during the complementary feeding period in which the peak is reached between 12 and 18 months (Headey, 2014), hence, appropriate care and feeding practices in terms of quantity, quality and hygienic preparation and storage can alter such trajectory positively.

Child care practices in the context of child feeding remain integral part of addressing child malnutrition globally. The “first 1000 days” has been considered very critical in terms of vulnerability for malnutrition and as also an important stage whereby interventions have the maximum benefit. Hence, this Ph.D. work addressed this critical period in terms of understanding the patterns of caregiver child interaction, describing the different indices of infant and young child and their utility as indicators of nutrient intake and growth. Moreover, the influence of agricultural seasons on dietary quality, growth and feeding practices has been addressed in this setting where majority of the population are reliant on own production from a rain fed agriculture.

From our earlier publications, it was observed that intake and supply of n-3 LC-PUFA has been almost nil which can be reflected on the content of breast milk in those mothers residing in the same community (Michaelsen et al., 2011). Moreover, the current practice of breast feeding for the target children is suboptimal compared to the recommendations even if breast feeding is universally practiced in Ethiopia. Hence, supplementing lactating mothers and enriching a processed complementary food with essential fatty acids was warranted.

Therefore, this chapter discusses the main study findings, implications of the studies to a wider context of child care practices, methodological issues and research and policy recommendations which need to be taken up subsequently.

7.2. Main research findings

Based on the different studies conducted under this Ph.D. work, the following are the main findings.

1. As feeding styles are embedded in ethno-cultural behaviors, control over feeding of one’s child could vary across cultures. Hence, rural mothers in Derashe Special District practiced responsive feeding compared to laissez-fair feeding and controlling feeding styles. Most of the caregivers are aware that eating or food acceptance or intake of food decreases in response to illness or if caregivers are exercising force feeding. Significant proportion of caregivers claimed that they practiced responsive feeding in case of refusal. Caregivers other than biological mothers practiced laissez-fair and controlling feeding styles (**Chapter 3**).

2. Even if there was no significant difference in terms of DDS and intake of essential micronutrients in both HS and PHS, it was very clear that children were taking poorly diversified diet with possibility low micronutrient content and bioavailability. Most of the children in both seasons were dependent on cereals and tubers for their micronutrient intake from which micronutrients are less available. There was no difference in age adjusted MMDA between the two seasons. Strangely, there was no statistically significant difference in mean DDS between the two seasons. However, there was significantly higher prevalence of morbidity and lower HAZ in PHS. It is expected that high quality food groups are less likely to be consumed in PHS even if there was no difference in MMDA and DDS between the two seasons. A low DDS (≤2) predicted low MMDA; however, it failed to predict higher MMDA as most of the children took few food groups in the previous 24 hours (**Chapter 4**).

3. The level of linear growth restriction (expressed as mean of LAZ(SD)) and prevalence of stunting during infancy is high for HS , PHS and follow up children even if it was lower than the national average for the EDHS 2011. The prevalence of wasting was considered high in both seasons both for 6-12 and 12-18 month old children. The DDS and ICFI were not different in the two seasons and there was no association of DDS or ICFI with growth, LAZ and WHZ in the two seasons for the 6-12 month old infants. After adjusting for important confounding factors, low DDS was associated with stunting in HS and in the pooled data (from 6-12 months old children at HS and PHS). Moreover, LAZ at follow up (in PHS) was associated with DDS at baseline (HS) and the presence of diarrhea within season at follow-up negatively affected LAZ (Chapter 5).

4. In the supplementation with n-3 LC-PUFA via breast milk and processed supplementary food, we found no significant effect in any of the intervention arms on linear growth; however, a statistically significant group difference was detected on ponderal growth expressed as WLZ (P=0.013). Compared to the control arm, significantly larger monthly increases in WLZ were detected in the CI and MCI arms though the effect sizes were small; whereas no significant effect was found in the MI arm. Although not statistically significant, we found a trend towards larger monthly MUAC increases in the CI and MCI arms compared to the control arm. No significant effect of any of the interventions was found on the nutritional status of children

(expressed in term of stunting, wasting, and anemia), occurrence of systemic inflammation and common childhood illnesses (**Chapter 6**).

5. In the sub-group of 164 children from which bloodspot sample was taken, the supplementation with n-3 LC-PUFA resulted in significantly higher blood DHA in CI, MI and MCI arms compared to the control. The blood concentration of EPA was also higher in the CI and MCI arms and there was lower ratio of ARA/DHA+EPA in all interventions arms (**Chapter 6**).

In sum, the aforementioned findings demonstrate that responsive feeding style is adopted by majory of caregivers which is the recommended behavior for the age group studied (6-23 months)(PAHO/WHO 2003). However, such behaviors are culture specific and generalization of the observation to other regions in Ethiopia or beyond is problematic. We found also that IYCF indicators can be used as important tools for population level assessment, targeting, monitoring and evaluation. The DDS was stable across seasons as proxy for nutritional quality of complementary foods. However, there is lack of robustness in the association of some of the indicators with growth and nutritional status across seasons. Moreover, enriching a breast milk and a processed supplementary food with n-3 LC-PUFA did not affect the primary endpoint (linear growth) in children even though there was an increase in the concentration of EPA and DHA and a decrease in AA and the ratio of ARA to EAP+DHA in the intervention arms which signify tissue uptake. There was also an effect on pondral growth (WLZ) in the in MCI and CI arms.

7.3. Caregiver-child interaction and child outcomes

Caregiver-child interaction (caregiver feeding styles) is found to be an important aspect of care during complementary feeding. Feeding practices and styles are interchangeably used but feeding practices refer to “what” caregivers do; while caregiver styles refer to “how” they do it (Jansen et al., 2012). We found that responsive feeding was practiced by majority of the mothers in Derashe district (Wondafrash et al., 2012). Ruel et al (1999) demonstrated that a “care index” involving breastfeeding, complementary feeding, food quality, and three active feeding behaviors and preventive health care services were associated with child nutritional status especially among mothers from poor households and with less than a secondary education(Ruel et al., 1999). Children of responsive or proactive caregivers are more likely to take more food (Dearden et al., 2009, Flax et al., 2011, Bentley et al., 2011) or such feeding behavour increases the number of mouthfuls accepted (Abebe et al., 2017) and it is an important factor for healthy eating behavior and self-regulation (Harbron and Booley, 2013). We also found that caregivers who breastfed frequently and those other than biological mothers were less responsive (Wondafrash et al., 2012) even if we were not able to see the effect in terms of intake of complementary food or nutritional status. Non responsive feeding , force feeding, or controlling type of feeding have been associated with either avoidance and undernutrition or over eating with long term effect of poor regulation of eating and overweight or obesity(Engle and Pelto, 2011, Hurley et al., 2011).

There are quite few studies looking at the short term and long term benefits of appropriate caregiver-child interaction (active or responsive feeding) for the child even if such practice is being recommended as part of the optimal complementary feeding practice for children 6-24 months (Engle and Pelto, 2011); however, disaggregating the effect of responsive or active feeding on child outcome could be challenging and most of the studies which included responsive feeding failed to disentangle its effect. Two recent studies in the Western part of Ethiopia used direct observation to assess the infant-caregiver interaction during feeding. Abebe et al found that caregiver's active positive feeding (encouragement to eat) and responsive positive feeding (identifying and responding to child's cues) were associated with mouthful accepted and LAZ(Abebe et al., 2017) among infants. Moreover, mothers who breastfeed mostly are less responsive. Another analysis in the same study areain Ethiopia demonstrated that responsive positive feeding style was associated with increased energy intake after controlling for confounding factors (Baye et al., 2017). However, the latter studies were cross-sectional and temporal relations cannot be ascertained.

7.4. Infant and young child feeding practice indicators, dietary quality and child growth

It is acknowledged that appropriate IYCF is an important determinant of child growth and development. Apart from the study on "how" feeding occurs (Wondafrash et al., 2012), we assessed the adherence to some of the components of guiding principles of complementary feeding among the breastfed (PAHO/WHO 2003) using the different indicators (WHO, 2008). We also looked at the quality of the diet through the measurement of diversity of the diet and creating a composite index comprising of different dimensions of feeding. In agreement with few researches conducted in developing countries, we found that DDS was associated with dietary quality expressed in terms of MMDA in two distinct seasons in which consumption of ≤2 food groups best predicted low micronutrient intake (Wondafrash et al., 2016) and intake of this amount was also associated with stunting in HS and in the pooled data from HS and PHS(Wondafrash et al., 2017). However, ICFI was not associated with growth or nutritional status in both seasons.

As a result of multidimensional and age specific nature of IYCFs, constructing an appropriate and valid indicator has been a research agenda by various individuals and expert groups starting from early 2000. To this end, composite indices were created by accumulating scores assigned to individual components (indicators) which in principle are more robust in terms of predicting child health and nutrition outcomes as introduced in **Chapter 1.** Association of the DDS and composite indices with dietary quality (Hatloy et al., 1998, Kennedy et al., 2007, Moursi et al., 2008a) and nutritional status or growth (Onyango et al., 1998, Sawadogo et al., 2006, Ntab et al., 2005, Arimond and Ruel, 2004, Bork et al., 2012, Moursi et al., 2008b, Marriott et al., 2012, Reinbott et al., 2015, M'Kaibi et al., 2016 , Onyango et al., 2014) was reported before. However, there was lack of consistency in terms of the association of IYCF indicator (s) or composite indices and dietary quality, nutritional or health status (Kourlaba and Panagiotakos, 2009). In situation where children's diets are composed of few food groups, the DDS is unable to predict higher micronutrient intake as the two are highly correlated in many studies. The lack of predictive ability of DDS in low income settings has been found by other researchers (Steyn et al., 2006, Moursi et al., 2008a, Kennedy et al., 2007). We found a relatively higher energy and micronutrient intake in PHS possibly due to the buffering of young children during lean seasons compared to adults or adolescents (Maxwell, 1996). The latter could be in the form of higher portion size per food group or higher frequency which could not be captured by DDS. Even if we were unable to measure maternal intake, it is proposed that maternal intake in relation to their requirement is reduced during lean seasons or in times of food insecurity at which time children are believed to be buffered through consumption of a more nutritious foods(Arsenault et al., 2014, Arsenault et al., 2013).

Majority of the researches regarding the relationship between child feeding indicators and dietary quality or nutritional status were conducted using DDS instead of composite indices. Among others, the number of components needed, relative weights (attribution of values) of the components, the cut off values have been challenging in the use of composite indices. For instance, young infants who are breastfed for extended period of time are more likely to be given a high score for breast feeding in a composite index and they may be taking less complementary foods. It was found previously that breast feeding for extended period of time was negatively associated with growth in which mothers extend breast feeding particularly for those undernourished children(Marquis et al., 1997a, Simondon and Simondon, 1998) and children who were not breastfed were more likely to be given solid or semisolid food(CSA [Ethiopia] and ORC Macro, 2011). On the contrary, those children who were bottle fed were having better LAZ in PHS which we were not able to explain from the data (Wondafrash et al., 2017). However, bottle feeding is not recommended at any age as it is associated with early cessation of breast feeding and expose children to unhygienic food which further compromises their health and nutritional status (WHO, 2010a). Moreover, the local dietary pattern and nutritional composition of foods and the varieties of the food crops used locally complicate the development of an index to be used across all regions. For instance, near to 90% of the dietary energy is coming from grains, roots and tubers in Africa and significant portion of the daily dietary consumption constitutes roots and tubers with all other food groups contributing less than 10% (Berti and Jones, 2013) which makes dietary diversity (a component of ICFI) less important in the index .

Albeit the paradox, our findings demonstrate that dietary diversity, but not ICFI, can be used as an important tool to indicate dietary quality (expressed in terms of MMDA) and such findings have been replicated by many researchers even if the number of food groups used to calculate DDS, the cut offs, the recall period and the scoring systems are variable among the different settings which implies that the indicator is robust. Infant and child feeding index is expected to have a number of dimension of feeding compared to DDS, however it was not associated with linear growth or nutritional status in both seasons. This finding necessitates further improvement of the index by including other dimensions and through the use of longitudinal design (Moursi et al., 2008b, Ma et al., 2012). Moreover, the relative importance of each component of ICFI in relation to a particular child outcome is not known and DDS could actually be the one determining such association as observed by some researchers.

The findings that dietary diversity (measured with a 24 hour dietary recall) was associated with stunting (chronic undernutrition which develops quite early during childhood) may ascertain the existence of clustering of practices as described before by other authors (Arimond and Ruel, 2003b). The aforementioned observations adds to the importance of this indicator as an important tool to assess dietary quality at population level, target interventions and for monitoring and evaluation of programs aiming at improving dietary quality and nutritional status of children without requiring to collect detailed quantitative intake of foods.

7.5. Essential fatty acids and child health and growth

Even if there is evidence of no or little effect of supplementation with n-3LC-PUFA on child growth (Makrides et al., 1999, Simmer et al., 2011 , Wieland and Santesso, 2016) in the Western populations, there is little or no evidence of an effect on linear growth or various outcomes in low income settings (van der Merwe et al., 2007). Unlike the suggestion that essential fatty acids could be the missing link in the relationship between interventions aimed at improving of complementary food intake and child growth and other outcomes (Adu-Afarwuah et al., 2007, Dewey and Adu-Afarwuah, 2008), there is little or no evidence of improvement in linear or ponderal growth with additional supplementation with n-3 LC-PUFAs (Simmer et al., 2011). A recent systematic review also failed to demonstrate an effect on growth irrespective of the amount of AA relative to DHA in the supplement (Cristina Campoy, 2012). However, there is consistent evidence that there is a dose dependent and linear relationship between dietary intake of DHA and EPA with plasma or RBC level of these fatty acids and most of the n-3 LC-PUFA supplementation trials which measured biomarkers concluded that plasma or RBC levels of EPA and DHA significantly increased in the supplemented group. Based on analysis of supplementation studies conducted on animal models, the latter observations could indicate an improvement in tissue uptake of EPA and DHA(Gurzell et al., 2014, Fenton et al., 2016). Our supplementation trial **(Chapter 6**) demonstrated lack of an effect on linear growth after n-3 LC-PUFA was supplemented to children through breast milk and processed complementary food for a period 12 months but there was a significant effect on monthly changes in weight for length z-score and the intervention arms had significantly higher amount of RBC DHA, EPA, EPA+DHA (omega-3 index) and lower ratio of AA/DHA +EPA. The plasma fatty acid response is more rapid compared to the RBC fatty acid response in which there is a steady increase in the plasma level of the phospholipids for about four weeks and a steady state is reached. While the RBC level of these fatty acids takes about four to six months before it levels off after the supplementation is started and the steady state is maintained even if the supplementation is continued beyond (Arterburn et al., 2006). Supplementation with DHA compared to ALA or EPA brings about the highest levels this fatty acid in the plasma and RBC (Arterburn et al., 2006).

Despite clear theoretical mechanisms, we found no effect on common childhood infection and systemic inflammation in all intervention arms **(Chapter 6)**. The n-3 LC-PUFAs are involved in signal transduction, immune modulation via production of anti-inflammatory eicosanoids; while n-6 LC-PUFAs have pro-inflammatory characteristics through production of pro-inflammatory eicosanoids. Animal studies showed that DHA supplementation leads to drastic reduction in pro-inflammatory cytokine release through their effect on dendritic cells (DC) maturation by decreasing activation of nuclear factor κB(NF-κB). The latter is involved in the expression of pro-inflammatory genes including cytokines (IL-1, TNF-α) (Draper et al., 2011, Kong et al., 2010). However, there are no studies which unequivocally show the effect of n-3 LC-PUFA supplementation on amelioration systemic or gut inflammation as a primary outcome (Calder, 2009, Dewey and Mayers, 2011). Since, the existing evidences are neither to be rejected nor accepted unequivocally, there is a need for additional intervention studies using measures of inflammation as a primary outcome with adequate sample size.

In presence of deficiency of DHA and EPA and at the background of low n-6 fatty acids in the diet, humans can convert ALA to EPA and DHA in a good amount. However, typical diets both in the West and many developing countries contain higher amount of LA in relation to ALA or preformed n-3 fatty acids (EPA and DHA)(Arterburn et al., 2006, Michaelsen et al., 2011) which make the conversion of ALA to EPA and DHA minimal. Hence, direct intake of the latter fatty acids is crucial especially at critical ages in the life cycle (pregnancy, lactation, and early childhood). In infants the conversion is specifically very low and the local dietary pattern also complicate the conversion. Maize, sorghum and teff (*Eragrostis tef)* are predominantly consumed in our study areas. As maize and many other cereals are consumed after some decortication and refining, the crude fat is also reduced whereby reducing the content of n-3 and n-6 fatty acids (Baye, 2014). Teff is consumed without decortication as the grains are very small and the natural composition of essential nutrients including essential fatty acids is preserved. However, teff contains predominantly unsaturated fatty acids which include oleic acid (24%) and linoleicacids (44.2%) (Hundera, 1998) with the LA: ALA ratio of ~7:1 (Baye, 2014). When the ratio reaches 5 or above, the phospholipid in the liver are said to be rich in AA compared to EPA which in turn affects further conversion to DHA (Gibson et al., 2011). The deficiency of important micronutrients in the local food crops is reflected on human intake(Girma, 2016) in Ethiopia which in turn are implicated in decreased conversion of ALA to EPA and DHA. However, our supplementation trial addressed this by providing multiple micronutrients to all arms and this alone may not explain the lack of effect on the primary and secondary endpoints (**Chapter 6**).

7.6. Implications of findings of the research

Despite international commitment towards addressing undernutrition globally, significant number of countries from low resource settings are still failing to keep pace withimportant development goals including reduction of undernourishment and underweight. This has been attributed to the relatively slower reduction in poverty rate (proportion living on less than 1.90 USD) per day(The World Bank, 2015), growing income inequality within the population, recurrent conflict, erratic climatic conditions, environmental degradation or over exploitation (United Nations, 2015a). Even if the rate of underweight is cut by half since 1990, nearly 90 million children under the age of five years remain underweight and a third of those are found in sub-Saharan Africa (United Nations, 2015a) and currently the prevalence of stunting is still one of the highest in this region. The findings in our study and the recent EDHS report call for a concerted effort in addressing undernutrition during the most critical period of the life cycle.

Among the different interventions, IYCF has been pivotal in curbing undernutrition with its short and long term impacts. Very recently, nutrition has not been given due attention by the Ethiopian government as stipulated in the successive National Nutrition Programs (NNP) (FMoH/UNICEF/EU, 2016). Understanding the cross cutting nature of the problem, the government pursued a multisectoral approach in addressing nutrition in its 2013- 2015 National Nutrition Program (FDRE, 2013) involving Health, Education, Industry, Water and Energy, Trade, Agriculture, Labor and Social Affairs, Finance and Economic Development, and Women, Children, and Youth Affair. The latest NNP has already been endorsed and the “first 1000 days” has been one of the focus areas(FDRE, July, 2017) as a means of eliminating chronic undernutrition in Ethiopia by 2030 through the Seqota Declaration(Compact2025 [Ethiopia], 2016).

As stipulated in the Growth and Transformation Plan of the country (which is under implementation); one of the element is social sector development which is very much interrelated to achieving the nutritional goals (FDRE, May, 2016). Achieving a skilled, innovating and productive workforce requires a very good preparation from childhood through good nutrition and health care. The other social sector development plan encompasses the health sector which is considered an important driver of the growth and transformation agenda. Ethiopia has achieved so much in the health related MDG goals, however, the rate of undernutrition, maternal and child mortality rates are still high. In order to address this issues, it is important that the "first 1000 days" is targeted for a maximum benefit for the current and the generation to come. Moreover, addressing chronic malnutrition and hunger has also been incorporated directly under the Sustainable Development Goals (SDG target 2.2) (United Nations, 2015b).

It is known that investing in the complementary feeding period would contribute the lion's share in preventing chronic undernutrition and its short and long-term consequences in terms of overall health, educational attainment, and economic productivity. The validation of the existing IYCF indicators is; hence, an important contribution in the assessment, targeting and monitoring and evaluation of programs at population level. They provide quick information about the level of implementation of programs and projects targeting infants and young children at the population level.

Even if we were unable to demonstrate the association of feeding styles with child outcomes, we found that majority of the biological mothers in the rural setting practiced responsive feeding. Despite the lack of randomized trials which clearly isolate the effect of appropriate feeding styles and behaviors (responsive feeding), there is an increasing acknowledgement that such behaviors could enhance food acceptance, intake and whereby influencing growth and development of infants and young children (Bentley et al., 2011, Flax et al., 2013, Abebe et al., 2017, Baye et al., 2017). In settings where there is an ongoing food insecurity, behavior change interventions involving responsive feeding should be accompanied by nutrition specific interventions (e.g. good supplementation) (Aboud et al., 2009) for maximum outcome. At the same time, nutrition interventions which are focused on only food or feeding without considering the feeding context are most likely to be less effective as the feeding context is equally important as the provision of nutritious foods or practicing appropriate feeding (e.g. exclusive breast feeding, consumption of solid or semisolid foods, etc.).

The so called “gold standard” methods of assessment of micronutrient status rely on identifying and measuring biomarkers of those nutrients (in blood, urine, tissues, etc.) which cannot be performed by lower level practitioners and are time taking, expensive in terms of sample collection, transportation, and analysis. Such methods cannot be used in the community settings for monitoring and evaluation of nutrition programs aiming at alleviating micronutrient deficiencies. Therefore, there is a need to standardize the existing indicators of IYCF in line with their ability to predict changes in micronutrient status with improvement in the consumption of diversified foods. We demonstrated that simple count of food groups are associated with micronutrient intake (expressed in terms of MMDA). Even if different methods of accounting intake of foods is employed (for instance food groups, food items), such indicator was found to be robust in predicting dietary quality. In our case DDS was calculated with no restriction to amount of intake of a certain food item to be included in the food group which obviously decrease the specificity of DDS as indicator of nutrient adequacy or density; however, its practical significance should be considered as it is mostly challenging to estimate the portion size of the food items consumed. Imposing minimum to the counting of intake of food items or improving the portion size could increase the specificity and decrease the percentage of misclassification (Kennedy et al., 2007, Daniels et al., 2009, Moursi et al., 2008a) but it may not be pragmatic as pointed out by the respective researchers.

Dietary diversity score is especially crucial for the monitoring and evaluation of programs involved in a food based approaches and dietary diversification in order to address micronutrient deficiencies. Such approaches are said to be more sustainable and exert minimal burden on the health system on supplying micronutrient supplements and treating micronutrient deficiency disorders. They can be analyzed and interpreted at the decentralized level. DDS, however, may not be appropriate in situations where fortified products or micronutrient supplements are used in the prevention and treatment of micronutrient deficiencies among the target populations. In conclusion, even if we were not able to standardize all indicators of IYCF, they are very useful for assessment of complementary feeding practices in large scale national or regional surveys. They can address key dimensions of complementary feeding, and they can be useful for tracking the level of implementation of the National Nutrition Program in Ethiopia.

It is known that brain accretion of DHA occurs during the latter part of pregnancy and the first two years of life which coincides with the highest rate of physical growth and this is the period where much of the brain growth occurs compared to the rest of the stages in the life cycle. It has been recognized that the diet globally is changing and becoming more pronounced in the countries undergoing nutrition transition with consumption of vegetable oil with very high n-6: n-3 LC-PUFA ratio and low content of preformed EPA and DHA. It is then intuitive to assume that the tissue levels of n-3 LC-PUFAs would be low in settings where the diet is poor in n-3 LC-PUFAs or there is high intake of n-6 fatty acids. As a result, supplementation with preformed n-3 LC-PUFAs would be expected to support physical growth and development. However, we found no effect on linear growth even if there was an effect on ponderal growth in the intervention arms.

The evidence so far is not also supportive of an effect on linear growth upon supplementation with n-3 LC-PUFA; hence, longer follow-up starting from early age with relatively higher dose of preformed EPA and DHA has been suggested (Simmer et al., 2011, Delgado-Noguera et al., 2015). However, the quality of the ponderal increase in the intervention groups cannot be verified in our research as we didn't measure body composition. Even if there is an ongoing dispute, there is an evidence for an increase in fat mass or fat deposition associated with increased of n-6 LC-PUFA in relation to n-3 LC-PUFA consumption (Lapillonne and Carlson, 2001, Ailhaud et al., 2006, Ailhaud et al., 2007). An increase in MUAC and a relatively delayed onset of increased skin fold thickness has also been observed among infants supplemented with fish oil in The Gambian trial (van der Merwe et al., 2013)but the mechanism of such effect is not well elucidated. Systematic reviews of the effect of n-3 LCPUFA intake during pregnancy and postnatally on short and long-term effects on body composition of infants and young children (adiposity, BMI or body weight), did not produce unequivocal result(Muhlhausler et al., 2010, Rodríguez et al., 2012). Some of the reviewed trials showed an inverse association, some had no effect at all, and two studies where mothers were supplemented with n-LCPUFA during lactation resulted in higher BMI, adipose tissue and waist circumference of children at 2·5 and but such finding did not persist at the age of 7 years(Asserhøj et al., 2009).In a recent RCT, children born from mothers who were supplemented with DHA during the latter half of pregnancy were followed up at 3 and 5 years of age. After adjusting for a genetic predisposition to obesity, maternal intake of DHA did not affect the BMI or body composition of children at 3 or 5 years of age(Muhlhausler et al., 2016).Nevertheless, mechanisms for the observed direct association of perinatal n-3 LCPUFA supplementation and body composition during childhood are not understood.

Despite the lack of effect on primary endpoints, the blood levels of EPA and DHA in children increased significantly among the n-3 LC-PUFA supplemented arms in our study. Even if the biological functions of the EPA and DHA are well documented by many researchers, the lack of consistent effect on growth could be due to the variation in the diet consumed by the different populations including breast feeding mothers. It is widely documented that breast feeding is universal in Ethiopia and all of the children in our trial were breastfed, but the quality of fat in the breast milk is dependent on the maternal diet especially in terms of content of LA and ALA. Hence, the complete profile of the fat content of the local diet needs to be known to make a sound conclusion even if we provided the preformed n-3 LC-PUFAs for both the mothers and infants in the intervention arms. Low levels of LA has been given a lot of attention more than the ALA content when we are considering the food based approach in improving the EPA and DHA status(Wood et al., 2015). Keeping the LA low was associated with increasing level of EPA and DHA even if the ALA level was kept constant (Liou et al., 2007, Wood et al., 2014). Provision of preformed n-3 LC-PUFA is said to be unsustainable globally and consumption of low amount of LA should be given emphasis so that conversion of ALA to EPA and DHA from local foods can be enhanced (Gibson et al., 2011).

Ethiopia acquires most of its edible oil through an import and palm oil is the main imported oil over the past decade and the bulk of importation has grown rapidly over years and reached to 350, 000 metric tons per year in 2016 or an increase by 94% between 2010 and 2015(USDA (Foreign Agricultural Service), 2017). Palm oil, has a relatively high amount of LA and trace amount of ALA (with possible high LA:ALA ratio) (Godswill et al., 2016) which is less suitable for use for young children and breastfeeding women as the rate of conversion from ALA to EPA and DHA will be significantly affected as explained above. The risk of increased childhood adiposity with increasing n-6 to n-3 fatty acid ratio has also been reported by Ailhaud et al (2006, 2007) and at most attention should be given for the critical periods in the life cycle in terms of the type of oil or other fatty acid sources they are consuming (Ailhaud et al., 2006, Ailhaud et al., 2007).

Several researchers described the role of n-3 LC-PUFAs in inflammation and immune reactions among both animal studies and human subjects(Calder, 2009, Calder, 2010)through incorporation in the phospholipids of immune cells and the production of bioactive lipids known as eicosanoids. The incorporation of n-3 LCPUAs in the phospholipids of immune cells leads to inhibition of the eicosanoids (2-series prostaglandins and leukotrienes) generated by n-6 LC-PUFA (arachidonic acid). Relatively recently two lipid derivatives of EPA and DHA were found to be anti-inflammatory substances involved in resolution of inflammation. They are termed resolvins and protectins (Calder, 2013). Given extensive investigation into anti-inflammatory effect of EPA and DHA, not much is known about their role in the prevention of infectious morbidity and gut inflammation. In our analysis relatively smaller proportion of children were diagnosed with common childhood illness and systemic inflammation. As a result the effect of n-3 LC-PUFA could have been weakened and no effect was found. Furthermore, children in all arms received multiple micronutrients and deworming tables as well which might have contributed to the reduction in the morbidity and systemic inflammation. Hence, it is impossible to conclude about the impact of n-3 LC-PUFA on common childhood morbidity and systemic inflammation in our study.

7.7. Methodological considerations

A number of study limitations were mentioned corresponding to the papers in the previous chapters, the following methodological considerations/limitations are worth mentioning in this section.

Most of the studies conducted already to assess the feeding styles of caregivers are based on different questionnaires descried in the general introduction section. They are not adapted and validated in different ethno-cultural settings in Ethiopia. We used the locally adapted questionnaire based on the Caregiver’s Feeding Style Questionnaire. Relevant items were taken from the questionnaire and pretested on sub sample forunderstandability, cultural sensitivity and the amount of time it takes to administer. Direct observation should have also been made to capture the caregiver-child interaction for proper classification of the caregiver and child behavior during feeding. As it is now, there are different definitions and measurement methods used for the different assessment feeding styles. For instance, various questionnaires or direct observational methods are used to measure responsive feeding in interventions involving behavior change. The study did not assess the different outcomes of feediny styles (acceptance of food intake, mouthful of bites taken, energy intake, weight status, etc.) and such outcomes should also be standardized to increase their validity.

The aim of **chapter 3**was mainly to assess pattern of caregiver-child interaction in the context of feeding; however, we should have used the WHO indicators of IYCF strictly for our assessment breast feeding and complementary feeding practices of the caregivers. WHO indicators are used for the assessment of IYCF in the population and they are meant to capture the multidimensional and dynamic nature of infant and young child feeding. Moreover, they can be used to track changes in feeding practices in a population not only within a country but also across countries in a region or globally. The failure of use of such indicators reduces the comparability of the findings with other studies on various dimension of feeding practices.Moroever, the area where the feeding style assessment was made is a relatively small district in the southern Ethiopia where specific ethnic groups live. Based on the 2007 Census conducted by the Central Statistical Agencyof Ethiopia (CSA), there are eight ethnic groups in the woreda but the largest ones being Derashe (45%) andGawwada(28%) the rest account for the remaining percentages and half of them are protestant by religion(CSA [Ethiopia], 2008). The finding in this district might not be generalizable to other populations in Ethiopia as parenting or feeding style is ethno-cultural in nature. Hence, the findings should be used with such limitations taken into consideration.

In our study, we were not able to assess child outcomes in relation to the different feeding styles adopted by mothers or caregivers. Moreover, temporality between caregiver feeding style and child outcomes (with regards to child food acceptance or intake, growth or development) are difficult to ascertain. Hence, in order to isolate its effect a longitudinal study which start early in infancy should be undertaken using responsive feeding (supposedly beneficial feeding style) as one intervention arm.

As it has been mentioned elsewhere, complementary feeding behaviors are multidimensional in nature, they change with age of the childand complement breast milk intake (which is likely to be variable) and the effect on child outcomes are considered to be cumulative. However, most studies including ours assessed the cross sectional association of indicators of complementary feeding with child outcomes. As a result of difficulty of ascertaining temporality, longitudinal measurement of feeding indicators and child outcomes should be the way forward.

The discussion about the limitations of the dietary diversity is still going on. It has been found in our study and elsewhere that DDS can serve as a proxy for dietary quality expressed in term of the different quality measures at the population level (not at individual or household level). However, defining the minimum quantity of food items to be counted as consumed in the score is still pragmatically challenging. The relationship between food group diversity and dietary quality were stronger when small quantities were not counted as consumed (Kennedy et al., 2007) but not all studies had the same observation and it may not work in many low income countries where the diet of the population is not well diversified (Moursi et al., 2008a). The issue of counting ingredients in mixed dishes is critical and there is no easy and practical solution to it if we have to make the collection of information concerning the food consumption over a period of time as easy as possible. One of the other limitations of the problem of the use of DDS is the difficulty to deal with enriched foods consumed by the study subjects. Even if we haven't found any difference in terms of the average number of food groups consumed by the study children, seasonality affects the availability and the type of foods consumed by the population; which in turn affect the dietary quality for each DDS. Hence, care must be taken in making conclusion based on dietary diversity measured in different seasons. Calculation of dietary diversity could be context specific (a number of food groups determined for one setting may not necessary work everywhere). It depends on availability, frequency of consumption, and nutritional composition. Moreover, species of a certain cereal or legumes could vary from place to place and so does their nutrient composition. This and other findings highlight the need to operationalize the use of simple food group based indicators like DDS to specific situations rather than generalizing to all low income settings.

We employed list-based questionnaire to gather information from the caregiver about intake of food items which might sometimes be incomplete as the child might have consumed other foods not included in the predefined list. There are no studies however showing the comparison between the list-based or open recall methods in terms of capturing all the foods/food groups consumed by the child over the previous day/days. For the quantitative 24 hour recall, we employed an open recall technique and the caregivers were given the chance to recall all the foods, food groups, recipes or drinks given or consumed by the child. We also used a different food intake data for DDS and

MMDA calculation so that we avoided a higher chance of correlation which is mainly due to the fact that the same data is used to create both.

The food composition table prepared for use in Ethiopia lack the composition of some foods items which are locally consumed; as a result we had to impute or use values from analysis papers. The Ethiopian food composition tables have values for raw and as well as cooked foods and it was possible to directly convert food intake to nutrient intake. Hence, there was no need to do conversion for those foods reported as cooked in which case it is expected that there are some nutrient loses. The calculation of nutrient intake from food intake was made by a Lucille Food Intake Program developed by the Department of Food Safety and Quality Ghent University (DFSFQ, n.d); hence, there was no chance for introducing errors in the process. However, we had some food items for which there was missing nutrient values on the Ethiopian food composition tables and we had to input from those values from the same food item in another food composition table based on previously suggested formula (Gibson and Ferguson, 2008). In so doing, adjustment was made for different moisture content of foods or whether the food was cooked, raw or fresh (Gibson and Ferguson, 2008). We used analysis paper from relatively similar area in the southern region (Abebe et al., 2007) to be complete. Care has been taken for mixed dishes by gathering recipe information from the study mothers/caregivers from the study site and record ingredients and their corresponding weight while preparing the food. Those weights are used in the calculation of nutrient intake from the mixed complementary food. Due to lack of up-to-date and complete food composition table, we used different food composition tables and a recipe book which might have used different analytical techniques to determine the nutrient content of the foods/recipes which potentially introduce bias into the nutrient intake data. In line with analytical techniques used, we haven't made individual scrutiny to ensure that currently acceptable techniques are employed to determine nutrient values. Moreover, as a result of variation in the nutrient composition of foods based on geography, a locally developed food composition tables should have been used to avoid systematic error.

The validity of the interactive 24 hours dietary recall method could be affected by the lack of prior validation on similar population groups and as a result there is a potential for the introduction of systematic error (Gibson and Ferguson, 2008); however, maximum effort has been made to training the caregivers on portion size estimation, distribution of measurement materials from the research team, calibrating and standardization of household measures and advising the mothers/caregivers to use those martials during the survey period, using pictures of local food items and fruits to facilitate portion size estimation.

One of the dimensions we haven't captured in our analysis was the association of DDS with household level food security. As mentioned elsewhere, we found no significant difference in DDS between the HS and PHS even if there is a clear difference in term of agricultural production in the two seasons. There could be other sources whereby households offset the seasonality of agricultural production. Moreover, the intra household distribution in terms of age and gender should have been captured to see if there is buffering of children during lean season.

We collected the information for the construction of infant and child feeding index from different components of feeding practices obtained from brief recall techniques. Such index has its own limitations in terms of its association with different child outcomes. As a result, measurement of such feeding practice indicators longitudinally from the same children and constructing a longitudinal IYCF index has been recommended.

We provided a moderate amount of EPA and DHA (500mg in total) for mothers only and children only arms and around 1000mg for the intervention arm where both mothers and infants were grouped. The study subjects were followed for a period of around 12 months starting from 6 month of age and the sample size was adequately powered to at least detect the difference in the primary endpoint (linear growth).In low income countries environmental enteropathy is common starting from as early as 3 months of age (Lunn et al., 1991) and it was associated with linear growth retardation(Campbell et al., 2002, Campbell et al., 2003). Hence, there could be a benefit from starting with additional n-3 LC-PUFA very early either through higher dose of maternal supplementation or direct consumption in a formula food. Even if the sample was not calculated taking differences in gut permeability into consideration, measuring gut permeability directly would have provided an important information regarding the benefits of supplementation with n-3 LC-PUFA.

A Cochrane review showed that there was no effect of LC-PUFA supplementation on physical growth expressed in terms of weight, length and head circumference. The conclusion made was that there is no sufficient evidence for the routine supplementation of term infants with LC-PUFA on physical growth (Simmer et al., 2011). The sample size, dose of DHA/AA, duration of supplementation, and type of fatty acid supplemented were some of the reason mentioned. However, there was no effect even if children were followed up till the age of 18 months. Length, weight, and head circumference did not differ between intervention and control groups after supplementation during pregnancy and the first four months after birth with LC-PUFA(Delgado-Noguera et al., 2015). Even if the studies reviewed had significant limitations in terms of power and sample size to detect differences between groups, LC-PUFAs during pregnancy prevented preterm delivery and increased head circumference (Szajewska et al., 2006, Makrides et al., 2006). The role of the latter finding in terms of later growth outcomes is thus difficult to be ascertained. All the above reviews were also made based on studies conducted in high income countries.

7.8. Conclusion

Undernutrition during early childhood remains the single most important public health problem in low income countries and the plight might have started during the prenatal period. The problem is compounded by the emergence of overweight and obesity in those countries undergoing nutrition transition and in affluent areas of developing countries. Apart from the short term effects, undernutrition is associated with poor development, work performance and overall socioeconomic productivity during adulthood. The rise of chronic diet related diseases in low and middle income countries has been explained by the early undernutrition. The prevalence of both linear growth restriction and low ponderal growth in our study area are similar to other settings in Ethiopia.

Among others, poor IYCF practices contribute a significant share in the causation of undernutrition. Programs aimed at addressing early child undernutrition require monitoring and evaluation tools which can capture the extent of implementation and outcomes among the target populations. Before implementing those local or large scale programs, knowledge about care practices pertaining to feeding and the extent of compliance to the infant and young child recommendation is crucial. Moreover, the existence of locally adapted and validated indicators for assessment and evaluation of IYCF practices have paramount importance in keeping track of the changes in the dietary practices and nutrient intake. We demonstrated the possible role of simple food group based indicators as an important tool in the assessment and monitoring and evaluations of programs aimed at improving nutrient intake. Such indicators were also assessed for their ability to predict micronutrient intake and their stability across two distinct agricultural seasons. To address the multidimensional nature of feeding, we constructed a composite index and its association with child growth across seasons was evaluated.

It has been suggested that essential fatty acids are lacking in complementary foods in developing countries and little is known regarding the role of essential fatty acids in the diet and hence we demonstrated the effect of supplementation with n-3 LC-PUFA through breast milk and enriched complementary foods on child growth, morbidity, and systemic inflammation.

Based on the findings this Ph.D. study the following conclusions can be made:

There is a dearth of information regarding the pattern of interactionbetween caregivers and their children in different ethno-cultural contexts in Ethiopia. For such researches to be conducted, there is an urgent need to adapt and locally validate a tool to measure the different parental/caregiver feeding styles. Responsive feeding was found to be the commonest feeding style among the rural caregivers in the study area but such finding may not be generalizable to all similar rural regions in Ethiopia.

A simple indicator developed from collection of information regarding food group consumption can be used as an important tool to assess micronutrient intake in setting where there is high burden of micronutrient deficiency. The usability of DDS for predicating “higher intake” of micronutrients is not known as most of the children were taking suboptimal amount of essential micronutrients.

We found no significant difference in terms of the mean DDS between HS and PHS and DDS remains associated with measure of nutrient intake showing that the food group indicator can be useful in spite of a likely seasonality in food intake and availability. Dietary diversity score at HS was also associated with linear growth at follow up (six months later) in PHS after controlling all measured confounding factors.

The use of composite index has been recommended to encompass various dimension of feeding. There was no significant difference in the tertiles of the index for infants in two seasons. However, the ICFI was not associated with child growth in both seasons.The use of longitudinal ICFI and including more dimensions such as food consistency, care practices into the construction of the index could mitigate the lack of the association with child outcomes.

Despite the growing interest in the addition of essential fatty acids in the diet or through supplementation to support linear or ponderal growth, prevent or ameliorate gut inflammation (environmental enteropathy) or systemic inflammation, supplementation of children with n-3 LC-PUFA through breast milk and additional supplementary food did not affect linear growth. Even if it is known with considerable certainty that essential fatty acid intake was small among children in the study area and the hygienic condition is expected to be suboptimal, the intervention did not bring about a difference in common childhood infection and systemic inflammation.

7.9. Recommendations for policy and further research

The return on investment by intervening during the critical period of the first 2 years is very high. Addressing problems related to complementary feeding contributes significantly in the reduction of the level of chronic undernutrition and its implication for health, education, socioeconomic development of the country. Moreover, lessons should be learnt from the missed opportunities during the implementation of the MDGs in addressing the level of chronic undernutrition through nutrition specific and nutrition sensitive interventions. Based on the findings of this Ph.D. work, the following policy issues and further research areas are recommended.

7.9.1. ***Recommendations for policy***

- As the world is starting to implement the SDGs, it is important to consider nutrition as both an input and output of most of the development goals adopted by the countries. Nutrition is mentioned directly under SDG2; however, most of the development goals are either affected by nutrition or are the consequences of achievement of good nutrition. Preventing linear growth faltering (stunting) starting from early life through addressing the complementary feeding practices is crucial for achievement of the rest of the development goals in a long term.
- Incorporate indicators of IYCF in the national information management system so that changes in the level of compliance to feeding recommendations, energy and micronutrient intake can be estimated and tracked for timely action.
- It is observed that children consume very few food groups irrespective of season. Enhancing the dietary diversity should be given emphasis if intake of essential

nutrients is to be adequate. Improving nutrition relevant agricultural biodiversity should be the way forward as it has already been associated with increased consumption or availability of diversified foods and nutritional status in previous studies. Hence, in order to offset the effect of season on food intake and nutritional status, cultivation of diverse and nutritionally meaningful crops combined with improving market access and integration should be considered without displacing nutritionally important local foods.

- Despite the lack of effect of n-3 LC-PUFA supplementation on linear growth, addressing the essential fatty acid intake during the complementary feeding period should not wait another longitudinal study in countries like Ethiopia where the supply and intake of these fatty acid is much less than the current recommendation for daily intake for children in the first two years. Exploration of local sources of n-3 fatty acids and reducing intake of n-6 sources from the diet should be underscored in this age group.

7.9.2. ***Recommendationsfor further research***

- There are reports of feeding styles and nutritional outcomes in few developing countries both from cross sectional and observational studies; hence, there is a need to further look at feeding styles and their effect on food acceptance/intake, nutritional or health status using a longitudinal design.

- Responsive feeding has been incorporated in the recommendation for appropriate complementary feeding by various governments or organizations. However, there are limited prospective studies which evaluated the effect on dietary intake, growth, and psychosocial development by using responsive feeding as one intervention arm. Moreover, most of the follow up studies assume a unidirectional relationship and do not give any information about the direction of the relationship between the feeding style and the child outcomes in which the degree of food acceptance or eating by the child could lead to responsive feeding behavior or the vice versa. Hence, this reciprocal nature of the relationship should be addressed and a more objective methods of observation should be incorporated (e.g. videotaping the child-mother interaction in non-intrusive way).

- The child feeding style or questionnaire used in our study has not been validated in our country and there needs to be proper adaptation and validation of a locally suitable instrument based on the principles of appropriate parenting in relation to feeding (feeding style). As parental or caregivers feeding styles are part of the ethno-cultural practices in childrearing, further adaptation of an instrument that fits to a given community or group is needed so that the instrument captures the actual practice.

- The development of indicators for IYCF was useful for various programs aimed at improvement of IYCF practices to document their progress and outcomes. Apart from DDS, the other indicators of IYCF have not been validated and local adaptation should be made so that we make sure that they measure what they are intended to. Almost a decade after they were published, there are only few validation studies done but only for DDS as indicator of dietary quality and nutritional status. Hence, the remaining ones need to be validated against a gold standard or specific child outcome.
- Revisiting the composite index of IYCF is needed as it was not associated with child outcomes in our study and in much of the studies conducted elsewhere. Its validity after incorporating other dimensions of child feeding or care practice should be evaluated. There is also an emerging problem of overweight and obesity in low and middle income countries, hence, IYCF should include indicators related to the prevention of overweight or obesity. Moreover, prospective measurement of feeding practices and other components of ICFI is important to capture the dynamic nature of feeding and usual practices.
- The association of DDS and dietary quality were not affected by season, but all seasons in the area are not covered by this study. Households or individual caregivers are expected to have a relatively greater disposable income in the post-harvest season as compared to the pre-harvest season and that could affect the diversity of the diet provided to their children. Moreover, agricultural seasons and the types of food crops grown vary from place to place in Ethiopia. Hence, the stability and performance of DDS as indicator of dietary quality should be investigated in various agro-ecologies and seasons.
- Most of the food consumption surveys, including ours, use qualitative 24 hour dietary recall to estimate food intake and calculate DDS. There is a need to validate the

qualitative methods using a food group diversity indicators derived from detailed quantitative dietary intake surveys.

- Accounting intake of nutrients from breast milk based on “average intake” of breast milk by similar population in other countries might introduce bias. Such values should be estimated based on breast milk intake values from the same population. The same applies for the assumption of energy density in the presence of variation in nutrient density of locally available foods used for complementary feeding.
- There is a need to prepare a food composition table which includes local foods and recipes, and the bioavailability of some critical nutrients (e.g. iron, zinc, calcium) needs to be known to make sure that the nutrient intake and density adequacies we calculate represent the local situation. We employed the nationally prepared food composition table with some micronutrients not analyzed and with a lot of missing values.
- The food composition table for use in Ethiopia should be critically updated considering the lack of composition forsome micronutrients, saturated, mono unsaturated, and polyunsaturated fat fractions, fatty acids (including essential fatty acids) and phytate. Attention should also be given to missing values of some nutrients, missing foods, composition of fortified staples and traditional foods and recipes. The FAO/International Network of Food Data System (FAO/INFOODS) exercise in West Africa should also be replicated in the Eastern African region in which a standardized food composition table is developed based of list of staples, traditional foods, recipes, fermented and fortified products. Such effort requires the involvement of the policy makers, research institutions, the agriculture sector and non-governmental organization working on food composition databases such as the one mentioned above. Regional collaboration, capacity building regarding generation of high quality food composition databases and awareness creation concerning its importance should be underscored.
- Our research did not assess the contamination of local food used for complementary feedingin relation to child growth or other outcomes. Previous study based on maize sample from the subsample of the households of the same population concluded that the daily intake of Dichlorodiphenyltrichloroethane (DDT) was above the recommendations (Mekonen et al., 2015). Moreover, given the storage practices by

the farmers in the study area, mycotoxins contamination of the maize commonly used for complementary feeding is expected to be high even if we were not able to quantity it and considered a missed opportunity in this research. Among other detrimental environmental hazards, mycotoxins are implicated in growth faltering among young children. Subsequent research should assess the level of contamination of complementary foods with mycotoxins and their effect on health and nutritional status of infants and young children.

- There is consistent assertion that n-3 LC-PUFAs are important in child growth, development and amelioration of intestinal inflammation (environmental enteropathy); hence, there should be replication of similar supplementation trial with higher dose of EPA and DHA in which supplementation is started early through maternal route (supplementation during pregnancy or lactation). Adequate sample size should be used to identify differences with age, gender, socioeconomic status and hygiene status to ensure adequate statistical power. Moreover, the studies should have better compliance, adequate blinding of participants and investigators and with very low attrition rates.
- The later part of pregnancy and the first two years of life are characterized by "brain growth spurt" which requires adequate amount of brain-selective nutrients such as preformed n-3 LC-PUFAs (e.g.DHA). Several studies show this period is strongly associated with significant DHA accretion. However, the effect of both prenatal and postnatal LCPUFA supplementation on child development are rather inconsistent and much of the evidences are coming from developed countries and no or little is known in low-income countries. Hence,determining the effect of n-3 LC-PUFA supplementation on child development measured with a specific test battery is recommended to generate evidence in settings like ours.
- Environmental enteropathy has been associated with linear growth faltering; however, we did not directly measure gut permeability and there is only one n-3 LC-PUFAs supplementation study which used measures of gut inflammation as an outcome in Africa. Hence, a randomized trial involving the promotion of hygienic practices and n-3 LC-PUFAs supplementation with direct measurement of gut permeability could provide a valuable evidence for future programmatic interventions aiming at improving child growth.

References

AAP (Committee on Nutrition) (1988) Clinical Testing Of Infant Formulas With Respect To Nutritional Suitability for Term Infants; Task Force on Clinical Testing of Infant Formulas. Available at : https://wayback.archive-it.org/7993/20161022111601/http://www.fda.gov/Food/GuidanceRegulation/GuidanceDocumentsRegulatoryInformation/InfantFormula/ucm170649.htm.

Abay K. & Hirvonen K. (2016) Does market access mitigate the impact of seasonality on child growth? Panel data evidence from northern Ethiopia. *The Journal of Development Studies*, 1-16.

Abebe Y., Bogale A., Hambidge K.M., Stoecker B.J., Bailey K. & Gibson R.S. (2007) Phytate, zinc, iron and calcium content of selected raw and prepared foods consumed in rural Sidama, Southern Ethiopia, and implications for bioavailability. *Journal of Food Composition and Analysis***20,** 161-168.

Abebe Z., Haki G.D. & Baye K. (2017) Child feeding style is associated with food intake and linear growth in rural Ethiopia. *Appetite***116,** 132-138.

Aboud F.E. & Alemu T. (1995) Nutrition, maternal responsiveness and mental development of Ethiopian children. *Social Science & Medicine***41,** 725-732.

Aboud F.E., Moore A.C. & Akhter S. (2008) Effectiveness of a community-based responsive feeding programme in rural Bangladesh: a cluster randomized field trial. *Maternal & Child Nutrition***4,** 275-286.

Aboud F.E., Shafique S. & Akhter S. (2009) A Responsive Feeding Intervention Increases Children's Self-Feeding and Maternal Responsiveness but Not Weight Gain. *The Journal of Nutrition***139,** 1738-1743.

Abu-Saad K. & Fraser D. (2010) Maternal nutrition and birth outcomes. *Epidemiologic reviews***32,** 5-25.

Adair L.S., Fall C.H.D., Osmond C., Stein A.D., Martorell R., Ramirez-Zea M., et al. (2013) Associations of linear growth and relative weight gain during early life with adult health and human capital in countries of low and middle income: findings from five birth cohort studies. *The Lancet***382,** 525-534.

Addo O.Y., Stein A.D., Fall C.H., Gigante D.P., Guntupalli A.M., Horta B.L., et al. (2013) Maternal Height and Child Growth Patterns. *The Journal of Pediatrics***163,** 549-554.e541.

Adu-Afarwuah S., Lartey A., Brown K.H., Zlotkin S., Briend A. & Dewey K.G. (2007) Randomized comparison of 3 types of micronutrient supplements for home fortification of complementary foods in Ghana: effects on growth and motor development. *The American Journal of Clinical Nutrition* **86,** 412-420.

Agosti M., Tandoi F., Morlacchi L. & Bossi A. (2017) Nutritional and metabolic programming during the first thousand days of life. *La Pediatria Medica e Chirurgica* **39**.

Agostoni C., Giovannini M., Sala D., Usuelli M., Livio L., Francescato G., et al. (2007) Double-blind, placebo-controlled trial comparing effects of supplementation of two micronutrient sprinkles on fatty acid status in Cambodian infants. *Journal of Pediatric Gastroenterology and Nutrition* **44,** 136-142.

Ahmed T., Auble D., Berkley J.A., Black R., Ahern P.P., Hossain M., et al. (2014) An evolving perspective about the origins of childhood undernutrition and nutritional interventions that includes the gut microbiome. *Annals of the New York Academy of Sciences* **1332,** 22-38.

Ahmed T., Mahfuz M., Ireen S., Ahmed A.M.S., Rahman S., Islam M.M., et al. (2012) Nutrition of Children and Women in Bangladesh: Trends and Directions for the Future. *Journal of Health Population and Nutrition* **30**.

Ailhaud G., Massiera F., Alessandri J.-M. & Guesnet P. (2007) Fatty acid composition as an early determinant of childhood obesity. *Genes & Nutrition* **2,** 39-40.

Ailhaud G., Massiera F., Weill P., Legrand P., Alessandri J.-M. & Guesnet P. (2006) Temporal changes in dietary fats: Role of n−6 polyunsaturated fatty acids in excessive adipose tissue development and relationship to obesity. *Progress in lipid research* **45,** 203-236.

Alder E.M., Williams F.L., Anderson A.S., Forsyth S., Florey C.d.V. & Van der Velde P. (2004) What influences the timing of the introduction of solid food to infants? *British Journal of Nutrition* **92,** 527-531.

Anderson M. & Fritsche K.L. (2002) (n-3) Fatty Acids and Infectious Disease Resistance. *The Journal of Nutrition* **132,** 3566-3576.

Andrew Sunil Rajkumar, Christopher Gaukler & Jessica Tilahun (2012) Combating malnutrition in Ethiopia : an evidence-based approach for sustained results ; Africa human development series , The World Bank Washington DC.

Arabi M., Frongillo E.A., Avula R. & Mangasaryan N. (2012) Infant and Young Child Feeding in Developing Countries. *Child development* **83,** 32-45.

Arimond M. & Ruel M. (2004) Dietary diversity is associated with child nutritional status: evidence from 11 demographic and health surveys. *J Nutr***134,** 2579 - 2585.

Arimond M. & Ruel M.T. (2002a) Assessing care: progress towards the measurement of selected childcare and feeding practices, and implications for programs. *Food And Nutrition Technical Assistance*.

Arimond M. & Ruel M.T. (2002b) Progress in developing an infant and child feeding index: an example using the Ethiopia Demographic and Health Survey 2000. Discussion Paper.

Arimond M. & Ruel M.T. (2003a) Generating indicators of appropriate feeding of children through 23 months from the KPC 2000+. Food and Nutrition Technical Assistance Project, FANTA:Washington, DC.

Arimond M. & Ruel M.T. (2003b) Measureing childcare practices: approahes, indicators and implication for programs. *Food And Nutrition Technical Assistance*.

Arimond M., Zeilani M., Jungjohann S., Brown K.H., Ashorn P., Allen L.H., et al. (2015) Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. *Maternal & Child Nutrition***11,** 31-61.

Arsenault J.E., Nikiema L., Allemand P., Ayassou K.A., Lanou H., Moursi M., et al. (2014) Seasonal differences in food and nutrient intakes among young children and their mothers in rural Burkina Faso. *Journal of Nutritional Science***3,** e55.

Arsenault J.E., Yakes E.A., Islam M.M., Hossain M.B., Ahmed T., Hotz C., et al. (2013) Very Low Adequacy of Micronutrient Intakes by Young Children and Women in Rural Bangladesh Is Primarily Explained by Low Food Intake and Limited Diversity. *The Journal of Nutrition***143,** 197-203.

Arterburn L.M., Hall E.B. & Oken H. (2006) Distribution, interconversion, and dose response of n−3 fatty acids in humans. *The American Journal of Clinical Nutrition***83,** S1467-1476S.

Asserhøj M., Nehammer S., Matthiessen J., Michaelsen K.F. & Lauritzen L. (2009) Maternal Fish Oil Supplementation during Lactation May Adversely Affect Long-Term Blood Pressure, Energy Intake, and Physical Activity of 7-Year-Old Boys. *The Journal of Nutrition***139,** 298-304.

Assis A., Barreto M., Santos L., Fiaccone R. & da Silva Gomes G. (2005) Growth faltering in childhood related to diarrhea: a longitudinal community based study. *European Journal of Clinical Nutrition***59,** 1317.

Barceló-Coblijn G. & Murphy E.J. (2009) Alpha-linolenic acid and its conversion to longer chain n− 3 fatty acids: Benefits for human health and a role in maintaining tissue n− 3 fatty acid levels. *Progress in lipid research* **48,** 355-374.

Baye K. (2014) *Teff: nutrient composition and health benefits.* Intl Food Policy Res Inst.

Baye K., Tariku A. & Mouquet-Rivier C. (2017) Caregiver–infant's feeding behaviours are associated with energy intake of 9-11 month-old infants in rural Ethiopia. *Maternal & Child Nutrition*, e12487-n/a.

Bentley M., Caulfield L., Torun B., Schroeder D. & Hurtado E. (1992) Maternal feeding behavior and child appetite during acute diarrhea and subsequent health in Guatemala. *FASEB J* **6**.

Bentley M., Stallings R.Y., Fukumoto M. & Elder J. (1991) Maternal feeding behavior and child acceptance of food during diarrhea episodes, convalescence, and health in the Central NorthernSierra of Peru. *Am J Public Health* **83**.

Bentley M.E., Wasser H.M. & Creed-Kanashiro H.M. (2011) Responsive Feeding and Child Undernutrition in Low- and Middle-Income Countries. *The Journal of Nutrition* **141,** 502-507.

Berndt S.I., Gustafsson S., Mägi R., Ganna A., Wheeler E., Feitosa M.F., et al. (2013) Genome-wide meta-analysis identifies 11 new loci for anthropometric traits and provides insights into genetic architecture. *Nature genetics* **45,** 501-512.

Berti P.R. & Jones A.D. (2013) Biodiversity's contribution to dietary diversity. *Diversifying food and diets: using agricultural biodiversity to improve nutrition and health*, 400.

Bhutta Z.A. (2013) Early nutrition and adult outcomes: pieces of the puzzle. *The Lancet* **382,** 486-487.

Bhutta Z.A., Ahmed T. & Black R.E. (2008) What works? Interventions for maternal and child undernutrition and survival. *Lancet* **371**.

Bhutta Z.A., Das J.K., Rizvi A., Gaffey M.F., Walker N., Horton S., et al. (2013) Evidence-based interventions for improvement of maternal and child nutrition: what can be done and at what cost? *The Lancet* **382,** 452-477.

Bhutta Z.A. & Salam R.A. (2012) Global Nutrition Epidemiology and Trends. *Annals of Nutrition and Metabolism* **61(suppl 1),** 19-27.

Birch E.E., Khoury J.C., Berseth C.L., Castaneda Y.S., Couch J.M., Bean J., et al. (2010) The impact of early nutrition on incidence of allergic manifestations and common respiratory illnesses in children. *J Pediatr* **156**.

Birch L.L. (1998) Psychological Influences on the Childhood Diet. *The Journal of Nutrition* **128,** 407S-410S.

Birch L.L. & Fisher J.A. (1995) Appetite and eating behavior in children. *PediatrClin N Am* **42**.

Birch L.L. & Fisher J.O. (2000) Mothers' child-feeding practices influence daughters' eating and weight. *Am J ClinNutr* **71**.

Black R., Allen L., Bhutta Z., Caulfield L., de Onis M. & Ezzati M. (2008) Maternal and child undernutrition: global and regional exposures and health consequences. *Lancet* **371,** 243 - 260.

Black R.E., Victora C.G., Walker S.P., Bhutta Z.A., Christian P., de Onis M., et al. (2013) Maternal and child undernutrition and overweight in low-income and middle-income countries. *The Lancet* **382,** 427-451.

Bland R.M., Rollins N.C., Solarsh G., Van den Broeck J. & Coovadia H.M. (2003) Maternal recall of exclusive breast feeding duration. *Archives of Disease in Childhood* **88,** 778.

Bork K., Cames C.c., Barigou S., Cournil A. & Diallo A. (2012) A Summary Index of Feeding Practices Is Positively Associated with Height-for-Age, but Only Marginally with Linear Growth, in Rural Senegalese Infants and Toddlers. *The Journal of Nutrition* **142,** 1116-1122.

Bredenkamp C., Buisman L.R. & Van de Poel E. (2014) Persistent inequalities in child undernutrition: evidence from 80 countries, from 1990 to today. *International Journal of Epidemiology* **43,** 1328-1335.

Brenna J.T., Akomo P., Bahwere P., Berkley J.A., Calder P.C., Jones K.D., et al. (2015) Balancing omega-6 and omega-3 fatty acids in ready-to-use therapeutic foods (RUTF). *BMC Medicine* **13,** 1-4.

Brenna J.T., Varamini B., Jensen R.G., Diersen-Schade D.A., Boettcher J.A. & Arterburn L.M. (2007) Docosahexaenoic and arachidonic acid concentrations in human breast milk worldwide. *The American Journal of Clinical Nutrition* **85,** 1457-1464.

Briend A., Darmon N., Ferguson E. & Erhardt J.G. (2003) Linear programming: a mathematical tool for analyzing and optimizing children's diets during the complementary feeding period. *J Pediatr Gastroenterol Nutr* **36,** 12-22.

Briend A., Dewey K.G. & Reinhart G.A. (2011) Fatty acid status in early life in low-income countries–overview of the situation, policy and research priorities. *Maternal & Child Nutrition* **7,** 141-148.

Brown K.H. (1997) Complementary feeding in developing countries: factors affecting energy intake. *Proc Nutr Soc* **56,** 139-148.

Brown K.H., Dickin K.L., Bentley M.E., Oni G.A., Obassajju V.T., Esrey S.A., et al. (1988) Consumption of weaning foods from fermented cereals: Kwara State, Nigeria. InImproving Young Child Feeding in Eastern and SouthernAfrican Household-level Food Technology. In: Proceedings of a Workshop, pp 181-197 [D Alnwick, S Moses and OG Schmidt, editors] Ottawa, Ont: IDR.

Bryce J., Coitinho D., Darnton-Hill I., Pelletier D. & Pinstrup-Andersen P. (2008) Maternal and child undernutrition: effective action at national level. *Lancet* **371,** 510-526.

Butte N.F., Lopez-Alarcon M.G. & Garza C. (2002) Nutrient adequacy of exclusive breastfeeding for the term infant during the first six months of life.

Butte N.F., Wong W.W., Hopkinson J.M., Heinz C.J., Mehta N.R. & Smith E.O. (2000) Energy requirements derived from total energy expenditure and energy deposition during the first 2 y of life. *Am J Clin Nutr* **72,** 1558-1569.

C E West, F Pepping, C R Temalilwa & editors (1988) The composition of foods commonly eaten in East Africa ; Wageningen Agricultural University.

Calder P.C. (2002) Dietary modification of inflammation with lipids. *Proceedings of the Nutrition Society* **61,** 345-358.

Calder P.C. (2006) Polyunsaturated fatty acids and inflammation. *Prostaglandins, leukotrienes and essential fatty acids* **75,** 197-202.

Calder P.C. (2008) Polyunsaturated fatty acids, inflammatory processes and inflammatory bowel diseases. *Molecular nutrition & food research* **52,** 885-897.

Calder P.C. (2009) Polyunsaturated fatty acids and inflammatory processes: New twists in an old tale. *Biochimie* **91,** 791-795.

Calder P.C. (2010) Omega-3 fatty acids and inflammatory processes. *Nutrients* **2**.

Calder P.C. (2013) n-3 fatty acids, inflammation and immunity: new mechanisms to explain old actions. *Proc Nutr Soc* **72**.

Campbell D., Elia M. & Lunn P. (2003) Growth faltering in rural Gambian infants is associated with impaired small intestinal barrier function, leading to endotoxemia and systemic inflammation. *The Journal of Nutrition* **133,** 1332-1338.

Campbell D.I., Lunn P.G. & Elia M. (2002) Age-related association of small intestinal mucosal enteropathy with nutritional status in rural Gambian children. *British Journal of Nutrition* **88,** 499-505.

Campoy C., Escolano-Margarit M.V., Anjos T., Szajewska H. & Uauy R. (2012) Omega 3 fatty acids on child growth, visual acuity and neurodevelopment. *British Journal of Nutrition***107,** S85-S106.

Checkley W., Buckley G., Gilman R.H., Assis A.M., Guerrant R.L., Morris S.S., et al. (2008) Multi-country analysis of the effects of diarrhoea on childhood stunting. *International Journal of Epidemiology***37,** 816-830.

Christian P., Lee S.E., Donahue Angel M., Adair L.S., Arifeen S.E., Ashorn P., et al. (2013) Risk of childhood undernutrition related to small-for-gestational age and preterm birth in low- and middle-income countries. *International Journal of Epidemiology***42,** 1340-1355.

Clark H.R., Goyder E., Bissell P., Blank L. & Peters J. (2007) How do parents' child-feeding behaviours influence child weight? Implications for childhood obesity policy. *Journal of Public Health***29,** 132-141.

Cogill B. (2003) *Anthropometric indicators measurement guide.* Food and Nutrition Technical Assistance Project, Academy for Educational Development, Washington, D.C.

Coly A., Milet J., Diallo A., Ndiaye T., Benefice E. & Simondon F. (2006) Preschool stunting, adolescent migration, catch-up growth, and adult height in young senegalese men and women of rural origin. *J Nutr***136,** 2412 - 2420.

Compact2025 [Ethiopia] (2016) Ending Hunger & Undernutrition Challenges & Opportunities; Draft Scoping Report for Roundtable Discussion , March 2016.

Crane R.J., Jones K.D.J. & Berkley J.A. (2015) Environmental enteric dysfunction: An overview. *Food and Nutrition Bulletin***36,** S76-S87.

Crouch P., O'Dea J.A. & Battisti R. (2007) Child feeding practices and perceptions of childhood overweight and childhood obesity risk among mothers of preschool children. *Nutrition & Dietetics***64,** 151-158.

CSA [Ethiopia] (2008) Summary and statistical report of the 2007 population and housing census results. In: Addis Ababa.

CSA [Ethiopia] (2016) Key findings of the 2015/2016 (2008 E.C) agricultural sample surveys.

CSA [Ethiopia] & ORC Macro (2001) *E*thiopia Demographic and Health Survey 2000. Addis Ababa, Ethiopia and Calverton, Maryland, USA: Central Statistical Authority and ORC Macro.

CSA [Ethiopia] & ORC Macro (2006) Ethiopia Demographic and Health Survey 2005. Addis Ababa, Ethiopia and Calverton, Maryland, USA: CSA and ORC.

CSA [Ethiopia] & ORC Macro (2011) Ethiopia Demographic and Health Survey 2011, Addis Ababa, Ethiopia and Calverton, Maryland, USA: Central Statistical Agency and ORC Macro.

CSA [Ethiopia] & ORC Macro (2014) Ethiopia Demographic and Health Survey 2014, Addis Ababa, Ethiopia and Calverton, Maryland, USA: Central Statistical Agency and ORC Macro.

CSA [Ethiopia] and ICF (2016) Ethiopia Demographic and Health Survey 2016: Key Indicators Report. Addis Ababa, Ethiopia, and Rockville, Maryland, USA. CSA and ICF.

Damsgaard C.T., Lauritzen L., Kjær T.M.R., Holm P.M.I., Fruekilde M.-B., Michaelsen K.F., et al. (2007) Fish Oil Supplementation Modulates Immune Function in Healthy Infants. *The Journal of Nutrition* **137,** 1031-1036.

Daniels M.C., Adair L.S., Popkin B.M. & Truong Y.K. (2009) Dietary diversity scores can be improved through the use of portion requirements: an analysis in young Filipino children. *European Journal of Clinical Nutrition* **63,** 199-208.

Darling N. & Steinberg L. (1993) Parenting style as context: An integrative model. *Psychological Bulletin* **113,** 487-496.

de Onis M., Blössner M. & Borghi E. (2012) Prevalence and trends of stunting among pre-school children, 1990–2020. *Public Health Nutrition* **15,** 142-148.

de Onis M., Blössner M., Borghi E., Frongillo E.A. & Morris R. (2004a) EStimates of global prevalence of childhood underweight in 1990 and 2015. *JAMA* **291,** 2600-2606.

de Onis M. & Branca F. (2016) Childhood stunting: a global perspective. *Maternal & Child Nutrition* **12,** 12-26.

de Onis M., Dewey K.G., Borghi E., Onyango A.W., Blössner M., Daelmans B., et al. (2013) The World Health Organization's global target for reducing childhood stunting by 2025: rationale and proposed actions. *Maternal & Child Nutrition* **9,** 6-26.

de Onis M., Garza C., Victora C.G., Onyango A.W., Frongillo E.A. & Martines J. (2004b) The WHO Multicentre Growth Reference Study: Planning, Study Design, and Methodology. *Food and Nutrition Bulletin* **25,** S15-S26.

Dearden K.A., Hilton S., Bentley M.E., Caulfield L.E., Wilde C., Ha P.B., et al. (2009) Caregiver Verbal Encouragement Increases Food Acceptance among Vietnamese Toddlers. *The Journal of Nutrition* **139,** 1387-1392.

Delgado-Noguera M.F., Calvache J.A. & Bonfill Cosp X. (2010) Supplementation with long chain polyunsaturated fatty acids (LCPUFA) to breastfeeding mothers for improving child growth and development (Review). *Cochrane Database of Systematic Reviews* **Issue 12. Art. No. : CD007901. DOI: 10.1002/14651858.CD007901.pub2**.

Delgado-Noguera M.F., Calvache J.A., Bonfill Cosp X., Kotanidou E.P. & Galli-Tsinopoulou A. (2015) Supplementation with long chain polyunsaturated fatty acids (LCPUFA) to breastfeeding mothers for improving child growth and development. *The Cochrane database of systematic reviews* **7,** CD007901-CD007901.

Deribew A., Tessema G.A., Deribe K., Melaku Y.A., Lakew Y., Amare A.T., et al. (2016) Trends, causes, and risk factors of mortality among children under 5 in Ethiopia, 1990–2013: findings from the Global Burden of Disease Study 2013. *Population Health Metrics* **14,** 42.

Dettwyler K.A. (1989) Styles of Infant Feeding: Parental/Caretaker Control of Food Consumption in Young Children. *American Anthropologist* **91,** 696-703.

Dettwyler K.A. & Fishman C. (1992) Infant Feeding Practices and Growth. *Annual Review of Anthropology* **21**.

Development Initiatives (2017) Global Nutrition Report 2017: Nourishing the SDGs. Bristol, UK: Development Initiatives.

Dewey K.G. (2013) The Challenge of Meeting Nutrient Needs of Infants and Young Children during the Period of Complementary Feeding: An Evolutionary Perspective. *The Journal of Nutrition* **143,** 2050-2054.

Dewey K.G. & Adu-Afarwuah S. (2008) Systematic review of the efficacy and effectiveness of complementary feeding interventions in developing countries. *Matern Child Nutr* **4 Suppl 1,** 24-85.

Dewey K.G. & Begum K. (2011) Long-term consequences of stunting in early life. *Maternal & Child Nutrition* **7,** 5-18.

Dewey K.G. & Brown K.H. (2003) Update on technical issues concerning complementary feeding of young children in developing countries and implications for intervention programs. *Food Nutr Bull.* **24,** 5-28.

Dewey K.G. & Huffman S.L. (2009) Maternal, infant, and young child nutrition: combining efforts to maximize impacts on child growth and micronutrient status. *Food and Nutrition Bulletin* **30,** S187-S189.

Dewey K.G. & Mayers D.R. (2011) Early child growth: how do nutrition and infection interact? *Maternal & Child Nutrition* **7,** 129-142.

Dewey K.G., R. J. Cohen, M. Arimond & M. T. Ruel (2006) Developing and Validating Simple Indicators of Complementary Food Intake and Nutrient Density for Breastfed Children in Developing Countries. Washington, DC: the Food and Nutrition Technical Assistance (FANTA) Project, Academy for Educational Development (AED).

Dewey K.G. & Vitta B.S. (2013) Strategies for ensuring adequate nutrient intake for infants and young children during the period of complementary feeding. *Washington: Alive & Thrive* **7**.

DFSFQ (n.d) Lucille Food Intake Application ; Available at : http://www.foodscience.ugent.be/nutriFOODchem/foodintake ; last accessed, March 2017.

Diggle P.J., Heagerty P.J., Liang K.-Y. & Zeger S.L. (2004) Analysis of longitudinal data second Edition. *Oxford statistical science series* **1,** ALL-ALL.

Disha A., Rawat R., Subandoro A. & Menon P. (2012) Infant and Young Child Feeding (IYCF) practices in Ethiopia and Zambia and their association with child nutrition: analysis of Demographic and Health Survey data. *African Journal of Food, Agriculture, Nutrition and Development* **12,** 5895-5914.

Disha A.D., Tharaney M., Abebe Y., Alayon S. & Winnard K. (2015) Factors associated with infant and young child feeding practices in Amhara Region and nationally in Ethiopia: Analysis of the 2005 and 2011 Demographic and Health Surveys. Washington, DC: Alive & Thrive.

Draper E., Reynolds C.M., Canavan M., Mills K.H., Loscher C.E. & Roche H.M. (2011) Omega-3 fatty acids attenuate dendritic cell function via NF-κB independent of PPARγ. *The Journal of Nutritional Biochemistry* **22,** 784-790.

Dror D.K. & Allen L.H. (2011) The Importance of Milk and other Animal-Source Foods for Children in Low-Income Countries. *Food and Nutrition Bulletin* **32,** 227-243.

Egal S., Hounsa A., Gong Y.Y., Turner P.C., Wild C.P., Hall A.J., et al. (2005) Dietary exposure to aflatoxin from maize and groundnut in young children from Benin and Togo, West Africa. *International Journal of Food Microbiology* **104,** 215-224.

EHNRI (1998a) Food composition table for use in Ethiopia. Part III. 1995-1997. Ethiopia Health and Nutrition Research Institute /Food Agriculture Organization: Addis Ababa Ethiopia.

EHNRI (1998b) Food composition table for use in Ethiopia. Part IV. 1995–1997. Ethiopia Health and Nutrition Research Institute /Food Agriculture Organization: Addis Ababa Ethiopia.

Engle P. (1999) The role of caring practices and resources for care in child survival, growth, and development: South and Southeast Asia. *Asian Development Review* **17,** 132-167.

Engle P. & Ricciuti H. (1995) Psychosocial aspects of care and nutrition. *Food Nutr Bull* **16,** 77.

Engle P.L., Bentley M. & Pelto G. (2000) The role of care in nutrition programmes: current research and a research agenda. *ProcNutrSoc* **59**.

Engle P.L. & Lhotska L. (1999) The role of care in programmatic actions for nutrition: Designing programmes involving care. *Food and Nutrition Bulletin* **20,** 121-135.

Engle P.L., Menon P. & Haddad L. (1999) Care and nutrition: concepts and measurement. *World Development* **27,** 1309-1337.

Engle P.L. & Pelto G.H. (2011) Responsive Feeding: Implications for Policy and Program Implementation. *The Journal of Nutrition* **141,** 508-511.

Engle P.L. & Zeitlin M. (1996) Active feeding behavior compensates for low interest in food among young Nicaraguan children. *J Nutr.* **Jul;126,** 1808-1816.

ENI (MOH) (1980) *Ethiopian Traditional Recipes.* First Edition edn.ENI, Addis Ababa ,Ethiopia.

Eshel N., Daelmans B., Mello M.C.d. & Martines J. (2006) Responsive parenting: interventions and outcomes. *Bulletin of the World Health Organization* **84,** 991-998.

Faith M.S. & Kerns J. (2005) Infant and child feeding practices and childhood overweight: the role of restriction. *Maternal & Child Nutrition* **1,** 164-168.

Faith M.S., Scanlon K.S., Birch L.L., Francis L.A. & Sherry B. (2004) Parent-Child Feeding Strategies and Their Relationships to Child Eating and Weight Status[ast][ast]. *Obesity* **12,** 1711-1722.

FAO (2008) Ethiopia Nutrition Profile – Nutrition and Consumer Protection Division, Food and Agriculture Organization.

FAO (2010) Fats and fatty acids in human nutrition; Report of an expert consultation;10-14 November 2008 Geneva ; FAO food and nutrition paper 91 , Rome.

FAO (2016) Nutrition country profile available at http://www.fao.org/ag/agn/nutrition/eth_en.stm

FAO/WHO (1992) International Conference on Nutrition Goals. World Declaration and Plan of Action. FAO, Rome and WHO, Geneva.

FAO/WHO (2004) Vitamin and mineral requirements in human nutrition. Rome and Geneva: FAO and WHO;.

FAO/WHO (2008) Interim Summary of Conclusions and Dietary Recommendations on Total Fat & Fatty Acids From the Joint FAO/WHO Expert Consultation on Fats and Fatty Acids in Human Nutrition, WHO, Geneva.

http://www.fao.org/ag/agn/nutrition/docs/Fats%20and%20Fatty%20Acids%20Summary.pdf.

FAO/WHO/UNU (2004) Human energy requirements. Food and Nutrition Technical Report Series No 1. UNU/WHO/FAO: Rome.

FDRE-PCC (2008) Summary and statistical report of the 2007 population and housing census of Ethiopia. *Addis Ababa*.

FDRE (2013) National Nutrition Programme, June 2013-June 2015.

FDRE (July, 2017) National Nutrition Program, 2016-2020.

FDRE (May, 2016) Growth and Transformation Plan II (GTP II) (2015/16-2019/20) ; National Planning Commission, Addis Ababa, Ethiopia.

FDRE (MOH) [Ethiopia] (2010) Health Sector Development Programme IV 2010/11 – 2014/15.

Fenton J.I., Gurzell E.A., Davidson E.A. & Harris W.S. (2016) Red blood cell PUFAs reflect the phospholipid PUFA composition of major organs. *Prostaglandins, Leukotrienes and Essential Fatty Acids (PLEFA)***112,** 12-23.

Ferrucci L., Cherubini A., Bandinelli S., Bartali B., Corsi A., Lauretani F., et al. (2006) Relationship of Plasma Polyunsaturated Fatty Acids to Circulating Inflammatory Markers. *The Journal of Clinical Endocrinology & Metabolism***91,** 439-446.

Field C.J., Van Aerde J.E., Robinson L.E. & Clandinin M.T. (2008) Effect of providing a formula supplemented with long-chain polyunsaturated fatty acids on immunity in full-term neonates. *Br J Nutr***99**.

Fisher J.O. & Birch L.L. (1999a) Restricting Access to Foods and Children's Eating. *Appetite***32,** 405-419.

Fisher J.O. & Birch L.L. (1999b) Restricting access to palatable foods affects children's behavioral response, food selection, and intake. *The American Journal of Clinical Nutrition* **69,** 1264-1272.

Flax V.L., Mäkinen S., Ashorn U., Cheung Y.B., Maleta K., Ashorn P., et al. (2011) Responsive feeding and child interest in food vary when rural Malawian children are fed lipid-based nutrient supplements or local complementary food. *Maternal & Child Nutrition*, no-no.

Flax V.L., Mäkinen S., Ashorn U., Cheung Y.B., Maleta K., Ashorn P., et al. (2013) Responsive feeding and child interest in food vary when rural Malawian children are fed lipid-based nutrient supplements or local complementary food. *Maternal & Child Nutrition* **9,** 369-380.

Flock M.R., Harris W.S. & Kris-Etherton P.M. (2013) Long-chain omega-3 fatty acids: time to establish a dietary reference intake. *Nutrition Reviews* **71,** 692-707.

FMoH/UNICEF/EU (2016) Situation Analysis of the Nutrition Sector in Ethiopia: 2000-2015. Ethiopian Federal Ministry of Health, UNICEF and European Commission Delegation. Addis Ababa, Ethiopia

Food and Nutrition Board (Institute of Medicine) (2002) Dietary Fats: Total Fat and Fatty Acids. Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein, and Amino Acids. Washington, D.C.: National Academies Press. 422-541.

Fritsche K.L. (2015) The Science of Fatty Acids and Inflammation. *Advances in Nutrition* **6,** 293S-301S.

Galler J.R., Ramsey F.C., Harrison R.H., Brooks R. & Weiskopf-Bock S. (1998) Infant Feeding Practices in Barbados Predict Later Growth. *J Nutr* **128**.

Gebhardt S.E., Lemar L.E., Pehrsson P.R., Exler J., Haytowitz D.B., Showell B.A., et al. (2010) USDA national nutrient database for standard reference, release 23.

German J.B. (2011) Dietary lipids from an evolutionary perspective: sources, structures and functions. *Maternal & Child Nutrition* **7,** 2-16.

Gibson R.A., Muhlhausler B. & Makrides M. (2011) Conversion of linoleic acid and alpha-linolenic acid to long-chain polyunsaturated fatty acids (LCPUFAs), with a focus on pregnancy, lactation and the first 2 years of life. *Maternal & Child Nutrition* **7,** 17-26.

Gibson R.S. (1990) *Principles of Nutritional Assessment.* Oxford University Press, New York.

Gibson R.S., Abebe Y., Hambidge K.M., Arbide I., Teshome A. & Stoecker B.J. (2009) Inadequate feeding practices and impaired growth among children from subsistence farming households in Sidama, Southern Ethiopia. *Maternal & Child Nutrition* **5,** 260-275.

Gibson R.S. & Ferguson E.L. (2008) An Interactive 24-Hour Recall for Assessing the Adequacy of Iron and Zinc Intakes in Developing Countries. International Life Sciences Institute, Washington, D.C.

Gibson R.S., Ferguson E.L. & Lehrfeld J. (1998) Complementary foods for infant feeding in developing countries: their nutrient adequacy and improvement. *European Journal of Clinical Nutrition* **52,** 764-770.

Girma K. (2016) Minerals and trace elements in the soil-plant-animal continuum in Ethiopia: a review. *African Journal of Food, Agriculture, Nutrition and Development* **16,** 11219-11235.

Girma W. & Timotiows G. (2002) *Determinants of Nutritional Status of Women and Children in Ethiopia: further analyis of Ethiopia Demographic and Health Survey 2000.* ORC Macro, Calverton, Maryland, USA.

Gittelsohn J., Shankar A.V., West Jr K.P., Faruque F., Gnywali T. & Pradhan E.K. (1998) Child feeding and care behaviors are associated with xerophthalmia in rural Nepalese households. *Social Science & Medicine* **47,** 477-486.

Glewwe P. & Miguel E.A. (2007) Chapter 56 The Impact of Child Health and Nutrition on Education in Less Developed Countries. In Handbook of Development Economics (Vol. 4, pp. 3561-3606). (Handbook of Development Economics; Vol. 4).

Godswill N.-N., Martin B.J., Kingsley T.-M., Albert D.-M.J., Thierry K.S., Sastile M.N., et al. (2016) Effects of Dietary Fatty Acids on Human Health: Focus on Palm oil from Elaeis guineensis Jacq. and Useful Recommendations. *Food and Public Health* **6,** 75-85.

Golden M.H. (2009) Proposed recommended nutrient densities for moderately malnourished children. *Food Nutr Bull* **30,** S267-342.

Gordon J.I., Dewey K.G., Mills D.A. & Medzhitov R.M. (2012) The Human Gut Microbiota and Undernutrition. *Science Translational Medicine* **4,** 137ps112-137ps112.

Goto R., Mascie-Taylor C. & Lunn P.G. (2009) Impact of intestinal permeability, inflammation status and parasitic infections on infant growth faltering in rural Bangladesh. *British Journal of Nutrition* **101,** 1509-1516.

Gottrand F. (2008) Long-chain polyunsaturated fatty acids influence the immune system of infants. *The Journal of Nutrition***138,** 1807S-1812S.

Grantham-McGregor S., Cheung Y.B., Cueto S., Glewwe P., Richter L., Strupp B., et al. (2007) Developmental potential in the first 5 years for children in developing countries. *Lancet***369,** 60-70.

Gurzell E.A., Wiesinger J.A., Morkam C., Hemmrich S., Harris W.S. & Fenton J.I. (2014) Is the omega-3 index a valid marker of intestinal membrane phospholipid EPA+DHA content? *Prostaglandins, Leukotrienes and Essential Fatty Acids (PLEFA)***91,** 87-96.

Ha P.B., Bentley M.E., Pachón H., Sripaipan T., Caulfield L.E., Marsh D.R., et al. (2002) Caregiver styles of feeding and child acceptance of food in rural Viet Nam. *Food and Nutrition Bulletin***23,** 92-98.

Haile D., Belachew T., Berhanu G., Setegn T. & Biadgilign S. (2014) Stability of infant and child feeding index over time and its association with nutritional status of HIV exposed infants in Sidama Zone, Southern Ethiopia: A longitudinal study. *Early Human Development***90,** 815-820.

Hansen A.E., Wiese H.F., Boelsche A.N., Haggard M.E., Adam D.J.D. & Davis H. (1963) ROLE OF LINOLEIC ACID IN INFANT NUTRITION. *Clinical and Chemical Study of 428 Infants Fed on Milk Mixtures Varying in Kind and Amount of Fat***31,** 171-192.

Harbron J. & Booley S. (2013) Responsive feeding: establishing healthy eating behaviour early on in life. *South African Journal of Clinical Nutrition***26,** S141-S149.

Harris W.S. (2007) The omega-6/omega-3 ratio and cardiovascular disease risk: Uses and abuses. *Current Cardiovascular Risk Reports***1,** 39-45.

Harris W.S., Mozaffarian D., Lefevre M., Toner C.D., Colombo J., Cunnane S.C., et al. (2009) Towards establishing dietary reference intakes for eicosapentaenoic and docosahexaenoic acids. *The Journal of Nutrition***139,** 804S-819S.

Hatløy A., Hallund J., Diarra M.M. & Oshaug A. (2000) Food variety, socioeconomic status and nutritional status in urban and rural areas in Koutiala (Mali). *Public Health Nutrition***3,** 57-65.

Hatloy A., Torheim L.E. & Oshaug A. (1998) Food variety–a good indicator of nutritional adequacy of the diet? A case study from an urban area in Mali, West Africa. *Eur J Clin Nutr***52**.

Headey D. (2014) An analysis of trends and determinants of child undernutrition in Ethiopia, 2000–2011. *International Food Policy Research Institute (IFPRI)*.

Heird W.C. (1997) Statistically significant versus biologically significant effects of long-chain polyunsaturated fatty acids on growth. *Developing Brain and Behaviour: the Role of Lipids in Infant Formula.(Dobbing, J., ed.). Academic Press, New York, NY***,** 169-205.

Hirvonen K., Taffesse A.S. & Worku Hassen I. (2015) Seasonality and household diets in Ethiopia. *Public Health Nutrition***FirstView,** 1-8.

Hoddinott J., Alderman H., Behrman J.R., Haddad L. & Horton S. (2013a) The economic rationale for investing in stunting reduction. *Maternal & Child Nutrition***9,** 69-82.

Hoddinott J., Behrman J.R., Maluccio J.A., Melgar P., Quisumbing A.R., Ramirez-Zea M., et al. (2013b) Adult consequences of growth failure in early childhood. *The American Journal of Clinical Nutrition*.

Hoddinott J., Headey D. & Dereje M. (2015) Cows, Missing Milk Markets, and Nutrition in Rural Ethiopia. *The Journal of Development Studies***51,** 958-975.

Hoerr S., Hughes S., Fisher J., Nicklas T., Liu Y. & Shewchuk R. (2009) Associations among parental feeding styles and children's food intake in families with limited incomes. *Int J BehavNutrPhys Act***6**.

Holman R.T., Johnson S.B. & Hatch T.F. (1982) A case of human linolenic acid deficiency involving neurological abnormalities. *The American Journal of Clinical Nutrition***35,** 617-623.

Hop L.T., Gross R., Giay T., Sastroamidjojo S., Schultink W. & Lang N.T. (2000) Premature complementary feeding is associated with poorer growth of vietnamese children. *J Nutr***130,** 2683-2690.

Horie T., Nakamaru M. & Masubuchi Y. (1998) Docosahexaenoic acid exhibits a potent protection of small intestine from methotrexate-induced damage in mice. *Life Sciences***62,** 1333-1338.

Hossain M.I., Nahar B., Hamadani J.D., Ahmed T., Roy A.K. & Brown K.H. (2010) Intestinal mucosal permeability of severely underweight and non-malnourished Bangladeshi children, and effects of nutritional rehabilitation. *Journal of Pediatric Gastroenterology and Nutrition***51,** 638-644.

Hotz C. & Brown K. (2004) International Zinc Nutrition Consultative Group (IZiNCG) technical document: Assessment of the risk of zinc deficiency in populations and options for its control. *Food Nutr Bull***25,** 94 - 203.

Hsieh J.-C., Liu L., Zeilani M., Ickes S., Trehan I., Maleta K., et al. (2015) High-Oleic Ready-to-Use Therapeutic Food Maintains Docosahexaenoic Acid Status in Severe Malnutrition. *Journal of Pediatric Gastroenterology and Nutrition* **61,** 138-143.

Huffman S.L., Harika R.K., Eilander A. & Osendarp S.J.M. (2011) Essential fats: how do they affect growth and development of infants and young children in developing countries? A literature review. *Matern Child Nutr* **7 Suppl 3,** 44-65.

Hughes S.O., Power T.G., Orlet Fisher J., Mueller S. & Nicklas T.A. (2005) Revisiting a neglected construct: parenting styles in a child-feeding context. *Appetite* **44,** 83-92.

Humphrey J.H. (2009) Child undernutrition, tropical enteropathy, toilets, and handwashing. *The Lancet* **374,** 1032-1035.

Hundera F. (1998) Variations of morpho-agronomic characters and grain chemical composition of released varieties of tef (Eragrostis tef (Zucc.) Trotter)[Ethiopia]. *Journal of Genetics & Breeding (Italy)*.

Hupkens C.L.H., Knibbe R.A., van Otterloo A.H. & Drop M.J. (1998) Class differences in the food rules mothers impose on their children: a cross-national study. *Social Science & Medicine* **47,** 1331-1339.

Hurley K.M. & Black M.M. (2011) Introduction to a Supplement on Responsive Feeding: Promoting Healthy Growth and Development for Infants and Toddlers. *The Journal of Nutrition* **141,** 489.

Hurley K.M., Cross M.B. & Hughes S.O. (2011) A Systematic Review of Responsive Feeding and Child Obesity in High-Income Countries. *The Journal of Nutrition* **141,** 495-501.

Huxley R.R., Shiell A.W. & Law C.M. (2000) The role of size at birth and postnatal catch-up growth in determining systolic blood pressure: a systematic review of the literature. *Journal of Hypertension* **18,** 815-831.

Iannotti L.L., Dulience S.J.L., Green J., Joseph S., François J., Anténor M.-L., et al. (2014) Linear growth increased in young children in an urban slum of Haiti: a randomized controlled trial of a lipid-based nutrient supplement. *The American Journal of Clinical Nutrition* **99,** 198-208.

Ichihara K.i., Waku K., Yamaguchi C., Saito K., Shibahara A., Miyatani S., et al. (2002) A convenient method for determination of the C20–22 PUFA composition of glycerolipids in blood and breast milk. *Lipids* **37,** 523-526.

IFPRI (2005) An assessment of the causes of malnutrition in Ethiopia. A Contribution to the Formulation of a National Nutrition Strategy for Ethiopia. Edited by T. Benson, International Food Policy Research Institute. Washington D.C. Available at: http://ebrary.ifpri.org/cdm/ref/collection/p15738coll2/id/58081 ; accessed in Nov 2016.

IFPRI/WHH/Concern Worldwide/UN (2016) Global Hunger Index: Getting to zero hunger Washington, DC/Dublin/Bonn

IHSR (Jimma University) (2011) Brief Introduction about Gilgel Gibe Field Research Center ; available at http://www.indepth-network.org/Profiles/Gilgel%20HDSS.pdf.

Imdad A., Yakoob M.Y. & Bhutta Z.A. (2011) Impact of maternal education about complementary feeding and provision of complementary foods on child growth in developing countries. *BMC Public Health* **11 Suppl 3,** S25.

Imhoff-Kunsch B., Stein A.D., Martorell R., Parra-Cabrera S., Romieu I. & Ramakrishnan U. (2011) Prenatal docosahexaenoic acid supplementation and infant morbidity: randomized controlled trial. *Pediatrics* **128,** e505-e512.

Innis S.M. (1991) Essential fatty acids in growth and development. *Progress in lipid research* **30,** 39-103.

Innis S.M. (2007) Human milk: maternal dietary lipids and infant development. *Proceedings of the Nutrition Society* **66,** 397-404.

Innis S.M. (2009) Omega-3 Fatty Acids and Neural Development to 2 Years of Age: Do We Know Enough for Dietary Recommendations? *Journal of Pediatric Gastroenterology and Nutrition* **48,** S16-S24.

Innis S.M. (2011) Metabolic programming of long-term outcomes due to fatty acid nutrition in early life. *Maternal & Child Nutrition* **7,** 112-123.

Innis S.M., Novak E.M. & Keller B.O. (2013) Long chain omega-3 fatty acids: Micronutrients in disguise. *Prostaglandins, Leukotrienes and Essential Fatty Acids (PLEFA)* **88,** 91-95.

Institute of Health Science Research (Jimma University) (October, 2011) Population and Household Heads' Demographics , Policy Brief Number 1, Jimma , Ethiopia. .

Institute of Medicine (1997) *Dietary reference intakes for calcium, phosphorus, magnesium, vitamin D, and fluoride. National Academy Press, Washington, DC.*

Institute of Medicine (2001) *Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc. National Academy Press, Washington, DC.*

Irwin L.G., Siddiqi A. & Hertzman C. (2007) Early child development: A powerful equalizer. *Final report to the WHO Commission on social determinants of health, Geneva*.

Jansen E., Daniels L.A. & Nicholson J.M. (2012) The dynamics of parenting and early feeding – constructs and controversies: a viewpoint. *Early Child Development and Care* **182,** 967-981.

Joan J. & Mesfin B.H. (2008) *Report on review of Incorporation of Essential Nutrition Actions into Public Health Programs in Ethiopia; Food and Nutrition Technical Assistance (FANTA) Project, Academy for Educational Development (AED).* Addis Ababa, Ethiopia.

Johnson S.L. & Birch L.L. (1994) Parents' and Children's Adiposity and Eating Style. *Pediatrics* **94,** 653-661.

Jones A.D., Ickes S.B., Smith L.E., Mbuya M.N., Chasekwa B., Heidkamp R.A., et al. (2014) World Health Organization infant and young child feeding indicators and their associations with child anthropometry: a synthesis of recent findings. *Maternal & Child Nutrition* **10,** 1-17.

Jones G., Steketee R.W., Black R.E., Bhutta Z.A. & Morris S.S. (2003) How many child deaths can we prevent this year? *The Lancet* **362,** 65-71.

Jones K.D., Ali R., Khasira M.A., Odera D., West A.L., Koster G., et al. (2015) Ready-to-use therapeutic food with elevated n-3 polyunsaturated fatty acid content, with or without fish oil, to treat severe acute malnutrition: a randomized controlled trial. *BMC Medicine* **13,** 1-14.

Kant A.K. (2004) Dietary patterns and health outcomes. *Journal of the American Dietetic Association* **104,** 615-635.

Katona P. & Katona-Apte J. (2008) The Interaction between Nutrition and Infection. *Clinical Infectious Diseases* **46,** 1582-1588.

Kau A.L., Ahern P.P., Griffin N.W., Goodman A.L. & Gordon J.I. (2011) Human nutrition, the gut microbiome and the immune system. *Nature* **474,** 327-336.

Kennedy G.L., Pedro M.R., Seghieri C., Nantel G. & Brouwer I. (2007) Dietary diversity score is a useful indicator of micronutrient intake in non-breast-feeding Filipino children. *Journal of Nutrition* **137,** 472-477.

Keusch G.T., Rosenberg I.H., Denno D.M., Duggan C., Guerrant R.L., Lavery J.V., et al. (2013) Implications of Acquired Environmental Enteric Dysfunction for Growth and Stunting in Infants and Children Living in Low- and Middle-Income Countries. *Food and Nutrition Bulletin* **34,** 357-364.

Khlangwiset P., Shephard G.S. & Wu F. (2011) Aflatoxins and growth impairment: A review. *Critical Reviews in Toxicology* **41,** 740-755.

Koletzko B., Lattka E., Zeilinger S., Illig T. & Steer C. (2011) Genetic variants of the fatty acid desaturase gene cluster predict amounts of red blood cell docosahexaenoic and other polyunsaturated fatty acids in pregnant women: findings from the Avon Longitudinal Study of Parents and Children. *The American Journal of Clinical Nutrition* **93,** 211-219.

Koletzko B., Lien E., Agostoni C., Böhles H., Campoy C., Cetin I., et al. (2008) The roles of long-chain polyunsaturated fatty acids in pregnancy, lactation and infancy: review of current knowledge and consensus recommendations. In: Journal of Perinatal Medicine.

Kong W., Yen J.-H., Vassiliou E., Adhikary S., Toscano M.G. & Ganea D. (2010) Docosahexaenoic acid prevents dendritic cell maturation and in vitro and in vivo expression of the IL-12 cytokine family. *Lipids in Health and Disease* **9,** 12.

Korpe P.S. & Petri W.A. (2012) Environmental Enteropathy: Critical implications of a poorly understood condition. *Trends in Molecular Medicine* **18,** 328-336.

Kosek M., Haque R., Lima A., Babji S., Shrestha S., Qureshi S., et al. (2013) Fecal markers of intestinal inflammation and permeability associated with the subsequent acquisition of linear growth deficits in infants. *Am J Trop Med Hyg* **88**.

Kotloff K.L., Nataro J.P., Blackwelder W.C., Nasrin D., Farag T.H., Panchalingam S., et al. (2013) Burden and aetiology of diarrhoeal disease in infants and young children in developing countries (the Global Enteric Multicenter Study, GEMS): a prospective, case-control study. *The Lancet* **382,** 209-222.

Kourlaba G. & Panagiotakos D.B. (2009) Dietary quality indices and human health: A review. *Maturitas* **62,** 1-8.

Lango Allen H., Estrada K., Lettre G., Berndt S.I., Weedon M.N., Rivadeneira F., et al. (2010) Hundreds of variants clustered in genomic loci and biological pathways affect human height. *Nature* **467,** 832-838.

Lapillonne A. & Carlson S.E. (2001) Polyunsaturated fatty acids and infant growth. *Lipids* **36,** 901-911.

Lapillonne A., Clarke S.D. & Heird W.C. (2003) Plausible mechanisms for effects of long-chain polyunsaturated fatty acids on growth. *The Journal of Pediatrics* **143,** 9-16.

Lapillonne A., Clarke S.D. & Heird W.C. (2004) Polyunsaturated fatty acids and gene expression. *Current Opinion in Clinical Nutrition & Metabolic Care* **7,** 151-156.

Lapillonne A., Pastor N., Zhuang W. & Scalabrin D.M. (2014) Infants fed formula with added long chain polyunsaturated fatty acids have reduced incidence of respiratory illnesses and diarrhea during the first year of life. *BMC Pediatrics* **14,** 1-8.

Lauritzen L. & Carlson S.E. (2011) Maternal fatty acid status during pregnancy and lactation and relation to newborn and infant status. *Maternal & Child Nutrition* **7,** 41-58.

Lauritzen L., Fewtrell M. & Agostoni C. (2015) Dietary arachidonic acid in perinatal nutrition: a commentary. *Pediatr Res* **77,** 263-269.

Lauritzen L., Hansen H.S., Jørgensen M.H. & Michaelsen K.F. (2001) The essentiality of long chain n-3 fatty acids in relation to development and function of the brain and retina. *Progress in lipid research* **40,** 1-94.

Lauritzen L., Hoppe C., Straarup E.M. & Michaelsen K.F. (2005a) Maternal Fish Oil Supplementation in Lactation and Growth during the First 2.5 Years of Life. *Pediatr Res* **58,** 235-242.

Lauritzen L., Jorgensen M.H., Olsen S.r.F., Straarup E.M. & Michaelsen K.F. (2005b) Maternal fish oil supplementation in lactation: effect on developmental outcome in breast-fed infants. *Reprod. Nutr. Dev.* **45,** 535-547.

Lawn J.E., Cousens S., Zupan J. & Team L.N.S.S. (2005) 4 million neonatal deaths: when? Where? Why? *The Lancet* **365,** 891-900.

Leroy J., Ruel M., Habicht J.-P. & Frongillo E. (2014) Linear growth deficit continues to accumulate beyond the first 1000 days in low- and middle-income countries: global evidence from 51 national surveys. *J Nutr* **144,** 1460 - 1466.

Leroy J., Ruel M., Habicht J.-P. & Frongillo E. (2015) Using height-for-age differences (HAD) instead of height-for-age z-scores (HAZ) for the meaningful measurement of population-level catch-up in linear growth in children less than 5years of age. *BMC Pediatrics* **15,** 145.

Lin A., Arnold B.F., Afreen S., Goto R., Huda T.M.N., Haque R., et al. (2013) Household Environmental Conditions Are Associated with Enteropathy and Impaired Growth in Rural Bangladesh. *The American Journal of Tropical Medicine and Hygiene* **89,** 130-137.

Lindenmayer G.W., Stoltzfus R.J. & Prendergast A.J. (2014) Interactions between Zinc Deficiency and Environmental Enteropathy in Developing Countries. *Advances in Nutrition: An International Review Journal* **5,** 1-6.

Liou Y.A., King D.J., Zibrik D. & Innis S.M. (2007) Decreasing Linoleic Acid with Constant α-Linolenic Acid in Dietary Fats Increases (n-3) Eicosapentaenoic Acid in Plasma Phospholipids in Healthy Men. *The Journal of Nutrition* **137,** 945-952.

Liu L., Johnson H.L., Cousens S., Perin J., Scott S., Lawn J.E., et al. (2012) Global, regional, and national causes of child mortality: an updated systematic analysis for 2010 with time trends since 2000. *Lancet* **379**.

López-Pedrosa J.M., Ramírez M., Torres M.I. & Gil A. (1999) Dietary Phospholipids Rich in Long-Chain Polyunsaturated Fatty Acids Improve the Repair of Small Intestine in Previously Malnourished Piglets. *The Journal of Nutrition* **129,** 1149-1155.

Lunn P.G. (2000) The impact of infection and nutrition on gut function and growth in childhood. *Proceedings of the Nutrition Society* **59,** 147-154.

Lunn P.G., Northrop-Clewes C.A. & Downes R.M. (1991) Intestinal permeability, mucosal injury, and growth faltering in Gambian infants. *The Lancet* **338,** 907-910.

Lutter C.K., Daelmans B.M.E.G., de Onis M., Kothari M.T., Ruel M.T., Arimond M., et al. (2011) Undernutrition, Poor Feeding Practices, and Low Coverage of Key Nutrition Interventions. *Pediatrics* **128**.

M'Kaibi F.K., Steyn N.P., Ochola S.A. & Du Plessis L. (2016) The relationship between agricultural biodiversity, dietary diversity, household food security, and stunting of children in rural Kenya. *Food Science & Nutrition*.

Ma J.-Q., Zhou L.-L., Hu Y.-Q., Liu J.-R., Liu S.-S., Zhang J., et al. (2012) A summary index of infant and child feeding practices is associated with child growth in urban Shanghai. *BMC Public Health* **12,** 568.

Maccoby E.E. (1992) The role of parents in the socialization of children: An historical overview. *Developmental Psychology* **28,** 1006-1017.

Magnus Domellöf, Bo Lönnerdal, Steven A Abrams & Olle Hernell (2002) Iron absorption in breast-fed infants: effects of age, iron status, iron supplements, and complementary foods. *The American Journal of Clinical Nutrition* **76,** 198-204.

Makrides M., Collins C.T. & Gibson R.A. (2011) Impact of fatty acid status on growth and neurobehavioural development in humans. *Maternal & Child Nutrition* **7,** 80-88.

Makrides M., Duley L. & Olsen S.F. (2006) Marine oil, and other prostaglandin precursor, supplementation for pregnancy uncomplicated by pre-eclampsia or intrauterine growth restriction. *Cochrane Database of Systematic Reviews*.

Makrides M., Gibson R.A., Udell T., Ried K. & Investigators t.I.L. (2005) Supplementation of infant formula with long-chain polyunsaturated fatty acids does not influence the growth of term infants. *The American Journal of Clinical Nutrition* **81,** 1094-1101.

Makrides M., Neumann M.A., Simmer K. & Gibson R.A. (1999) Dietary Long-Chain Polyunsaturated Fatty Acids Do Not Influence Growth of Term Infants: A Randomized Clinical Trial. *Pediatrics* **104,** 468-475.

Maluccio J. & Flores R. (2005) Impact evaluation of a conditional cash transfer program: the Nicaraguan Red de Protección Social. International Food Policy Research Institute (IFPRI).

Mamidi R.S., Shidhaye P., Radhakrishna K.V., Babu J.J. & Sudhershan Reddy P. (2011) Pattern of growth faltering and recovery in under-5 children in India using WHO growth standards — A study on first and third national family health survey. *INDIAN PEDIATRICS* **48,** 855-860.

Mangani C., Cheung Y.B., Maleta K., Phuka J., Thakwalakwa C., Dewey K., et al. (2014) Providing lipid-based nutrient supplements does not affect developmental milestones among Malawian children. *Acta Paediatrica* **103,** e17-e26.

Marquis G.S., Habicht J.P., Lanata C.F., Black R.E. & Rasmussen K.M. (1997a) Association of breastfeeding and stunting in Peruvian toddlers: an example of reverse causality. *Int J Epidemiol* **26,** 349-356.

Marquis G.S., Habicht J.P., Lanata C.F., Black R.E. & Rasmussen K.M. (1997b) Breast milk or animal-product foods improve linear growth of Peruvian toddlers consuming marginal diets. *The American Journal of Clinical Nutrition* **66,** 1102-1109.

Marriott B.P., White A., Hadden L., Davies J.C. & Wallingford J.C. (2012) World Health Organization (WHO) infant and young child feeding indicators: associations with growth measures in 14 low-income countries. *Maternal & Child Nutrition* **8,** 354-370.

Martin L., Hossain S., Casanovas C. & Guyon A. (2008) Learning from large-scale community-based programmes to improve breastfeeding practices.

Martorell R. & Zongrone A. (2012) Intergenerational Influences on Child Growth and Undernutrition. *Paediatric and Perinatal Epidemiology* **26,** 302-314.

Maxwell D.G. (1996) Measuring food insecurity: the frequency and severity of “coping strategies”. *Food policy* **21,** 291-303.

May A.L., Donohue M., Scanlon K.S., Sherry B., Dalenius K., Faulkner P., et al. (2007 Jul;) Child-feeding strategies are associated with maternal concern about children becoming overweight, but not children's weight status. *J Am Diet Assoc.* **107,** 1167-1175.

McKay S., Gaudier E., Campbell D.I., Prentice A.M. & Albers R. (2010) Environmental enteropathy: new targets for nutritional interventions. *International Health* **2,** 172-180.

Mekonen S., Lachat C., Ambelu A., Steurbaut W., Kolsteren P., Jacxsens L., et al. (2015) Risk of DDT residue in maize consumed by infants as complementary diet in southwest Ethiopia. *Science of the Total Environment* **511,** 454-460.

Michaelsen K.F., Dewey K.G., Perez-Exposito A.B., Nurhasan M., Lauritzen L. & Roos N. (2011) Food sources and intake of n-6 and n-3 fatty acids in low-income countries with emphasis on infants, young children (6–24 months), and pregnant and lactating women. *Matern Child Nutr* **7**.

Minns L.M., Kerling E.H., Neely M.R., Sullivan D.K., Wampler J.L., Harris C.L., et al. (2010) Toddler formula supplemented with docosahexaenoic acid (DHA) improves DHA status and respiratory health in a randomized, double-blind, controlled trial of US children less than 3 years of age. *Prostaglandins Leukot Essent Fatty Acids* **82**.

Moore A.C., Akhter S. & Aboud F.E. (2006) Responsive complementary feeding in rural Bangladesh. *Soc Sci Med* **62,** 1917-1930.

Morris S.S., Cousens S.N., Kirkwood B.R., Arthur P. & Ross D.A. (1996) Is prevalence of diarrhea a better predictor of subsequent mortality and weight gain than diarrhea incidence? *American Journal of Epidemiology* **144,** 582-588.

Moursi M., Arimond M., Dewey K., Treche S., Ruel M. & Delpeuch F. (2008a) Dietary diversity is a good predictor of the micronutrient density of the diet of 6- to 23-month-old children in Madagascar. *J Nutr* **138,** 2448 - 2453.

Moursi M.M., Martin-Prevel Y., Eymard-Duvernay S., Capon G., Treche S., Maire B., et al. (2008b) Assessment of child feeding practices using a summary index: stability over time and association with child growth in urban Madagascar. *American Journal of Clinical Nutrition* **87,** 1472-1479.

Moursi M.M., Treche S., Martin-Prevel Y., Maire B. & Delpeuch F. (2009) Association of a summary index of child feeding with diet quality and growth of 6-23 months children in urban Madagascar. *Eur J Clin Nutr* **63,** 718-724.

Muhlhausler B.S., Gibson R.A. & Makrides M. (2010) Effect of long-chain polyunsaturated fatty acid supplementation during pregnancy or lactation on infant and child body composition: a systematic review. *The American Journal of Clinical Nutrition* **92,** 857-863.

Muhlhausler B.S., Yelland L.N., McDermott R., Tapsell L., McPhee A., Gibson R.A., et al. (2016) DHA supplementation during pregnancy does not reduce BMI or body fat mass in children: follow-up of the DHA to Optimize Mother Infant Outcome randomized controlled trial. *The American Journal of Clinical Nutrition* **103,** 1489-1496.

Mukuria A.G., Monica T. Kothari & Noureddine Abderrahim (2006) Infant and Young Child Feeding Updates. Calverton, Maryland, USA: ORC Macro.

Muller O. & Krawinkel M. (2005) Malnutrition and health in developing countries. *Canadian Medical Association Journal* **173,** 279-286.

Neufeld L.M., Haas J.D., Grajéda R. & Martorell R. (2004) Changes in maternal weight from the first to second trimester of pregnancy are associated with fetal growth and infant length at birth. *The American Journal of Clinical Nutrition* **79,** 646-652.

Nishimura R.Y., Castro G.S.F.d., Junior A.A.J. & Sartorelli D.S. (2013) Breast milk fatty acid composition of women living far from the coastal area in Brazil. *Jornal de Pediatria (Versão em Português)* **89,** 263-268.

Ntab B., Simondon K., Milet J., Cisse B., Sokhna C., Boulanger D., et al. (2005) A young child feeding index is not associated with either height-for-age or height velocity in rural Senegalese children. *J Nutr* **135,** 457 - 464.

Nti C.A. & Lartey A. (2007) Effect of caregiver feeding behaviours on child nutritional status in rural Ghana. *International Journal of Consumer Studies* **31,** 303-309.

Olofin I., McDonald C.M., Ezzati M., Flaxman S., Black R.E., Fawzi W.W., et al. (2013) Associations of Suboptimal Growth with All-Cause and Cause-Specific Mortality in Children under Five Years: A Pooled Analysis of Ten Prospective Studies. *PLoS One* **8,** e64636.

Onis M.d., Onyango A.W., Borghi E., Siyam A., Nishida C. & Siekmann J. (2007) Development of a WHO growth reference for school-aged children and adolescents. *Bulletin of the World Health Organization* **85,** 660-667.

Onyango A., Koski K.G. & Tucker K.L. (1998) Food diversity versus breastfeeding choice in determining anthropometric status in rural Kenyan toddlers. *International Journal of Epidemiology* **27,** 484-489.

Onyango A.W. (2003) Dietary diversity, child nutrition and health in contemporary African communities. *Comp Biochem Physiol A Mol Integr Physiol* **136,** 61-69.

Onyango A.W., Borghi E., de Onis M., del Carmen Casanovas M. & Garza C. (2014) Complementary feeding and attained linear growth among 6–23-month-old children. *Public Health Nutrition* **17,** 1975-1983.

Onyango A.W., Receveur O. & Esrey S.A. (2002) The contribution of breast milk to toddler diets in western Kenya. *Bulletin of the World Health Organization* **80,** 292-299.

Owino V., Ahmed T., Freemark M., Kelly P., Loy A., Manary M., et al. (2016) Environmental Enteric Dysfunction and Growth Failure/Stunting in Global Child Health. *Pediatrics*.

Özaltin E., Hill K. & Subramanian S. (2010) Association of maternal stature with offspring mortality, underweight, and stunting in low-to middle-income countries. *JAMA* **303,** 1507-1516.

PAHO/WHO (2002) Guiding principles for breastfeeding and complementary feeding, Pan American Health Organization /World Health Organization.

PAHO/WHO (2003) Guiding Principles for Complementary Feeding of the Breastfed Child. Division of Health Promotion and Protection. Food and Nutrition Program: Washington, DC.

Panter-Brick C., Lunn P.G., Langford R.M., Maharjan M. & Manandhar D.S. (2008) Pathways leading to early growth faltering: an investigation into the importance of mucosal damage and immunostimulation in different socio-economic groups in Nepal. *British Journal of Nutrition* **101,** 558-567.

Pastor N., Soler B., Mitmesser S.H., Ferguson P. & Lifschitz C. (2006) Infants fed docosahexaenoic acid-and arachidonic acid-supplemented formula have decreased incidence of bronchiolitis/bronchitis the first year of life. *Clinical pediatrics* **45,** 850-855.

Patrick H., Nicklas T.A., Hughes S.O. & Morales M. (2005) The benefits of authoritative feeding style: caregiver feeding styles and children's food consumption patterns. *Appetite* **44,** 243-249.

Pelletier D.L. (1994) The relationship between child anthropometry and mortality in developing countries: implications for policy, programs and future research. *J Nutr Oct***124**.

Pelletier D.L., Frongillo E.A., Schroeder D.G. & Habicht J.P. (1995) The effects of malnutrition on child mortality in developing countries. *Bull World Health Organ***73.**

Pelto G.H. (2000) Improving complementary feeding practices and responsive parenting as a primary component of interventions to prevent malnutrition in infancy and early childhood. *Pediatrics***106,** 1300.

Petrou S. & Kupek E. (2010) Poverty and childhood undernutrition in developing countries: A multi-national cohort study. *Social Science & Medicine***71,** 1366-1373.

Pezzoli L., Pineda S., Halkyer P., Crespo G., Andrews N. & Ronveaux O. (2009) Cluster-sample surveys and lot quality assurance sampling to evaluate yellow fever immunisation coverage following a national campaign, Bolivia, 2007

Surveillance d'échantillons en grappe et échantillonnage par assurance qualité de lot pour évaluer la couverture de la vaccination contre la fièvre jaune suite à une campagne nationale en Bolivie en 2007

Estudios con muestreo por racimos (clusters) y con muestreo por lotes para garantizar la calidad para evaluar la cobertura de la inmunización contra la Fiebre Amarilla tras una campaña nacional en Bolivia en el 2007. *Tropical Medicine & International Health***14,** 355-361.

Phuka J.C., Maleta K., Thakwalakwa C., Cheung Y.B., Briend A., Manary M.J., et al. (2008) Complementary feeding with fortified spread and incidence of severe stunting in 6- to 18-month-old rural Malawians. *Arch Pediatr Adolesc Med***162,** 619-626.

Phuka J.C., Maleta K., Thakwalakwa C., Cheung Y.B., Briend A., Manary M.J., et al. (2009) Postintervention growth of Malawian children who received 12-mo dietary complementation with a lipid-based nutrient supplement or maize-soy flour. *The American Journal of Clinical Nutrition***89,** 382-390.

Poli A. & Visioli F. (2015) Recent evidence on omega 6 fatty acids and cardiovascular risk. *European Journal of Lipid Science and Technology***117,** 1847-1852.

Population Reference Bureau (2016) 2016 world population data sheet with a special focus on human needs and sustainable resources.

Prasad A.S. (2009) Zinc: role in immunity, oxidative stress and chronic inflammation. *Current Opinion in Clinical Nutrition & Metabolic Care***12,** 646-652.

Prendergast A. & Kelly P. (2012) Enteropathies in the developing world: neglected effects on global health. *Am J Trop Med Hyg***86**.

Prendergast A.J. & Humphrey J.H. (2014) The stunting syndrome in developing countries. *Paediatrics and International Child Health***34,** 250-265.

Prentice A.M. & Paul A.A. (2000) Fat and energy needs of children in developing countries. *The American Journal of Clinical Nutrition***72,** 1253s-1265s.

Prentice A.M. & van der Merwe L. (2011) Impact of fatty acid status on immune function of children in low-income countries. *Maternal & Child Nutrition***7,** 89-98.

Prentice A.M., Ward K.A., Goldberg G.R., Jarjou L.M., Moore S.E., Fulford A.J., et al. (2013) Critical windows for nutritional interventions against stunting. *The American Journal of Clinical Nutrition***97,** 911-918.

Prüss-Üstün A., Bos R., Gore F. & Bartram J. (2008) *Safer water, better health: costs, benefits and sustainability of interventions to protect and promote health.* World Health Organization.

Prüss-Üstün A. & Corvalán C. (2006) Preventing disease through healthy environments. *Geneva: World Health Organization*.

Racine R.A. & Deckelbaum R.J. (2007) Sources of the very-long-chain unsaturated omega-3 fatty acids: eicosapentaenoic acid and docosahexaenoic acid. *Current Opinion in Clinical Nutrition and Metabolic Care***10,** 123-128.

Ramakrishnan U., Martorell R., Schroeder D.G. & Flores R. (1999) Role of Intergenerational Effects on Linear Growth. *The Journal of Nutrition***129,** 544.

Regassa N. & Stoecker B.J. (2012) Household food insecurity and hunger among households in Sidama district, southern Ethiopia. *Public Health Nutrition***15,** 1276-1283.

Reinbott A., Kuchenbecker J., Herrmann J., Jordan I., Muehlhoff E., Kevanna O., et al. (2015) A child feeding index is superior to WHO IYCF indicators in explaining length-for-age Z-scores of young children in rural Cambodia. *Paediatrics and International Child Health***35,** 124-134.

Richard S.A., Black R.E., Gilman R.H., Guerrant R.L., Kang G., Lanata C.F., et al. (2013) Diarrhea in Early Childhood: Short-term Association With Weight and Long-term Association With Length. *American Journal of Epidemiology***178,** 1129-1138.

Rocquelin G., Tapsoba S., Kiffer J. & Eymard-Duvernay S. (2003) Human milk fatty acids and growth of infants in Brazzaville (The Congo) and Ouagadougou (Burkina Faso). *Public Health Nutrition* **6,** 241-247.

Rodríguez G., Iglesia I., Bel-Serrat S. & Moreno L.A. (2012) Effect of n-3 long chain polyunsaturated fatty acids during the perinatal period on later body composition. *British Journal of Nutrition* **107,** S117-S128.

Rollins N.C., Bhandari N., Hajeebhoy N., Horton S., Lutter C.K., Martines J.C., et al. (2016) Why invest, and what it will take to improve breastfeeding practices? *The Lancet* **387,** 491-504.

Ruel M. & Menon P. (2002) Child feeding practices are associated with child nutritional status in Latin America: innovative uses of the demographic and health surveys. *J Nutr* **132,** 1180 - 1187.

Ruel M.T. & Alderman H. (2013) Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *The Lancet* **382,** 536-551.

Ruel M.T., Brown K.H. & Caulfield L.E. (2003) Moving forward with complementary feeding: indicators and research priorities. International Food Policy Research Institute (IFPRI) discussion paper 146 (April 2003). *Food Nutr Bull* **24,** 289-290.

Ruel M.T., Levin C.E., Armar-Klemesu M., Maxwell D. & Morris S.S. (1999) Good Care Practices Can Mitigate the Negative Effects of Poverty and Low Maternal Schooling on Children's Nutritional Status: Evidence from Accra. *World Development* **27,** 1993-2009.

Ryan K.N., Stephenson K.B., Trehan I., Shulman R.J., Thakwalakwa C., Murray E., et al. (2014) Zinc or Albendazole Attenuates the Progression of Environmental Enteropathy: A Randomized Controlled Trial. *Clinical Gastroenterology and Hepatology* **12,** 1507-1513.e1501.

Sacco L.M., Bentley M.E., Carby-Shields K., Borja J.B. & Goldman B.D. (2007) Assessment of infant feeding styles among low-income African-American mothers: Comparing reported and observed behaviors. *Appetite* **49,** 131-140.

Saha K.K., Frongillo E.A., Alam D.S., Arifeen S.E., Persson L.Ã.k. & Rasmussen K.M. (2008) Appropriate infant feeding practices result in better growth of infants and young children in rural Bangladesh. *The American Journal of Clinical Nutrition* **87,** 1852-1859.

Savy M., Martin-Prével Y., Traissac P., Eymard-Duvernay S. & Delpeuch F. (2006) Dietary Diversity Scores and Nutritional Status of Women Change during the Seasonal Food Shortage in Rural Burkina Faso. *The Journal of Nutrition* **136,** 2625-2632.

Sawadogo P., Martin-Prevel Y., Savy M., Kameli Y., Traissac P., Traore A., et al. (2006) An infant and child feeding index is associated with the nutritional status of 6- to 23-month-old children in rural Burkina Faso. *J Nutr* **136,** 656 - 663.

Scrimshaw N. (1989) Energy cost of communicable diseases in infancy and childhood. *Activity, Energy Expenditure and Energy Requirements of Infants and Children*, 215-238.

Semba R.D., Trehan I., Li X., Moaddel R., Ordiz M.I., Maleta K.M., et al. (2017) Environmental enteric dysfunction is associated with carnitine deficiency and altered fatty acid oxidation. *EBioMedicine* **17,** 57-66.

Shloim N., Edelson L.R., Martin N. & Hetherington M.M. (2015) Parenting Styles, Feeding Styles, Feeding Practices, and Weight Status in 4–12 Year-Old Children: A Systematic Review of the Literature. *Frontiers in psychology* **6**.

Simmer K. (2001) Longchain polyunsaturated fatty acid supplementation in infants born at term. *Cochrane Database of Systematic Reviews*.

Simmer K., Patole S. & Rao S.C. (2008) Longchain polyunsaturated fatty acid supplementation in infants born at term. *Cochrane Database Syst Rev* **1**.

Simmer K., Patole S.K. & Rao S.C. (2011) Longchain polyunsaturated fatty acid supplementation in infants born at term. *Cochrane Database of Systematic Reviews*.

Simondon K.B., Costes R., Delaunay V., Diallo A. & Simondon F. (2001a) Children's height, health and appetite influence mothers' weaning decisions in rural Senegal. *International Journal of Epidemiology* **30,** 476-481.

Simondon K.B. & Simondon F. (1998) Mothers prolong breastfeeding of undernourished children in rural Senegal. *International Journal of Epidemiology* **27,** 490-494.

Simondon K.B., Simondon F., Costes R., Delaunay V. & Diallo A. (2001b) Breast-feeding is associated with improved growth in length, but not weight, in rural Senegalese toddlers. *The American Journal of Clinical Nutrition* **73,** 959-967.

Simopoulos A.P. (2002) The importance of the ratio of omega-6/omega-3 essential fatty acids. *Biomedicine & pharmacotherapy* **56,** 365-379.

Simopoulos A.P. (2016) An increase in the omega-6/omega-3 fatty acid ratio increases the risk for obesity. *Nutrients* **8,** 128.

Smit E.N., Muskiet F.A.J. & Boersma E.R. (2004) The possible role of essential fatty acids in the pathophysiology of malnutrition: a review. *Prostaglandins, leukotrienes and essential fatty acids* **71,** 241-250.

Smith H.E., Ryan K.N., Stephenson K.B., Westcott C., Thakwalakwa C., Maleta K., et al. (2014) Multiple Micronutrient Supplementation Transiently Ameliorates Environmental Enteropathy in Malawian Children Aged 12–35 Months in a Randomized Controlled Clinical Trial. *The Journal of Nutrition* **144,** 2059-2065.

Smith M.I., Yatsunenko T., Manary M.J., Trehan I., Mkakosya R., Cheng J., et al. (2013) Gut microbiomes of Malawian twin pairs discordant for kwashiorkor. *Science (New York, N.Y.)* **339,** 548-554.

Smuts C., Tichelaar H., Van Jaarsveld P., Badenhorst C., Kruger M., Laubscher R., et al. (1995) The effect of iron fortification on the fatty acid composition of plasma and erythrocyte membranes in primary school children with and without iron deficiency. *Prostaglandins, leukotrienes and essential fatty acids* **52,** 59-67.

Smuts C.M., Tichelaar H.Y., van Jaarsveld P.J., Badenhorst C.J., Kruger M., Laubscher R., et al. (1994) The effect of iron fortification on the fatty acid composition of plasma and erythrocyte membranes in primary school children with and without iron-deficiency. *Prostaglandins, leukotrienes and essential fatty acids* **51,** 277-285.

Stein A.D., Wang M., Martorell R., Norris S.A., Adair L.S., Bas I., et al. (2010) Growth patterns in early childhood and final attained stature: data from five birth cohorts from low- and middle-income countries. *Am J Hum Biol* **22,** 353-359.

Steyn N., Nel J., Nantel G., Kennedy G. & Labadarios D. (2006) Food variety and dietary diversity scores in children: are they good indicators of dietary adequacy? *Public Health Nutr* **9,** 644 - 650.

Szajewska H., Horvath A. & Koletzko B. (2006) Effect of n–3 long-chain polyunsaturated fatty acid supplementation of women with low-risk pregnancies on pregnancy outcomes and growth measures at birth: a meta-analysis of randomized controlled trials. *The American Journal of Clinical Nutrition* **83,** 1337-1344.

Tarini A., Bakari S. & Delisle H. (1999) [The overall nutritional quality of the diet is reflected in the growth of Nigerian children]. *Sante (Montrouge, France)* **9,** 23-31.

Teitelbaum J.E. & Allan Walker W. (2001) Review: the role of omega 3 fatty acids in intestinal inflammation. *The Journal of Nutritional Biochemistry* **12,** 21-32.

The World Bank (2006) Repositioning Nutrition as Central to Development: A Strategy for Large-Scale Action. The World Bank: Washington, DC.

The World Bank (2015) "World Bank Forecasts Global Poverty to Fall Below 10% for First Time; Major Hurdles Remain in Goal to End Poverty by 2030". http://www.worldbank.org/en/news/press-release/2015/10/04/world-bank-forecasts-global-poverty-to-fall-below-10-for-first-time-major-hurdles-remain-in-goal-to-end-poverty-by-2030 Retrieved 23 October 2017.

Thienprasert A., Samuhaseneetoo S., Popplestone K., West A.L., Miles E.A. & Calder P.C. (2009) Fish oil n-3 polyunsaturated fatty acids selectively affect plasma cytokines and decrease illness in Thai schoolchildren: a randomized, double-blind, placebo-controlled intervention trial. *J Pediatr***154**.

Tinoco S., Sichieri R., Setta C.L., Moura A.S. & Carmo M. (2009) n-3 polyunsaturated fatty acids in milk is associate to weight gain and growth in premature infants. *Lipids Health Dis***8,** 23.

Trumbo P., Schlicker S., Yates A.A. & Poos M. (2002) Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein and Amino Acids. *Journal of the American Dietetic Association***102,** 1621-1630.

Twisk J.W. (2003) *Applied longitudinal data analysis for epidemiology: a practical guide.* Cambridge University Press.

Uauy R. & Castillo C. (2003) Lipid Requirements of Infants: Implications for Nutrient Composition of Fortified Complementary Foods. *The Journal of Nutrition***133,** 2962S-2972S.

Uauy R. & Dangour A.D. (2009) Fat and fatty acid requirements and recommendations for infants of 0–2 years and children of 2–18 years. *Ann Nutr Metab***55**.

UNICEF (1990) Strategy for improved nutrition of children and women in developing countries , New York, UNICEF.

UNICEF (1997) The care initiative assessment, analysis, and action to improve care for nutrition. UNICEF Nutrition Section; New York

UNICEF (2012) Infant and young child feeding, programming guide ; Nutrition Section, Programmes, United Nations Children's Fund , New York.

UNICEF (2013) Improving child nutrition: the achievable imperative for global progress, New York: UNICEF.

UNICEF (October, 2016) From the first hour of life: Making the case for improved infant and young child feeding everywhere; United Nations Children's Fund ; New York ; USA.

UNICEF/WHO/The World Bank /UNDP (2015) Levels and Trends in Child Mortality Report 2015: Estimates Developed by the UN Inter-agency Group for Child Mortality Estimation. New York: UNICEF.

UNICEF/WHO/World Bank (2016) Levels and Trends in Child Malnutrition. UNICEF – WHO – World Bank Group joint child malnutrition estimates. Key findings of the 2016 edition

United Nations (2015a) *The millennium development goals report , New York.* United Nations Publications.

United Nations (2015b) Transforming our world: the 2030 agenda for sustainable development. New York (NY): (https://sustainabledevelopment.un.org/post2015/transformingourworld, accessed September 2017).

USDA (Foreign Agricultural Service) (2017) Ethiopia's Ag Imports Continue Growing; Global Agriculture Information Network , GAIN REPORT Number: ET1634 . Recovered on October 19, 2017 https://gain.fas.usda.gov/Recent%20GAIN%20Publications/Ethiopia%E2%80%99s%20Ag%20Imports%20Continue%20Growing_Addis%20Ababa_Ethiopia_2-7-2017.pdf.

Vaitla B., Devereux S. & Swan S.H. (2009) Seasonal Hunger: A Neglected Problem with Proven Solutions. *PLoS Medicine***6,** e1000101.

van der Merwe J., Kluyts M., Bowley N. & Marais D. (2007) Optimizing the introduction of complementary foods in the infant's diet: a unique challenge in developing countries. *Matern Child Nutr***3,** 259-270.

van der Merwe L.F., Moore S.E., Fulford A.J., Halliday K.E., Drammeh S., Young S., et al. (2013) Long-chain PUFA supplementation in rural African infants: a randomized controlled trial of effects on gut integrity, growth, and cognitive development. *The American Journal of Clinical Nutrition***97,** 45-57.

van Goudoever H., Guandalini S. & Kleinman R.E. (2011) *Early Nutrition: Impact on Short-and Long-Term Health.* Karger Medical and Scientific Publishers.

Vazir S., Engle P., Balakrishna N., Griffiths P.L., Johnson S.L., Creed-Kanashiro H., et al. (2013) Cluster-randomized trial on complementary and responsive feeding education to caregivers found improved dietary intake, growth and development among rural Indian toddlers. *Maternal & Child Nutrition* **9,** 99-117.

Ventura A.K. & Birch L.L. (2008) Does parenting affect children's eating and weight status? *International Journal of Behavioral Nutrition and Physical Activity* **5,** 15.

Victora C., Adair L., Fall C., Hallal P., Martorell R., Richter L., et al. (2008) Maternal and child undernutrition: consequences for adult health and human capital. *Lancet* **371**.

Victora C., de Onis M., Hallal P., Blossner M. & Shrimpton R. (2010) Worldwide timing of growth faltering: revisiting implications for interventions. *Pediatrics* **125,** e473 - 480.

Victora C.G., Bahl R., Barros A.J.D., França G.V.A., Horton S., Krasevec J., et al. (2016) Breastfeeding in the 21st century: epidemiology, mechanisms, and lifelong effect. *The Lancet* **387,** 475-490.

Vitta B. & Dewey K. (2012) Identifying Micronutrient Gaps in the Diets of Breastfed 6-11-Month-Old Infants in Bangladesh, Ethiopia and Viet Nam Using Linear Programming. *Washington, DC: Alive and Thrive*.

Vollmer R.L. & Mobley A.R. (2013) Parenting styles, feeding styles, and their influence on child obesogenic behaviors and body weight. A review. *Appetite* **71,** 232-241.

Vyas S. & Kumaranayake L. (2006) Constructing socio-economic status indices: how to use principal components analysis. *Health Policy and Planning* **21,** 459-468.

Wang A.Z., Shulman R.J., Crocker A.H., Thakwalakwa C., Maleta K.M., Devaraj S., et al. (2017) A Combined Intervention of Zinc, Multiple Micronutrients, and Albendazole Does Not Ameliorate Environmental Enteric Dysfunction or Stunting in Rural Malawian Children in a Double-Blind Randomized Controlled Trial. *The Journal of Nutrition* **147,** 97-103.

Webb A.L., Stein A.D., Ramakrishnan U., Hertzberg V.S., Urizar M. & Martorell R. (2006) A simple index to measure hygiene behaviours. *International Journal of Epidemiology* **35,** 1469-1477.

Weisz A.J., Manary M.J., Stephenson K., Agapova S., Manary F.G., Thakwalakwa C., et al. (2012) Abnormal Gut Integrity Is Associated With Reduced Linear Growth in Rural Malawian Children. *Journal of Pediatric Gastroenterology and Nutrition* **55,** 747-750.

Whincup P.H., Kaye S.J., Owen C.G., Huxley R., Cook D.G., Anazawa S., et al. (2008) Birth weight and risk of type 2 diabetes: a systematic review. *JAMA* **300,** 2886-2897.

WHO (1995) Physical Status: the Use and Interpretation of Anthropometry - Report of an Expert committee,Geneva.

WHO (1996) Trace elements in human nutrition and health. Geneva: WHO.

WHO (1998) Complementary feeding of young children in developing countries: a review of current scientific knowledge. WHO/NUT/98.1. Geneva.

WHO (1999) A critical link: Interventions for physical growth and psychological development: a review. Geneva: WHO Department of Child and Adolescent Health and Development.

WHO (2001) The optimal duration of exclusive breastfeeding. Report of an Expert Consultation. Geneva, World Health Organization

WHO (2005) Guiding principles for feeding non-breastfed children 6–24 months of age. Geneva.

WHO (2006) WHO Child Growth Standards: length/Height-for-age, weight-for-age, weight-for-length, weight-for-height and body mass index-for-age. Methods and development. In: WHO Child Growth Standards: Length/Height-for-age, weight-for-age, weight-for-length, weight-for-height and body mass index-for-age.

WHO (2007a) Report of a WHO Technical Consultation on Birth Spacing Geneva, Switzerland 13–15 , 2005.

WHO (2007b) WHO Anthro for personal computers, version 2, 2007: Software for assessing growth and development of the world's children. Geneva.

WHO (2008) *Indicators for assessing infant and young child feeding practices. Part 1: Definitions.* World Health Organization, Geneva.

WHO (2010a) Indicators for assessing infant and young child feeding practices, Part 3: country profiles. In: Indicators for assessing infant and young child feeding practices, Part 3: country profiles.

WHO (2010b) Indicators for assessing infant and young child feeding practices: Part 2: measurement.: Geneva WHO.

WHO (2010c) WHO Anthro for personal computers, version 3.2.2, 2011: Software and macros for assessing growth and development of the world's children. Geneva (http://www.who.int/childgrowth/software/en/).

WHO (2012) Resolution WHA65.6. Maternal, infant and young child nutrition. In: Sixty-fifth World Health Assembly, Geneva, 21–26 May. Resolutions and decisions, annexes. World Health Organization: Geneva. (WHA65/2012/REC/1).

WHO (2014) C-reactive protein concentrations as a marker of inflammation or infection for interpreting biomarkers of micronutrient status. Vitamin and Mineral Nutrition Information System. Geneva WHO.

WHO & UNICEF. (2003) *Global strategy for infant and young child feeding.* World Health Organization.

Wieland L.S. & Santesso N. (2016) A Summary of a Cochrane Review: Supplementation with long chain polyunsaturated fatty acids (LCPUFA) to breastfeeding mothers for improving child growth and development. *European Journal of Integrative Medicine* **8,** 113-114.

Willemsen L.E., Koetsier M.A., Balvers M., Beermann C., Stahl B. & van Tol E.A. (2008) Polyunsaturated fatty acids support epithelial barrier integrity and reduce IL-4 mediated permeability in vitro. *European journal of nutrition* **47,** 183-191.

Winichagoon P. (2008) Limitations and resolutions for dietary assessment of micronutrient intakes. *Asia Pac J Clin Nutr.* **17,** 296-298.

Wondafrash M., Amsalu T. & Woldie M. (2012) Feeding styles of caregivers of children 6-23 months of age in Derashe special district, Southern Ethiopia. *BMC Public Health* **12,** 1-8.

Wondafrash M., Huybregts L., Lachat C., Bouckaert K.P. & Kolsteren P. (2016) Dietary diversity predicts dietary quality regardless of season in 6–12-month-old infants in south-west Ethiopia. *Public Health Nutrition* **19,** 2485-2494.

Wondafrash M., Huybregts L., Lachat C., Bouckaert K.P. & Kolsteren P. (2017) Feeding practices and growth among young children during two seasons in rural Ethiopia. *BMC Nutrition* **3,** 39.

Wood K.E., Lau A., Mantzioris E., Gibson R.A., Ramsden C.E. & Muhlhausler B.S. (2014) A low omega-6 polyunsaturated fatty acid (n-6 PUFA) diet increases omega-3 (n-3) long chain PUFA status in plasma phospholipids in humans. *Prostaglandins, Leukotrienes and Essential Fatty Acids (PLEFA)* **90,** 133-138.

Wood K.E., Mantzioris E., Gibson R.A., Ramsden C.E. & Muhlhausler B.S. (2015) The effect of modifying dietary LA and ALA intakes on omega-3 long chain polyunsaturated fatty acid (n-3 LCPUFA) status in human adults: a systematic review and commentary. *Prostaglandins Leukot Essent Fatty Acids* **95**.

Working Group on IYCF Indicators (2006) Developing and Validating Simple Indicators of Dietary Quality and Energy Intake of Infants and Young Children in Developing Countries: Summary of findings from analysis of 10 data sets. Food and Nutrition Technical Assistance (FANTA) Project, Academy for Educational Development (AED), Washington, D.C., August 2006.

Wright K., Coverston C., Tiedeman M. & Abegglen J.A. (2006) Formula Supplemented with Docosahexaenoic Acid (DHA) and Arachidonic Acid (ARA): A Critical Review of the Research. *Journal for Specialists in Pediatric Nursing* **11,** 100-112.

Yuhas R., Pramuk K. & Lien E. (2006) Human milk fatty acid composition from nine countries varies most in DHA. *Lipids* **41,** 851-858.

Zhang J., Shi L., Wang J. & Wang Y. (2009) An infant and child feeding index is associated with child nutritional status in rural China. *Early Human Development* **85,** 247-252.

Zong X.-N. & Li H. (2014) Physical growth of children and adolescents in China over the past 35 years. *Bulletin of the World Health Organization* **92,** 555-564.

Annexes: questionnaires and forms

Annexes

Form 1: Feeding Pracitces and Styles of Caregivers in Derashie Special District; southern Ethiopia

1. Identification		
001	Family ID	
002	Name of the Kebele	

2. Socio demographic characteristics (household level information)

S.no	Questions	Responses	Skip to	Code
101.	Who feeds the child most of the time? *(Any person who will take care of the child and feed him/her in the home in most of the times.)* *Questions from 104 to 110 are about the immediate caregiver*	1. Mother 2. Father 3. Sister 4. Grand mother 5. Relatives 6. Other female Gardner 7. Other specify…………		
102.	Does mother/caregiver live in the same house?	1. Yes 2. No		
103.	Age of the mother/caregiver *(help her to remember by using social or cultural events if not documented)*	________ completed years		
104.	Marital Status	1. Married 2. Single 3. Widowed 4. Divorced 5. Separated 6. Other specify…………		
105.	Ethnicity	1. Derashe 2. Gawwada 3. Konso 4. Gamo 5. Other specify…………		
106.	Religion	1. Protestant 2. Orthodox 3. Muslim 4. Other specify…………		
107.	Occupational Status	1. Farmer 2. House wife 3. Daily laborer 4. Merchant 5. Self-employed 6. Government Employee 7. Other specify. ………		
108.	Literacy status *(Illiterate= no formal education attended)*	1. Illiterate (cannot read & write) → 2. Literate	110	

109.	What is level of education attained?	1. Only read and write without formal education 2. Elementary (grade 1-6) 3. Junior secondary school (grde 7-8) 4. Secondary school (grade 9-12) 5. Secondary school & certificate 6. College and above 7. Other, specify………..		
110.	Who is the head of the household?	1. The father 2. Mother 3. Other, specify _________________		
111.	Occupation of the head of the household	1. Farmer 2. House wife 3. Daily laborer 4. Merchant 5. Self-employed 6. Government Employee 7. Other specifies. ………		
112.	Monthly income of the head of the households if employed?	________________________________ 98. Farming and related activities are the only source of earning		
113.	Educational status of the head of the households? *(Illiterate= no formal education attended)*	1. Illiterate (cannot read & write) → 2. Literate	115	
114.	What is the highest level of education attained by the head of the household?	1. Read and write 2. Elementary (grade 1-6) 3. Junior secondary school (grade 7-8) 4. Secondary school (grade 9-12) 5. Secondary school & certificate 6. College and above 7. Other, specify………..		
115.	Family size (*number of permanent members of the household*)	____________________		
116.	Number of under five children in the household including the index child	____________________		
117.	Number of under two children in the household including the index child?	____________________		
118.	Monthly income in birr of the immediate mother/caregiver if	____________________		

	employed?	98. Housewife		
119.	Monthly income in birr of the household? (*estimate using the current price of own produces and divide that for the months*)	……………Birr		
120.	Are there conditions in which sometimes the immediate mother/caregiver is not available in the home?	1. Yes 2. No →	123	
121.	If yes, where will the child stays until the immediate mother/caregiver comes back home?	1. In the same home (in his/her home) 2. In the neighbor's house 3. Will travel with immediate mother/caregiver 4. others, specify ______________		
122.	If yes to Q120, Who else feeds the child in most of the times?	1. Anybody available 2. Father 3. Sister 4. Grand mother 5. Other relatives 6. Other specify…………		
123.	Is there death of child less than 5 years in the house hold?	1. Yes 2. No →	**125**	
124.	What is the cause of death (*probe about the diagnosis made at the health institution prior to the death of the child; or register if the mother/caregiver knows what the child was ill about*)	1. Malaria 2. Fever 3. Cough 4. Diarrhea 5. Malnutrition 6. Others, specify ______________		
125.	Parity of the caregiver? *(provide description for the mother)*	______________________		
3. Child related information				
S.No	**Questions**	**Responses**	**Skip to**	**Code**
201.	Age (in months) (*use cultural or social events to be able to guide the mother/caregiver to remember the age or birthdate*)	……… Months		
202.	Sex	1. Male 2. Female		

203.	Where did you take the child when he/she gets ill?	1. Health institutions 2. Traditional healers 3. Home management 4. Other specify _______________		
204.	Does the child get vaccinated?	5. Yes 6. . No →	*206*	
205.	For which condition was immunization provided? (*multiple answers are possible help the mother/caregiver about the schedules and route of administration to be able to determine which vaccine the child received; more than one response is possible depending on the age*)	1. Tuberculosis (BCG) 2. Polio 3. Diphtheria, Pertussis Tetanus 4. Measles 99. I don't know		
206.	Does the child has older brother or sister?	1. → 2.	**208**	
207.	Birth interval between the index child and the immediate older child? (*the count is between the time of the two births*)	__________months 99. I don't know		
208.	Does the child has younger brother or sister?	1. Yes 2. No →	**210**	
209.	Birth interval between the index child and the immediate younger child? (*the count is between the time of the two births*)	**1.**__________months **2.**Has no younger brother or sister **99.** I don't know		
210.	Birth order of the child in the family?	__________		
1. Breast feeding & complementary feeding knowledge & practices				
S.no	**Questions**	**Responses**	**Skip to**	**Code**
301.	Is the child being breastfed currently?	1. Yes -------------------------→ 2. No	303	

302.	At what age did the child stopped breast feeding?	______ Months 98. Since birth 99. I don't know		
303.	What was the frequency of breast feeding over the previous day? (How *many times the child was breast feed over the last 24 hours?*)	_________times		
304.	Up to when you planned to breast feed your child?	__________ months		
305.	At what age should infants start complementary food?	________ months		
306.	With what should complementary feeding started?	1. Liquid foods 2. Semisolid foods 3. Solid foods 4. I don't know		
307.	Do you separately prepare food for index child, using a separate material?	1. Yes 2. No		
308.	Do this child eat from a separate bowl (plate)?	1. Yes 2. No		
309.	What was the frequency of complementary feeding over the previous day? (*How many times the child was given complementary foods over the last 24 hours?)*	_________times		
310.	Do you give water to the child before six month?	1. Yes → 2. No	**312**	
311.	At what age did you start to give foods/drinks additional to breast milk to the child?	______ months 99. I don't know		
312.	Did you start to give anything additional to breast milk to the child before his/her 6 months of age?	1. Yes 2. No →	**312**	
313.	What were you giving additional to breast milk before six months of age?	1. Cow's milk 2. Water 3. Powder milk 4. Porridge 5. Family food 6. Other specify _______		

314.	Why did you start to give this (these) foods additional to breast milk before six month?	1. The infant was not satisfied with breast milk alone 2. Baby always crying 3. Not enough breast milk 4. Baby is hungry 5. Other specify ______________		
315.	What do you use to feed the child?	1. Hand 2. Cup and spoon 3. Cup 4. Bottle 5. Others, specify ______________		
316.	When children will be able to self-feed (using finger or spoon)?	1. _________ Months 99. I don't know		
317.	What is the consistency of complementary food you are giving to the child currently?	1. Solid 2. Liquid 3. Semi solid		
318.	What food/fluid is the child mostly receiving currently?	1. Cow's milk 2. Powder milk 3. Gruel 4. Family (adult) food 5. Other, specify ______________		
319.	Do you use bottle to feed your child?	1. Yes 2. No		
320.	How many times per day do you give additional food to your child?	________________times		

4	Child feeding style questions			
S.no	**Questions**	**Responses**	**Skip to**	**Code**
	Responsive feeding- Behavioral domains			
401.	Do you encourage your child either physically or verbally to eat or finish the portion of food given? *Interviewer: provide more explanation to the caregiver about the feeding behavior but it shouldn't be leading*	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify______________		
402.	What do you do to positively encourage your child?	____________________________________ ____________________________________ ____________________________________		
403.	Why do you feed your child in such a way?	____________________________________ ____________________________________ ____________________________________		
404.	Do you talk to the child about food or other things during meals?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify ______________		
405.	Do you sometimes give food to the child as a reward for good behavior?	5. Most of the time 6. Sometimes 7. Not at all 8. Other, specify ______________		
406.	Do you provide food when the child asks for food?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify ______________		
407.	Do you sit down with the child at meals?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify _________		
408.	How do you help your child to eat?	__________________________ __________________________ __________________________		
409.	Do you respond depending on child's hunger cues|?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify ______________		
410.	What signs of hunger (interest to eat) do you know?	1. Crying 2. Yawing 3. The child asks for food physically/verbally 4. Other, specify _____		

411.	How do you know if your child is full?	________________________ ________________________ ________________________ ________________________		
412.	How do you know your child needs additional food during the eating event?	________________________ ________________________ ________________________ ________________________		
413.	Did you teach (demonstrate) your child how to eat?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify ______________		
414.	When does the child eat more in most of the times? (more than one answer is possible)	________________________ ________________________ ________________________ ________________________		
415.	Do you encourage the child to eat when he/she refuses to eat	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify ______________		
416.	What do you do when your child refuses to eat ? (*more than one answer is possible*)	1. Encourage verbally and physically 2. offer alternative foods 3. offer the same food later 4. force the child to eat 5. I will not try anything once the child refuses to eat 6. Other, specify__________		
Responsive feeding style – belief domain				
417.	Children tend to eat more when they are encouraged to eat either physically or verbally	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don’t know		
418.	The child is capable of knowing how much he/she has to eat	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don’t know		

419.	The child is capable of knowing when he/she has to eat	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
420.	Child refuses to eat when he/she is full?	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
421.	Child refuses to eat when he/she is ill	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
422.	Child refuses to eat when he/she has not adequate attention from immediate caregiver	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
423.	Child refuses to eat when he/she is not encouraged verbally or physically.	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
424.	Child should be given food in response to hunger cues	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
425.	Child has favorite foods and the caregiver should respond to child preferences	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		

426.	Child eats more food when given on a separate plate, encouraged, and presented with alternate food in case he/she doesn't like.	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
427.	Children know when they are full or hungry and express them	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
Controlling feeding – Behavior domain				
428.	The child eats or finished the entire portion of the food given when forced or punished by the caregiver	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify		
429.	Do you withhold food from your child as a punishment?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify ______________		
430.	Do you make the decision as to when the child has to eat?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify _________		
Controlling feeding – belief domain				
431.	You are well able in feeding your child	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know _________		
432.	Responding to a child's food preferences will spoil a child	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know ____________		

433.	Child tends to eat more when he/she is forced to eat	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know __________		
434.	Children should not be allowed to eat during family meal time	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
435.	Children should only be allowed to eat at set meal times	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
436.	Children should always eat all the food on their plates	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
437.	Mothers/caregivers should make sure that their children eat enough	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know___________		
438.	Children should be allowed to eat food they don't like as those foods are often good for them	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know __________		
439.	Mothers/caregivers have to be sure that their children do not eat too much	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know _________		
Laissez faire feeding- behavior domains				

440.	How often do you only bring the food to the child's mouth and leave the decision to him/her either to eat or not (or to finish or not)?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify _________		
441.	How often keep track of what food the child has eaten?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify		Reverse coded
442.	How often do you support the child while eating or drinking from a cup/bottle?	1. Most of the time 2. Sometimes 3. Not at all 4. Other, specify ______________		Reverse coded
Laissez faire feeding- belief domains				
443.	A child should eat whatever is available in the house	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
444.	It is OK to leave the cup/bottle for the child to eat or drink from.	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		
445.	Give food whenever there is something to give to the child?	1. Strongly agree 2. Agree 3. Neutral 4. Disagree 5. Strongly disagree 99. I don't know		

Annexes

Questionnaire administered to the study participants to collect data on feeding patterns and practices and child growth in two season in the catchment of the Gilgel Gibe Field Research Center of Jimma University

1. Background Information		
101.	Child ID	|__|__|| __|
102.	Name of the Woreda	
103.	Name of the Kebele	
104.	Name of Gere	
105.	Name of the "got"	
106.	House number	
107.	Name of the household head	

2. Socio economic and demographic characteristics of caregivers			
	Questions	Coding categories	skip
201.	Relation of the respondent to the index infant	Mother ....1 Elder sibling ....2 Care giver ....3 Other 4	.
202.	What is the age of the mother/care giver (in years)	_____________	
203.	Marital status of the mother/ care taker	Married ....1 Single ....2 Divorced ....3 Separated ....4 Widowed 5	
204.	Religion of the mother/care giver	Islam .... 1 Orthodox ....2 Protestant..3 Catholic ....4 Other (specify)___________	
205.	Ethnicity of the mother or care taker	Oromo ....1 Amhara ....2 Tigre ....3 Answer refused ....77 Other (specify)___________	
206.	Educational status of the mother /care taker	Unable to read and write .... 1 Only read and write........................... 2 Attended formal education (specify the level of education attained) ______________	

207.	Educational status of the father	Unable to read and write .... 1 Only read and write ....2 Attended formal education (Specify the level of education attained) _____________________	
208.	How many children under 5 years of age usually live in this household?	Number children under 5 years ☐☐	
209.	What is the current main source of drinking water for members of your household? *Circle ONLY ONE answer*	Piped water ....1 Tube well or borehole ....2 Protected well ....3 Unprotected well ....4 Water from protected spring ....5 Water from unprotected spring ....6 Rainwater..................................10 Surface source (stream, river, pond, lake, canal) ....12 Don't know ....98 Answer refused ....77 Other (specify) .... 96 ________________________________	
210.	Do you treat your water in any way to make it safer to drink? *Circle ONLY ONE answer* *If yes, continue.* *If other answer, jump to question 30.*	Yes ....1 No ....2 Don't know ....98 Answer refused 77	
211.	What do you do <u>now</u> to the water to make it safer to drink? *Circle ALL applicable answers*	Boil ....1 Add bleach, chlorine or Agar ....2 Mix with leaves ....3 Strain it through a cloth ....4 Use a water filter ....5 Let it stand and settle ....6 Answer refused ....77 Don't know ....98 Other (specify) ....9 ________________________________	

212.	Where do members of your household usually go to relieve themselves? *Circle ONLY ONE answer*	Flush toilet ....1 Pit latrine with slab …………………2 Pit latrine without slab/open pit…….3 Composting toilet ....4 Bush or field ....5 On ground within compound ....6 Answer refused ....77 Other (specify) ....96 _____________________________ Don't know 98	
213.	What is the material from which your house is made up of?	Materials	
	214.1. Roof *Record observation*	Thached/Grass ....1 Corrugated iron sheet ....2 Other (specify)..........................96	
	214.2. Wall *Record observation*	Cement and wood ....1 Cement, wood and mud ....2 Wood and mud.............................3 Stone and mud.............................4 Wood...5 Other (specify)...........................96	
	214.3. Floor *Record observation*	Cemented....................................1 Ceramic tiles ....2 Carpet ....3 Dung ....4 Wooden.......................................5 Mud 6	
214.	What is now the primary source of income for this household? *Circle ONLY ONE answer*	Farming, including cash crops......1 Livestock ....2 Employment/salary.......................3 Petty trading (including sale of fire-wood, charcoal, grass, local brewery) Daily labor ....5 Handicrafts/artisan ....6 Remittances ....7 Answer refused ....77 Don't know ....98 Other (specify) ....96 _____________________________	
215.	I will now mention some animals, and I would like you to tell me how many animals of each type you have *Fill in NUMBER of each type*	Plow oxen: ____ Cows: ____ Hephers: ____ Calves: ____ Bulls: ____	

	of animal	Sheep and goats: Horses, donkeys, mules: Chickens:	
216.	How much land does your household own? Write in number of local units and the name of the local unit	Number of local units: Name of local unit: "Fechasa"……….. 1 "Timad" ………… 2 "Hectare"……….. 3 Other ………….. ....96 Specify: ________________ Don't know how much land………..98	
217.	Does your household have: *Circle 1 Or 2 For Each Item*	Yes No Electricity/generator: ....1 0 A sewing machine: ....1 0 A radio: 1..0 A television: ……………............................. 1 0 A mobile telephone: ....1 0 A fixed telephone: …………………1 0 A refrigerator: 1 ....0 A table: ……………………………1 0 A chair:……………………………1 0 A bed: ………………………………1 0 An electric "mitad": …………………1 0 A bicycle………………… 1 0 A motorcycle: ………………………1 0 A "Bajaj": ……………………………1 0 A kerosene or pressure lamp: ……1 0 A kerosene /electric stove:…………1 0	

3. Child characteristics and child feeding practice of the caregiver

301.	Ethiopian date of birth? 1. Reported by the mother 2. Recorded from birth certificate 3. Recorded from immunization card 4. Estimated using local events calendar	1. |__|__|/|__|__|/|__|__| (dd/mm/yy) 2. |__|__|/|__|__|/|__|__| (dd/mm/yy) 3. |__|__|/|__|__|/|__|__| (dd/mm/yy) 4. |__|__|/|__|__|/|__|__| (dd/mm/yy)	
302.	Sex of the child	Male…....1 Female... 0	
303.	Is the child ever breastfed?	Yes…….. 1 No………0	0⇨ 308
304.	Is the child currently breastfed?	Yes…….. 1 No ……...2	
305.	Was the child fed only breast milk over the last 24 hours?	Yes ……..1 No………2	
306.	How many times did you breastfed the child over the last 24 hours (day and night)? *<u>Probe the mother:</u> How many times did you breastfeed last night between sunset and sunrise and?* *How many times did you breastfeed yesterday during the daylight hours?*	|__|__|Times	
307.	For how long after birth was the child fed only breast milk? *(Probe the mother: for the additional foods, water, tea, fenugreek , etc,)* *Please use the local calendar to assist the mother to remember the duration*	|__|__|-days/ months Don't know---------------	
308.	Have you given additional food other than breast milk over the past seven days including yesterday?	Yes...... 1 No....... 0	
309.	Now I would like to ask you about the types of foods [name] has been fed over the last seven days, including yesterday. How many days during last seven days was [*name*] given each of the following? FOR EACH ITEM GIVEN AT LEAST ONCE IN LAST SEVEN DAYS, ASK:	Last 7 days number of days	Yesterday/ last night number of times the item was given

<table>
<tr><td></td><td>In total, how many times yesterday during the day or at night was [name] given:</td><td></td><td></td></tr>
<tr><td>310.</td><td>Grains /tubers/ roots;</td><td></td><td></td></tr>
<tr><td></td><td>1. Any porridge or gruel (made from grains other than teff)?</td><td>1. |__|__| DK ……8</td><td>1. |__|__|</td></tr>
<tr><td></td><td>2. Bread, pasta, rice, biscuits, cookies or any other food made from oats, maize, barley, wheat, sorghum, millet, or other grain?</td><td>2. |__|__| DK ……8</td><td>2. |__|__|</td></tr>
<tr><td></td><td>3. Any food made from teff, like injera, kita or porridge?</td><td>3. |__|__| DK ……8</td><td>3. |__|__|</td></tr>
<tr><td></td><td>4. Any white potatoes, white yams, bulla, kocho, cassava, or any other foods made from roots?</td><td>4. |__|__| DK …… 8</td><td>4. |__|__|</td></tr>
<tr><td></td><td>Legumes/nuts;</td><td></td><td></td></tr>
<tr><td></td><td>5. Any foods made from beans, peas, chickpea, lentils or pulses?</td><td>5. |__|__| DK …… 8</td><td>5. |__|__|</td></tr>
<tr><td></td><td>6. Any nuts or seeds such as peanuts (lewiz), sesame (selit) or sunflower seeds?</td><td>6. |__|__| DK …… 8</td><td>6. |__|__|</td></tr>
<tr><td></td><td>fruits and vegetables</td><td></td><td></td></tr>
<tr><td></td><td>7. Any pumpkin, carrots, squash or sweet potatoes that are yellow or orange inside?</td><td>7. |__|__| DK …… 8</td><td>7. |__|__|</td></tr>
<tr><td></td><td>8. Any ripe mangoes, papayas?</td><td>8. |__|__| DK …… 8</td><td>8. |__|__|</td></tr>
<tr><td></td><td>9. Any dark green, leafy vegetables like kale (gomen), spinach (kosta), lettuce (salata)</td><td>9. |__|__| DK …… 8</td><td>9. |__|__|</td></tr>
<tr><td></td><td>Other fruits and vegetables</td><td></td><td></td></tr>
<tr><td></td><td>10. Any other fruits or vegetables?</td><td>10. |__|__| DK …… 8</td><td>10. |__|__|</td></tr>
<tr><td></td><td>Flesh foods;</td><td></td><td></td></tr>
<tr><td></td><td>11. Any liver, kidneys, heart or other organ meats?</td><td>11. |__|__| DK ……8</td><td>11. |__|__|</td></tr>
<tr><td></td><td>12. Any beef, lamb, goat?</td><td>12. |__|__| DK …….8</td><td>12. |__|__|</td></tr>
<tr><td></td><td>13. Any chicken, duck or other birds?</td><td>13. |__|__| DK …… 8</td><td>13. |__|__|</td></tr>
<tr><td></td><td>14. Any fresh or dried fish or sea foods</td><td>14. |__|__| DK ……. 8</td><td>14. |__|__|</td></tr>
<tr><td></td><td>Eggs</td><td></td><td></td></tr>
<tr><td></td><td>15. Any eggs?</td><td>15. |__|__| DK …….8</td><td>15. |__|__|</td></tr>
<tr><td></td><td>Milk and dairy products</td><td></td><td></td></tr>
<tr><td></td><td>16. Any cheese or yogurt?</td><td></td><td></td></tr>
<tr><td></td><td>17. Any Cerifam, Fafa, Milupa, Babylac, Mother's Choice or other commercially fortified baby food?</td><td></td><td></td></tr>
<tr><td></td><td>Fats and oils</td><td></td><td></td></tr>
<tr><td></td><td>18. Any food made with oil, fat, butter,</td><td></td><td></td></tr>
</table>

	or galighee, red palm oil.	16. |__|__| DK …….8 17. |__|__| DK …….8 18. |__|__| DK …….8	16. |__|__| 17. |__|__| 18. |__|__|
311.	How many times was [name] fed mashed or pureed food or solid or semi-solid food yesterday during the day or at night? *(If 7 or more times, record '7')*	No. of times |__|__| DK ………… 98	
312.	Do you make special processing to improve the nutritional value / taste or other qualities of the additional food you give to your child?	Yes ………………..1 No …………………0	0⇨ **314**
313.	What processing techniques you use at home to prepare food for your child?	Removal of bran of grains and cereals……………1 Fermentation…………2 Soaking ……………..…3 De-hulling ……………4 Others………………..96 Specify_____________ Answer refused………77	
314.	Did (*name*) drink anything from a bottle with a nipple yesterday or last night?	Yes ………………….1 No …………………..0	

4. Child morbidity

401.	Try to remember the last two weeks. During this period, has *(name)* had diarrhea? **Definition**: *diarrhea is defined as having three or more loose stools during a 24-hour period* *circle only one answer*	Yes ....1 No ....2 Answer refused ....7 Don´t know 8
402.	Try to remember the last two weeks again. Has *(name)* had a cough? *circle only one answer*	Yes ....1 No ....2 Answer refused ....7 Don´t know 8
403.	Try to remember the last two weeks again. Has *(name)* had <u>difficulty breathing</u>? *Difficulty of breathing involves : chest in-drawing, grunting, and unusually rapid breathing* *circle only one answer*	Yes ....1 No ....2 Answer refused ....7 Don´t know 8

404.	Try to remember the last two weeks again; has *(name)* had a fever? *circle only one answer*	Yes .... 1 No .... 2 Answer refused .... 7 Don´t know 8	
5. Anthropometric measurement of the child			
501.	Child ID	|__|__|| __|	
502.	Child's weight.	Kilograms (kg) |__|__|.|__|	
503.	Child's length *Please measure length in recumbent position*	Length (cm) |__|__|| __|.|__|	
504.	Oedema	Present …………………1 Absent…………………..2 Not checked……………..3	

Questionnaires and forms used for the OME3JIM clinical trial in the catchment of Gilgel Gibe Field Research Center of Jimma Univeristy

1. የተሳታፊነትግምገማቅጽ/Unka hirmaatota filachuuf tajaajilu ; Screening form

ጠቅላላመረጃ/Odeeffannoo waliigalaa Background information |__| (**I**: ተካቷል/Fiilatameera/Included **E**: አልተካተተም/hin-filatamne/ Excluded)

<table>
<tr><td>S1. ቅጹየተሞላበትቀን (እ.ኢ.አ)/Guyyaa unki kuni itti guutame(ALI)/Screening date (E.C.)</td><td>|__|__|/|__|__|/|__|__|ቀን/ወር/ዓ.ም./gg/jj/ww dd/mm/yy</td></tr>
<tr><td>S2. የጥናቱቦታ/iddoo qo’annoo/Study site
1. ደጎሶ/Degosso /Degoso 3. ደነባ/Dannaba Deneba 5. ሰንዳቦ/Assandaaboo/Assendabo
2. ዲምቱ/Dimtuu/ Dimtu 4. ሰርቦ/Sarboo /Serbo 6. ቡርቃ/Burqaa /Burka</td><td>|__|</td></tr>
<tr><td>S3. የህፃኑመለያኮድ/Lakk. Eenyummaa Daa’imaa/ Child ID</td><td>C |__|__|__|</td></tr>
<tr><td>S4. የህፃኑስም/Maqaa daa’imma /Name child</td><td>|__|__|__|__|__|__|__|__|__|__|__|__|__|</td></tr>
<tr><td>S5. የልደትቀን (እ.ኢ.አ.)/Guyyaa dhaloota daa’imma (ALI) /Ethiopian DOB
S5a. በእናትየውየተነገረ/Kan haati daa’imma himte/Reported by the mother
S5b. ከልደትካርድየተሞላ/Sartifikeeta/ttii dhaloota irra argame/Birth certificate
S5c. ከክትባትካርድየተሞላ/Kan Kaardii talaallii irra argame/Immunization card
S5d. በየአካባቢክስተቶችየጊዜመቁጠርያየተገመተ/Mudannoo Naannoo fayyadamuun kan tilmaamame /Calendar</td><td>|__|__|/|__|__|/|__|__|ቀን/ወር/ዓ.ም./gg/jj/ww/ dd/mm/yy
|__|__|/|__|__|/|__|__|ቀን/ወር/ዓ.ም./gg/jj/ww/ dd/mm/yy
|__|__|/|__|__|/|__|__|ቀን/ወር/ዓ.ም./gg/jj/ww/ dd/mm/yy
|__|__|/|__|__|/|__|__|ቀን/ወር/ዓ.ም./gg/jj/ww/ dd/mm/yy</td></tr>
<tr><td>S6. የህጻኑእድሜ/Umurii daa’immaa/Age infant</td><td>|__|__| ወር/Ji’aan/months</td></tr>
<tr><td>S7. ህጻኑመንታአለዉ?/Daa’imni lakkuu dha?/Twin brother or sister?
1. አዎን/Eeyyen/Yes 0. አይአይደለም/Lakki /No 9. አላዉቅም/hinbeeku /Don’t know</td><td>|__|</td></tr>
<tr><td>S8. በቤተሰቡዉስጥስንትከአምስትአመትበታችህጻናትአሉ?/Mana kana keessa daa’imma waggaa 5 gadi meeqatu jiru?/Under-5 children in household?</td><td>|__|</td></tr>
<tr><td>S9. የህጻኑጾታ/Saalaa mucaa/ Sex of the child
1. ሴት/Dhala /Female 2. ወንድ/Dhiira /Male</td><td>|__|</td></tr>
<tr><td>S10. የህጻኑ/ኗዋናተንከባካቢወላጅእናትናት?/Guddiftuun daa’imaa haadha deessee dha?/ Mother is primary caregiver?
1. አዎንናት/Eeyyye ishi/Yes 0. አይአይደለችም/lakki ishii miti/No
9. አላዉቅም/hinbeeku / Don’t know</td><td>|__|</td></tr>
<tr><td>S11. የእናትመለያኮድ/Lakk eenyummaa haadhaa/Mother ID</td><td>M |__|__|__|</td></tr>
<tr><td>S12. የእናትስም/Maqaa haadha/Name mother</td><td>|__|__|__|__|__|__|__|__|__|__|__|__|__|</td></tr>
<tr><td>S13. የእናትእድሜ/Umurii haadha/Age mother</td><td>|__|__| ዓመት/waggaan/ Years</td></tr>
</table>

S14. ስንትልጆችወልደሻል፣ሞቶየተወለደወይምበወሊድጊዜናከዛበኋላየሞተንይጨምራል/ Ijoollee meeqa deessettaa? daa'imma midhame dhalatee yknYeeroo da'umsaa fi da'umsa booda kan midhamee yoo jiraates ni dabalata/ Parity	|__|__|
S15. የቤትቁጥር/Lakk manaa/House ID	|__|__|__|__|
S16. የቤተሰቡኃላፊስም/Maqaa abbaa warraa/ HH head	|__|__|__|__|__|__|__|__|__|__|__|__|__|
S17. የጤናኤክስቴንሽንሰራተኛስም/Maqaa hojjeettuu Ekstenshinii fayyaa/HEW	|__|__|__|__|__|__|__|__|__|__|__|__|__|
S18. የገሬዉስም/Maqaa Garee/Gere	|__|__|__|__|__|__|__|__|__|__|__|__|__|
S19. የዞንስም/Maqaa Zoonii/Zone	|__|__|__|__|__|__|__|__|__|__|__|__|__|
S20. የቀበሌዉስም/Maqaa Ganda/Kebele	|__|__|__|__|__|__|__|__|__|__|__|__|__|
S21. የምትኖሪበትንአካባቢየመልቀቅእቅድአለሽ?/Karoora bakka jireenyakee kana gadhiissuuf yaada qabda?/Planning to leave area permanently? **1.** አዎንአለኝ/Eeeyyeen/ Yes **0.** አይየለኝም/ Hin qabu/ No **9.** አላዉቅም/hinbeeku / Don't know	|__|
S22. ከ1 ወርበላይከምትኖሪበትአካባቢርቆየመሄድእቅድአለሽ? Ji'a tokkoo fi isaa olif bakka biraa faggaattee deemuf karoorfatteettaa?/Planning to leave area more than 1 month? **1.** አዎንአለኝ/Eeeyyeen/Yes **0.** አይየለኝም/ Hin qabu/ No **9.** አላዉቅም/hinbeeku / Don't know	|__|

የህጻንየሰዉነትልኬት/Safartii qaama Daa'imaa Child anthropometry

S23. ክብደት (በኪ.ግ.)/Ulfaatina/Weight (in kg)	|__|__|.|__|ኪ.ግ.kg
S24. ርዝመት (በሴ.ሜ.)/dheerina/Length (in cm)	|__|__|__|.|__| ሴ.ሜ.cm
S25. ክብደትለርዝመት/Ulfaatina gita hojjaan/Weight-for-Length Z-score	በ/Kabetween |__|__| SDእና/fi |__|__| SD መሃል/ gidduu
S26. የሁለቱምእግሮችእብጠት/Dhiittoo miilla lamaanii/Bilateral edema **1.** አለ/ni jira/ Present **0.** የለም/hin jiru/Absent **9.** ለማሳየትፍቃደኛአይደሉም/ agarsisuuf ni didan/Refusal	|__|__|

የህጻኑ/ኗክብደትለርዝመትከ-3 በታችከሆነወይምየሁለቱምእግሮችእብጠትካለለተቆጣጣሪውአሳውቅ/ቂ፡፡

Gitni Ulfaatina daa'imaa hojjaan yoo madaalamuu sadii (<3) gadi YKN Dhiittoon miilla daa'imaa lamaanirra yoo Jiraate, to'ataa keetiif bilbilli

If child's Weight-for-Length Z-score is <-3 or if he/she has bilateral edema, please call supervisor.

የህጻንአመጋገብ/Nyaachisa daa'imaaChild feeding

Annexes

S27. ህጻኑ/ኗባለፈዉአንድወርውስጥጠጣርወይምወፈርያለፈሳሽምግብቢያንስበቀን 1 ጊዜሲመገብ/ስትመገብነበር?/Ja'a darbe keessatti daa'imini kee guyyaatti yoo xiqqaate si'a tokko nyaata furdaa/ jajjaboo mooqa furdaa nyaateera/tti?/ Has the child eaten solid and/or semi-solid foods at least once per day in the last month?

1. አዎንነበር/Eeyyeen nyaateera/Yes **0.** አይአልነበረም/Lakki hinnyaatne /No **9.** አላዉቅም/hinbeeku/Don't know

S28. ህጻኑ/ኗበቀን (ቀንናለሊትንጨምሮ) ስንትግዜጡትይጠባል/ትጠባለች?/Daa'imni kee guyyaa gutuu (halkan dabalatee) si'a meeqa harma hodheera/hooteetti?/How many times per day (night included) is the child being breastfed?

0. ጡትአይጠባም/አትጠባም/Hin hodhu/hootu/Not breastfed **1.** በቀንከ1-3 ጊዜ/Guyyaatti si'a 1-3 /1-3 times per day

2. በቀንከ4-6 ጊዜ/Guyyaatti si'a 4-6/4-6 times per day **3.** በቀንከ7-9 ጊዜ/Guyyaatti si'a 7-9/7-9 times per day

4. በቀን 10 ጊዜናከዛበላይ/guyyaatti si'a 10'f isa ol/≥10times per day **9.** አላዉቀውም/Hinbeeku/ Don't know

የህጻንናእናትየጤናሁኔታ/Haal fayyaa daa'imaa fi haadhaa Child's and mother's health status

		ህጻን/Daa'ima Child		እናት/Haadha Mother
1. በአሁኑግዜዓልሚ-ምግብወይምንጥረ-ነገርእየወሰደ/ችነዉ?/Yeroo ammaa nyaataa dabalataa kan biroo fudhachaa jira/rtii?/ Currently taking nutritional supplements? **1.** አዎንነዉ/Eeyyeen /Yes **0.** አይአይደለም/Lakki /No **9.** አላዉቅም/Hinbeeku/Don't know	**S29.**	|__|	**S32.**	|__|
2. በአሁኑግዜህክምናላይነው/ናት፤መድሃኒትእየወሰደ/ችነዉ?/Yeroo ammaa qoricha fudhchaa / wal'aanamaa jiraa/rtii?/Currently under medical treatment? **1.** አዎንነዉ/Eeyyeen /Yes **0.** አይአይደለም/Lakki/No **9.** አላዉቅም/Hinbeeku/Don't know	**S30.**	|__|	**S33.**	|__|
3. የተፈጥሮየአካልመጉደል/ የጡትችግርአለበት/ባት?/Dhibeen wajjin kan dhalate/dhibee harmaa qabaa/dii?/ Congenitalmalformations/breast problems? **1.** አዎንአለ/ni jira /Present **0.** አይየለም/hin jiru /Absent **9.** ፍቃደኛአይደሉም/deebii kennuu ni didan/ Refusal	**S31.**	|__|	**S34.**	|__|

ህጻኑ/ኗወይምእናትየውዓልሚ-ምግብወይምንጥረ-ነገርእየወሰደ/ችከሆነ፤ህክምናላይከሆነ/ች፤ወይምየተፈጥሮየአካልመጉደልካለበት/ካለባትለተቆጣጣውአሳውቅ/ቂ፡፡

Yoo daa'imini YKN haati nyaata dabalataa kan biroo fudhachaa jiru ta'e akkasumas yoo wal'aansa fi dawaa fudhachaa jiru ta'e YKN dhibee wajiin dhalatani yoo qabaatan, gara to'ataa keetti bilbilli.

If child or mother are taking supplements, or is under treatment, or have congenital malformation, please call supervisor.

2. Socio-Economic & Demographic Form

Instruction: *Except questions **E33 E34 & E35**, all questions should be directed to the mother; whenever necessary, the father/household head can assist the mother.*

*መመሪያ፡ከመጨረሻዎቹሶስትጥያቄች***(E33 E34 & E35)***በስተቀርሁሉንምጥያቄዎችእናትውትጠየቅ፤አስፈላጊሲሆንየቤተሰቡኃላፊእናትየውንሊያግዝይችላል፡፡*

<u>Background information ጠቅላላመረጃ</u>

E1. Ethiopian date of interview የቃለ-መጠይቁቀንእ.ኢ.አ	|___|___|/|___|___|/|___|___| (dd/mm/yy) (ቀን/ወር/ዓ.ም.)
E2. Child ID የህጻኑመለያቁጥር	**C** |___|___|___|
E3. Name child የህጻኑስም	|___|___|___|___|___|___|___|___|___|___|___|___|
E4. Sex (**1.** Female / **2.** Male) ጾታ (1. ወንድ/ 2. ሴት)	|___|
E5. Name Mother የእናትስም	|___|___|___|___|___|___|___|___|___|___|___|__
E6. Name HH head የቤተሰቡኃላፊስም	|___|___|___|___|___|___|___|___|___|___|___|___|
E7. Name HEW የጤናኤክስቴንሽንሰራተኛስም	|___|___|___|___|___|___|___|___|___|___|___|___|
E8. House Number የቤትቁጥር	|___|___|___|___|___|___|___|___|___|___|___|___|
E9. Name Gere የገሬዉስም	|___|___|___|___|___|___|___|___|___|___|___|___|
E10. Name Zone የዞኑስም	|___|___|___|___|___|___|___|___|___|___|___|___|
E11. Name Kebele የቀበሌዉስም	|___|___|___|___|___|___|___|___|___|___|___|___|
E12. Phone Number ስልክቁጥር	|___|___|___|___|___|___|___|___|___|___|

Demographic & socio-economic information የዲሞግራፊክናማህበራዊ-ኢኮኖሚያዊመረጃ

E13. What is the age of the mother? የእናትእድሜ |__|__| years በዓመት

E14. What is the marital status of the mother? የእናትየጋብቻሁኔታ

1. Married ያገባች**2.** Single ያላገባች**3.** Divorced የፈታች**4.** Separated የተያየች**5.** Widowed ባልየሞተባት

E15. What is the religion of the mother? የእናትኃይማኖት

1. Islam እስላም **2.** Orthodox ኦርቶዶክስ**3.** Protestant ፕሮቴስታንት**4.** Catholic ካቶሊክ**5.** Other (specify) ሌላ፤ይጠቀስ: ______)

E16. What is the ethnicity of the mother? የእናትብሄር

1. Oromo ኦሮሞ 2. Amhara አማራ 3. Tigre ትግሬ 4. Other (specify) ሌላ፤ይጠቀስ: _______) 9. Refusal ፍቃደኛአይደሉም

E17. What is the educational status of the mother? የእናትየትምህርትሁኔታ

1. Unable to read & write መፃፍናማንበብአይችሉም 2. Only read & write መፃፍናማንበብይችላሉ

3. Attended formal education [tick the level attained] መደበኛትምህርትተከታትለዋል፤ያጠናቀቁትየክፍልደረጃይገለጽ

1. Primary school (grades 1-8) አንደኛደረጃ፤ከ1ኛ-8ኛክፍል

2. Secondary school (grades 9-12) ሁለተኛደረጃ፤ከ9ኛ-12ኛክፍል

3. Higher education (grades > 12) ከፍተኛደረጃ፤ከ12ኛክፍልበላይ

9. Not applicable አይመለከትም

E18. What is the educational status of the father? [If the child has no father ask about the household head] የአባትየት/ትሁኔታ? [አባትከሌለስለቤተሰቡኃላፊይጠየቅ]

1. Unable to read & write መፃፍናማንበብአይችሉም 2. Only read & write መፃፍናማንበብይችላሉ

3. Attended formal education [tick the level attained] መደበኛትምህርትተከታትለዋል፤ያጠናቀቁትየክፍልደረጃይገለጽ

1. Primary school (grades 1-8) አንደኛደረጃ፤ከ1ኛ-8ኛክፍል

2. Secondary school (grades 9-12) ሁለተኛደረጃ፤ከ9ኛ-12ኛክፍል

3. Higher education (grades > 12) ከፍተኛደረጃ፤ከ12ኛክፍልበላይ

9. Not applicable አይመለከትም

E19. How many people ate at this house today (including under-5 children)? በዛሬዉእለትእዚህቤትዉስጥስንትሰዎች (ከአምስትአመትበታችህፃናትንጨምሮ) ተመግበዋል) |__|__| people ሰዎች

E20. How many people live in this house (including under-5 children)? እዚህቤትዉስጥስንትሰዎች (ከአምስትአመትበታችህፃናትንጨምሮ) ይኖራሉ |__|__| people ሰዎች

E21. How many children of under 5 years usually live (eat & sleep) in this house? እዚህቤትዉስጥስንትእድሜያቸዉከአምስትአመትበታችየሆኑህፃናትይኖራሉ (ብዙዉንጊዜእዚህቤትዉስጥይመገባሉይተኛሉ |__|__| children ህፃናት

E22. What is the current main source of drinking water for members of your household? [Tick only one answer] በአሁኑጊዜየቤተሰቡዋናየመጠጥዉኃምንጭምንድነዉ፤አንድመልስብቻምረጭ

0	Pipe water into dwelling በግቢዉስጥከሚገኝቧንቧ	**4**	Protected spring ከተከለለምንጭ	**7**	Unprotected spring ካልተከለለምንጭ
1	Public tap / standpipe ከቦኖወይምከግቢውጭከሚገኝቧንቧ	**5**	Protected dug well ከተከለለከፍትየጉድጓድዉሃ	**8**	Rain water ከዝናብዉኃ
2	Tube well / borehole ከድፍንየጉድጓድዉሃ	**6**	Unprotected dug well ካልተከለለከፍትየጉድጓድዉሃ	**9**	Don't know or Refusal አላዉቅም/ፈቃደኛአይደሉም
3	Surface water (river, dam, lake, pond, stream, canal) ከመሬትበላይከሚገኝዉኃ፤እንደወንዝ፣ግድብ፣ሃይቅ፣ኩሬ፣ጅረት፣ቦይ			**10**	Other (specify) ሌላ፤ይጠቀስ: ________)

E23. Do you treat your water in any way to make it safer to drink (excluding washing the container)? የዉኃመቅጃዉን/ማጠራቀሚያዉንከማጠብዉጭ፤የምትጠጡትንዉኃንጹህናለጤናተስማሚለማድረግየምትጠቀሙትማንኛዉምዘዴአለ

0. No አይ፤የለም 1. Yes አዎን፤አለ 9. Don't know or refusal አላዉቅም/ፈቃደኛአይደሉም

→ Continue to question E25 if the answer to E23 is No or don’t know/refused to answer

→የጥያቄ E23 መልስየለምወይምአላዉቅም/ፈቃደኛአይደሉምከሆነ፤ወደጥያቄ E25 ይዘለል፡፡

E24. What do you do to make your water safer to drink? [Tick all possible answers]

የሚጠጡትንውኃንጹህለማድረግምንያደርጋሉ፤አንድመልስብቻምረጭ

1	Boil (እናፈላዋለን)	**2**	Add bleach/chlorine (‘wuha agar’, ‘bishan gari’) (እንደዉኃአጋርናቢሻንጋሪያሉኬሚካሎችንእንጨምርበታለን)
3	Strain it through cloth (በጨርቅእናጣራዋለን)	**4**	Solar disinfection (ውኃውንለፀሃይጨረርበማጋለጥአናክመዋለን)
5	Mix with leaves (ቅጠሎችንእንጨምርበታለን)	**6**	Use a water filter (ceramic, sand, composite, etc) (በአሸዋወይምሌላየውሃማጣሪያመሳሪያእናጣራዋለን)
7	Let it stand & settle (በማስቀመጥእስኪጠልእንጠብቃለን)	**8**	Other (specify) (ሌላ፤ይጠቀስ): ________)
9	Don’t know/refusal (አላውቅም/ፈቃደኛአይደሉም)		

E25. Where do members of your household usually go to relieve themselves? [Tick only one answer]የዚህቤተሰብአባላትባብዛኛዉየትይፀዳዳሉ፤አንድመልስብቻይመረጥ)

1	Bush or field (ሽንትቤትስለሌለን፤ሜዳ/ጫካእንጠቀማልን)	**5**	Ventilated Improved Pit latrine (VIP) (የአየርማስተላለፍያያለውየተሻሻለመፀዳጃቤት)
2	On ground within compound (ሽንትቤትስለሌለን፤ግቢያችንውስጥ/ጓሮእንጠቀማልን)	**6**	Composting toilet (ለማዳበርያነትበሚጠራቀምመፀዳጃቤት)
3	Pit latrine without slab/open pit (ወለሉበደንብያልተገነባየሽንትቤትጉድጓድ)	**7**	Other (specify) (ሌላ፤ይጠቀስ): ________)
4	Pit latrine with slab (ወለሉበደንብየተገነባየሽንትቤትጉድጓድ)	**9**	Don’t know or refusal (አላውቅም/ፈቃደኛአይደሉም)

E26. Where do you usually cook? [Tick only one answer] (ባብዛኛዉምግብየምታበስሉትየትነዉ፤አንድመልስብቻይመረጥ)

1. In the house (በምንኖርበትቤትዉስጥ) 2. In separate building (በተለየማብሰያቤት/ማድቤት) 3. Outdoors (ከቤትዉጭባዶቦታላይ)

E27. What type of fuel does your household mainly use? [Tick only one answer]

(ቤታችሁምግብለማብሰልብዙውንጊዜየምትጠቀሙትምንድንነዉ፤አንድመልስብቻይመረጥ)

E28. I will now mention some animals and I would like you to tell me how many animals of each type your household have. *[Write the number of animals in the boxes provided; write ‘000’ if no animal, ‘999’ for I don’t know/refusal]* ቀጥሎየምዘረዝራቸውንእንስሳትቤተሠቡስንትእንዳለወው·ይነግሩኛል፡፡ (ለእያንዳንዱእንስሳቤተሠቡያለውብዛትበቁጥርይቀመጥ፤ምንምከሌለ*‘000’፣አላውቅምካሉ/ፈቃደኛካልሆኑ ‘999’*ይጻፍ፡፡)

Plough oxen (የእርሻበሬዎች):	Cows (ላሞች): |___|___|	Horses, donkeys, mules (ፈረሶች፣

|___|___|		አህዮችናበቅሎዎች): |___|___|
Bulls (የሚደልቡበሬዎች): |___|___|	Calves (ጥጆች): |___|___|	Chicken (ዶሮዎች): |___|___|
Hephers (ጊደሮች): |___|___|	Sheep & goats (በጎችናፍየሎች): |___|___|	Bee hives (የንብቀፎዎች): |___|___|

E29. I would like you to tell me if your household own the things that I will mention you now. *[For each item, write '1' if yes, '0' if no, '9' if don't know or refusal]*

ቀጥሎከምዘረዝራቸውቁሳቁሶችዉስጥየትኞቹበቤትዎውስጥይገኛሉ፡፡ (ለእያንዳንዱቁስበቤቱዉስጥካለ'1'፣ከሌለ '0'፣ አላውቅምካለ/ፈቃደኛካልሆኑ '9' ይፃፍ፡፡)

Electricity (የኤሌክትሪክመብራት)	|___|	Kerosene/electric stove (የጋዝ/ኤሌክትሪክምድጃ)	|___|	Chair or bench (not stool) (ወንበር/አግዳሚወንበር፤ከበርጩማውጭ)	|___|
Kerosene lamp (ፋኖስ)	|___|	Animal-drawn cart (በእንስሳየሚጎተትጋሪ)	|___|	Bed with sponge/spring mattress (የስፖንጅ/ስፕሪንግፍራሽያለውአልጋ)	|___|
Pressure lamp (ማሾ)	|___|	Bicycle (ሳይክል)	|___|	Bed with cotton mattress (የጥጥፍራሽያለውአልጋ)	|___|
Generator (ጀነሬተር)	|___|	Motorcycle or '*Bajaj*' (ሞተርሳይክል/ባጃጅ)	|___|	Bed with straw mattress (የጭድፍራሽያለውአልጋ)	|___|
Radio (ራድዮ)	|___|	Car (መኪና)	|___|	Refrigerator (ማቀዝቀዣፍሪጅ)	|___|
Television (ቴሌቭዥን)	|___|	Diesel water pump (የውኃመሳቢያፓምፕ)	|___|	Land growing coffee or khat (የቡና/ጫትመሬት)	|___|
Mobile phone (ሞባይልስልክ)	|___|	Laundry machine (ልብስማጠብያ/ላውንደሪማሽን)	|___|	Mill house (የእህልወፍጮቤት)	|___|
Landline phone (የቤትስልክ)	|___|	Table (ጠረጴዛ)	|___|	Sewing machine (የልብስመስፊያማሽን)	|___|
Electric '*mitad'* (የኤሌክትሪክምጣድ)	|___|				

E30. What is the material of the roof made from? [Make your own observation & tick only one answer] (የቤቱጣራከምንእንደተሰራእራስሽበመመልከትምረጭ፤አንድመልስብቻይመረጥ)

1. Thached/Grass (ከሳር) 2. Corrugated iron sheet (ከቆርቆሮ) 3. Other (specify) (ሌላ፤ይጠቀስ): _______)

E31. What is the material of the walls made from? [Make your own observation & tick only one answer] (የቤቱግድግዳከምንእንደተሰራእራስሽበመመልከትምረጭ፤አንድመልስብቻይመረጥ)

1. Bricks/stone & cement (ከጡብ/ድንጋይናሲሚንቶ) 2. Bricks mud & cement (ከጡብ፣ከጭቃናሲሚንቶ)

3. Bricks & mud (ከጡብናከጭቃ) 4. Wood, mud & cement (ከእንጨት፣ከጭቃናሲሚንቶ)

5. Wood & mud (ከእንጨትናከጭቃ) 6. Wood only (ከእንጨትብቻ) 7. Other (specify) (ሌላ፤ይጠቀስ): __)

E32. What is the material of the floor made from? [Make your own observation & tick only one answer] (የቤቱወለልከምንእንደተሰራእራስሽበመመልከትምረጭ፤አንድመልስብቻይመረጥ)

1. Ceramic tiles (ከሴራሚክ) 2. Carpet (ከምንጣፍ) 3. Cement (with or without plastic carpet) (የሲሚንቶሊሾፕላስቲክምንጣፍያለዉ/የሌለዉ) 4. Wood (ከእንጨት) 5. Bricks (ከጡብ) 6. Mud with plastic carpet (ከአፈርየፕላስቲክምነጣፍያለዉ) 7. Mud (ከአፈር) 8. Other (specify) (ሌላ፤ይጠቀስ): ____)

→ Ask the mother to call for the household head and ask questions E33 to E35 to the household head.

→እናትየዉየቤተሰቡንኃላፊእንድትጠራልሽበማድረግከጥያቄ E33 እስከ E35 ያሉትንየቤተሰቡንኃላፊጠይቂ፡፡

E33. What is current the primary source of income for your household? [Tick only one answer]

በአሁኑግዜየቤተሰቡዋነኛየገቢምንጭምንድነው፤አንድመልስብቻይመረጥ፡፡

1	Farming of food crops (የምግብሰብሎችንበማምረት)	8	Handicrafts/artisan (በእደ-ጥበብስራዎች)
2	Farming of cash crops (like khat, coffee) (እንደቡናናጫትያሉየሽያጭሰብሎችንበማምረት)	9	Receiving money from relatives abroad (ከባህርማዶበሚላክገንዘብ)
3	livestock &/or bee keeping (በከብትርቢ/ንብበማነብ)	10	Supported by relatives in Ethiopia (ሀገርዉስጥካለዘመድከሚገኝድጎማ)
4	Employment/salary (በወርኃዊደሞዝ)	11	Other (specify) (ሌላ፤ይጠቀስ): ___)
5	Daily labour (በቀንስራ)	12	Refusal (አላውቅም/ፈቃደኛአይደሉም)
6	Small scale trade (like venders, small retailers, local brewery, selling of wood/charcoal, etc.) (በአነስተኛንግድ፤እንደጉልት፣ችርቻሮሱቅናጠላጠመቃበመሳሰሉ)		
7	Large scale trade (like wholesalers of agriculture produce) (ከፍባሉየንግድዓይነቶች፤ እንደአከፋፋይናየገበሬምርትተረክቦመሸጥበመሳሰሉ)		

E34. Do you have agricultural land of your own? የራስዎየሆነየግብርናመሬትአሎት

1. Yes (አዎን፤አለኝ) 0. No (አይ፤የለኝም) 9. Don't know or refusal (አላዉቅም/ፈቃደኛአይደሉም)

→ Skip question E35 if the answer to E34 is No or Don't know/refused to answer →ለጥያቄ E34 መልስየለምወይምአላዉቅም/ፈቃደኛአይደሉምከሆነ፤ጥያቄ E35ንዝለይ፡፡

E35. How much agricultural land do you own?

[Write the size of the land & tick the unit used; write '00' if no land is owned, '99' if don't know or refusal]

ያሎትየግብርናመሬትመጠንስንትነው Size of land (የመሬቱመጠን): |___|___|

Name of unit used (መሬቱየተለካበት): 1. Hectare (በሄክታር) 2. Fechasa/kert (በፈጫሳ/ቀርጥ) 3. Timad (በጥማድ) 4. Other (specify) (ሌላ፤ይጠቀስ): ________)

3. Hygiene Index Form

Background information: *[Confirm the child ID in the form with the mother card]*

ጠቅላላመረጃ፡በቅጹላይያለውንየህፃንመለያኮድከእናትየውካርድጋርበማመሳከርትክክለኛነቱንአረጋግጭ፡፡

<table>
<tr><td>H1. Ethiopian date of interview የቃለ-መጠይቁቀንእ.ኢ..አ</td><td>|___|___|/|___|___|/|___|___| (dd/mm/yy)
(ቀን/ወር/ዓ.ም.)</td></tr>
<tr><td>H2. Child ID የህጻኑመለያቁጥር</td><td>C |___|___|___|</td></tr>
<tr><td>H3. Name child የህጻኑስም</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
<tr><td>H4. Sex (1. Female / 2. Male) ጾታ (1. ወንድ/ 2. ሴት)</td><td>|___|</td></tr>
<tr><td>H5. Name Mother የእናትስም</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
<tr><td>H6. Name HH head የቤተሰቡኃላፊስም</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
<tr><td>H7. Name HEW የጤናኤክስቴንሽንሰራተኛስም</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
<tr><td>H8. House Number የቤትቁጥር</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
<tr><td>H9. Name Gere የገሬዉስም</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
<tr><td>H10. Name Zone የዞኑስም</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
<tr><td>H11. Name Kebele የቀበሌዉስም</td><td>|___|___|___|___|___|___|___|___|___|___|___|___|</td></tr>
</table>

Hygiene condition: *[Respondant should be mother of the index child]*

የንጽህናሁኔታ፡ (ሁሉንምጥያቄዎችእናትየዉንጠይቂ)

H12. When do you normally wash your hands? *[Tick all that apply]* አብዛኛዉንግዜእጅትንየመታጠቡትመቼነዉ፤ መልስየሆነዉሁሉይመረጥ፡፡

1	I don’t wash my hands at all (በማንኛዉምግዜእጄንአልታጠብም)	4	Before breastfeeding the child (ህፃኑንላጠባስል)	7	After cleaning child’s faeces (የህፃኑንአይነምድርካጸዳሁበኋላ)
2	After going to the toilet (ከሽንትቤትመልስ/ከተጸዳዳሁበኋላ)	5	Before meal (ከምግብበፊት)	8	When my hands are dirty (እጄሲቆሸሽ)
3	Before preparing food for the child (ለህፃኑምግብከማዘጋጀቴበፊት)	6	After meal (ከምግብበኋላ)	9	Don’t know/refusal (አላዉቅም/ፈቃደኛአይደሉም)

H13. What do you usually use for washing your hand? *[Tick only one answer]*
አብዛኛዉንግዜእጅትንየሚታጠቡተበምንድንነዉ፤አንድመልስብቻይመረጥ

1. Ash (በአመድ) **2.** Soap (በሳሙና) **3.** Only with water (በዉኃብቻ) **4.** Other (specify) (ሌላ፤ይጠቀስ):

H14. The last time when your child passed stools, how were the stools disposed? *[Tick only one answer]* ህፃኑለመጨረሻጊዜሲፀዳዳአይነምድሩየትተወገደ፤አንድመልስብቻይመረጥ

1	Put / rinsed into toilet or latrine (መጸዳጃቤትተደፋ)	**5**	Left in the open (ክፍትቦታላይተተወ)
2	Put / rinsed into drain or ditch (ፍሳሽማስወገጃመስመርውስጥተደፋ)	**6**	Other (specify) (ሌላ፤ይጠቀስ): ________)
3	Thrown in the garbage (ቆሻሻመጣያቦታተጣለ)	**9**	Don't know/refusal (አላውቅም/ፈቃደኛአይደሉም)
4	Buried (ጉድጓድውስጥተቀበረ)		

Hygiene spot-checks: *[Make your own observation & record in the boxes provided. Write '1' for yes, '0' for no, '8' for not applicable, and '9' if mother refuses observation].* **የሃይጅንስፖት-ቼክ፡**
(በራስሽበመመልከትእንደሚከተለውሙይ፡- *1*-አዎን፤አለ፣*0*-አይ፤የለም፣*8*-አይመለከተውም፣ 9-ለማሳየትፍቃደኛአይደሉም፡፡

	Drinking water የመጠጥዉኃ	**Observation true?** ታይቷል**?**
H15	Interior water storage container covered የቤትዉስጥየውኃማጠራቀሚያው·ተከድኗል	|___|
H16	Exterior water storage container is clean የቤትዉጭየውኃማጠራቀሚያውንጹህነው	|___|
H17	Water storage container contains water የውኃማጠራቀሚያውውኃይዟል	|___|

	Food ምግብ	**Observation true?** ታይቷል**?**
H18	Clean dishes are covered የታጠቡ/ንጹህሳህኖችተከድነዋል	|___|
H19	Clean dishes are kept high የታጠቡ/ንጹህሳህኖችከፍተደርገዉተቀምጠዋል	|___|
H20	All food is covered ሁሉምምግብተሸፍኗል	|___|
H21	Left-over food is present የተረፈምግብአለ	|___|

	Personal hygiene የግልንጽህና	**Observation true?** ታይቷል**?**
H22	Mother is wearing shoes እናትየውጫማአድርጋለች	|___|
H23	Mother's hands are clean የእናትየወእጆችንፁህናቸው	|___|
H24	Child's hands are clean የህፃኑእጆችንፁህናቸው	|___|
H25	Child uses diapers, underclothes ህፃኑየሽንትመቀበያጨርቅ (ዳይፐር) ተደርጎለታል	|___|
H26	Child's face is clean የህፃኑፊትንፁህነዉ	|___|
H27	Child's clothes are clean የህፃኑልብሶችንፁህናቸዉ	|___|
H28	Faeces on the ground አይነምድርመሬትላይአለ	|___|
H29	Child's playground is clean ህፃኑየሚጫወትበትቦታንፁህነዉ	|___|

	Household የቤትንጽህና	**Observation true? ታይቷል?**
H30	No trash outside house ከቤቱውጭቆሻሻየለም	|___|
H31	No trash inside house በቤቱውስጥቆሻሻየለም	|___|
H32	No unrestrained animal in patio or house በቤቱውስጥናዙሪያያልታሰረ/የተለቀቀእንስሳየለም	|___|
H33	No accumulation of dirty clothes የቆሸሹልብሶችአልተከማቹም	|___|
H34	Insignificant number of flies in house በቤቱውስጥእምብዛምዝንብአይታይም	|___|
H35	No standing water in patio or around house በቤቱውስጥናዙሪያያቆረውኃየለም	|___|

H36. Remarks (ልዩአስተያየት):

____

4. የህጻንሰዉነትልኬትመሙያቅጽ 1 Anthropometry Child Form CHILD ID: «cid»

A1. የህጻንመለያኮድ: **C«CID»A2.** የህጻንስም**:«NAMECH»A3.** ጾታ: **«SEX»A4.** የልደትቀንበኢትዮጵያአቆጣጠር: **«DOB»**(ቀን/ወር/ዓ.ም.)

A5. ምን:: **«SOURCE»A6.** እድሜበመጀመሪያልኬትጊዜ:**«AGEINCL»**ወር**A7.** የእናትስም: **«NAMEM»A8.** የቤተሰቡኃላፊስም:**«HH»**

A9. የጤናአክስቴንሽንሰራተኛስም: **«HEW»A10.** ገሬ: **«GERE»A11.** ዞን: **«ZONE»A12.** ቀበሌ: **«KEBELE»**

<table>
<tr><th rowspan="2"></th><th rowspan="2">ቀንበኢትዮጵያአቆጣጠር
(ቀን/ወር/ዓ.ም.)
Ethiopian date
(dd/mm/yy)</th><th rowspan="2">የጡንቻዙሪያ
(ሴ.ሜ.)
MUAC
(cm)</th><th rowspan="2">የጭንቅላትዙሪያ
(ሴ.ሜ.)
Head circum.
(cm)</th><th rowspan="2">ርዝመት
(ሴ.ሜ.)
Length
(cm)</th><th rowspan="2">ክብደት
(ኪ.ግ.)
Weight
(kg)</th><th rowspan="2">ክብደትለርዝመት
WLZ
1. < -3
2. = -3
3. > -3</th><th rowspan="2">የሁለቱምእግሮችእብጠት
Bilateral edema
1. አለ Present
0. የለም Absent</th><th rowspan="2">የቅጽቁጥር
(1 ወይም 2)
Data form
(1 or 2)</th><th colspan="2">የመረጃሰብሳቢቡድን
DC Team</th><th>ጥራትተቆጣጣሪ
DQM</th><th>መረጃአስገቢ
Data Clerk</th></tr>
<tr><th>ቁጥር
N°</th><th>ፊርማ
Sign</th><th>ፊርማ
Sign</th><th>ፊርማ
Sign</th></tr>
<tr><td>V1</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V2</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V3</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V4</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V5</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V6</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V7</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V8</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V9</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V10</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V11</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V12</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
<tr><td>V13</td><td>|__|__|/|__|__|/|__|__|</td><td>|__|__|.|__|</td><td>|__|__|.|__|</td><td>|__|__|__|.|__|</td><td>|__|__|.|__|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td>|__|</td><td></td><td></td><td></td></tr>
</table>

የቅጽመዝጊያEnd of form

A14. አስተያየትRemarks

____
____
____

A15. ያበቃበትቀንበኢትዮጵያአቆጣጠርEthiopian date of end of form:|___|___|/|___|___|/|___|___|(ቀን/ወር/ዓ.ም.)(dd/mm/yy)

A16. ያበቃበትምክንያትReason:|___|

1= በጥናቱመጀመሪያላይለመሳተፍፈቃደኛአልሆኑምdeclined to participate at enrolment

2= በጥናቱለመቀጠልፈቃደኛአይደሉምdeclined to continue in the study

3= ሁሉምጎብኝቶችተካሂደዋልall visits have been performed

4= የጥናቱንአካባቢለቀዉሄደዋልmoved out of study area

5= ህጻኑ/ኗከፍተኛየምግብእጥረትተገኝቶበታል/ተገኝቶባታልseverely acutely malnourished infant

6= የህጻንሞት፤የሞተበት/ችበትቀንበኢትዮጵያአቆጣጠርየተገመተ: |___|___|/|___|___|/|___|___| (ቀን/ወር/ዓ.ም.)infant death (estimated Ethiopian date of death)(dd/mm/yy)

7= የእናትሞት፤የሞተችበትቀንበኢትዮጵያአቆጣጠርየተገመተ: |___|___|/|___|___|/|___|___| (ቀን/ወር/ዓ.ም.)mother death (estimated Ethiopian date of death) (dd/mm/yy)

8= በህጻኑላይበተከሰተከፍተኛየጎንዮሽጉዳት (ይገለጽ) serious adverse event in infant (please specify): ……………………………………………………………..

9= በእናትየዉላይበተከሰተከፍተኛየጎንዮሽጉዳት (ይገለጽ) serious adverse event in mother (please specify): ……………………………………………………………

ህጻኑ/ኗከፍተኛየምግብእጥረትከተገኘበት/ባትለህክምናየመላኪያ(ሪፈራል) ቅጽተሞልቶባቅራቢያወደሚገኘዉጤናጣቢያ/ሆስፒታልይላክ/ትላክ::

In case infant is severely acutely malnourished, please refer him/her to the nearby health center/hospital with a completed referral slip.

የተመራማሪዉስምናፊርማInitials and Signature Researcher ………………………………………………..

5. Dhibee: Morbidity Form CHILD ID: C«ID»

M1. Koodii eenyummaa daa'imma:**C001M2.**Maqaa daa'imma:**LM3. Saala: M4.** Guyyaa (ALE):**(gg/jj/ww)** **M5.**Madda:**M6.**Umuri yeroo dawanna jalqabaa:**12,1ji'aM7.** Maqaa Haadha: **M8.** Maqaa abba waraa**: M9.** Maqaa hojjeettuu eksteenshinii fayyaa:**M10.**Maqaa gare: **LALIFTUM11.** Maqaa zoonii**: M12.** Maqaa gandaa**: AM14.** Tartiba lakk uunka : **01**

Haala neegeenya fayyaa daa'imaa torban darbee erga daawannaa hojjeetaa/ttu hawaasaa isa dhumaa ilaalee asi/booda Health condition of child in last week (since last visit of CM)

Haatii YKN daa'imma mana yoo hinjiree deddeebi'ii illaali. Yoo daa'imini jiraate gaaffii kanaa haadha wajjin raawadhu. Yoo haadha torban tokkoof marmaartee dhabde gaggeessaa qoranichaaf gabaasi. Return to HH if child and mother are not present. Perform interview only with mother and if child is present. Inform researcher if mother could not be reached for 1 full week.

	Guyyaa akka lakk Itoophiyaati (gg/jj/ww) Ethiopian date (dd/mm/yy)	Yaali[a] Treated	Olguruu (guyyaa) Vomit (days)	Garaa Kaasaa[b] (guyyaa) Diarrhea (days)	Qufaa (guyyaa) Cough (days)	Furri[c] Runny nose	Afuura baasu rakkachuu[d] Difficult breathing	Lakk.Hargan suu (lakkofsaan) Breathing rate (No°)	Gubaa qaamaa (°C) Body temperature (°C)	Busaa[e] (koodii) Malaria (code)	Yaada (fkn. yaalii kenname) Remarks (e.g. treatment)	Hojetaata hawwaasa Community Member		Suppervisarii Supervisor
												Lakk No°	Mallattoo Sign	Mallattoo Sign
V1	|__|__|/|__|__|/|__|__|	|__| |__|	|__|	|__|	|__|	|__|	|__|	|__|__|	|__|__|.|__|	|__|		|__|__|		
V2	|__|__|/|__|__|/|__|__|	|__| |__|	|__|	|__|	|__|	|__|	|__|	|__|__|	|__|__|.|__|	|__|		|__|__|		
V3	|__|__|/|__|__|/|__|__|	|__| |__|	|__|	|__|	|__|	|__|	|__|	|__|__|	|__|__|.|__|	|__|		|__|__|		
V4	|__|__|/|__|__|/|__|__|	|__| |__|	|__|	|__|	|__|	|__|	|__|	|__|__|	|__|__|.|__|	|__|		|__|__|		

[a] Deebiin hedduu laatamuu danda'a: **0.** hinwal'aanamnee no treatment / **1.** kellaa fayyaatii yaalame/te HP / **2.** buufata fayyaatii yaalame/te HC / **3.** Hoospitaallatii yaalame/te hospital / **4**. Qoorichaa faarmaasii irraa fayyadamte (<u>maqaa qorichaa fi maalif akka fayyadamte yaada keessatti katabi</u>) pharmacy / **5**. Qoricha aadaa fayyadamte (<u>maqaa qorichaa fi maalif akka fayyadamte yaada keessatti katabi</u>) traditional

[b] Garaa kaasaa jeechuun yooo albaatiin guyyaatti si'a sadii fi isa ol teessiseedha.

[c] **1.** Eyyeen / **0**. Lakki / **9**. Yoo ilaalchisuu diddee

[d] **0**. hin qabu / **1**. yoo daa'imni hargansiisu kan kuruufsisu (<u>gara buufata fayaatti ergi</u>) / **2**. yoo daa'imni hargansiisu kan kuruufsisu fi isa wajjiin wal qabatee sochii ribuu lapheerraa mul'atu (<u>gara buufata fayaatti ergi</u>) / **9**. Ni didde

[e] Yoo gubaan qaama mucaa 37.5 °C fi isaa ol ta'e YKN haatii layidaa/ qaama gubaa sa'aa 24 keessatti mudachuu isaa si himte Qoorannoo dhiigaa busaa raawadhu

➡ **Yeroo daawwannaa 4[ffaa], haadha mucaa akka ishiin nyaatni dabalataa akka rabsamuuf/ kennamuuf akkasumas akka kaardii mucaa ishee fiddee iddoo qorannoo akka dhuftu yaadachiisi**

Haadha ykn daa'ima ishii waraqaa referii wajjin ergi refer child/mother with referral slip:

- yoo daa'imni olguura ykn garaa kaasaa qaba ta'e gara (hojjettuu eksteenshinii fayyaatti) child has diarrhea or is vomiting (HEW)
- yoo yeroo hargansiisuu kurufsisa ta'e ykn sochiin ribuu laphee ni mul'ata ta'e gara buufata fayaatti child grunts or has chest indrawing (health center)
- yoo daa' imni ishii busaa qaba ta'e gara eksteenshinii fayyaatti test code 1 or 2 child tested positive on malaria, test code 1 or 2 (HEW)
- yoo haati mucaa dhibee addaa kan jabaa hintaane dheerte gara eksteenshinii fayyaatti child or mother reported other or not serious health concern(HEW)

Yoo dhibeen cimaan YKN duuti daa'ima mudate battalumatti Dr. Mekitie Wondafrash (047-1119299)
YKN obbo Alemayehu Argaw (09-33145853) tiif gabaasi.

Xumuraa uunkaa-End of form

M14. Guyyaa uunkaan itti dhume akka lakk Itiyoophiyaatii: |__|__|/|__|__|/|__|__|(gg/jj/ww)Ethiopian date of end of form (dd/mm/yy)

M15. Sababaa Reason: |__|

1= Qorannoo irratti guyyaa duraa hirmaachuu diduu declined to participate at enrolment

2= qo'annoo keessatti hirmaachuuf itti fufuu diduu (sababa isaa ibsi____) declined to continue in the study

3= daaw'anna hunda raawatameeraall visits have been performed

4= naannoo qo'annaa lakkisaniru moved out of study area

5= du'aati daa'imma (Guyyaa akka lakk Itiyophiyatii: |__|__|/|__|__|/|__|__| (gg/jj/ww))infant death andestimated Ethiopian DOD (dd/mm/yy)

6= du'aati haadhaa (Guyyaa akka lakk Itiyophiyatii: |__|__|/|__|__|/|__|__| (gg/jj/ww))mother death and estimated Ethiopian DOD (dd/mm/yy)

Qubee jalqabaa fi mallattoo gaggeessaa qo' annichaa Initials and Signature Researcher…………..……………………………………………………...

Mallattoo ragaa kan galchitu Signature Data Entry Clerk

6. Unka Qajeelfama Compliance Form

CHILD ID : «CID»

M1. Koodii eenyummaa daa'imma:**C«CID»M2.**Maqaa daa'imma:**«NAMECH»M3. Saala: «SEX»M4.** Guyyaa dhalootaa akka lakk Itoophiyaati:**«DOB»(gg/jj/ww)**

M5. Madda: **«SOURCE»M6.**Umuri yeroo dawanna jalqabaa:**«AGEINCL»ji'aM7.** Maqaa Haadha (Mother)**«NAMEM»**

M8. Maqaa abba waraa**: «HH»M9.** Maqaa hojjeettuu eksteenshinii fayyaa: **«HEW»M10.**Maqaa gare: **«GERE»**

M11. Maqaa zoonii**: «ZONE»M12.** Maqaa gandaa**: «KEBELE»M14.** Tartiba lakk uunka(Sequence number) |__|__|

	Guyyaa akka lakk Itoophiyaati (gg/jj/ww) Ethiopian date (dd/mm/yy)	**Guyyaa darbee keessa si'a meqaa harma hoosifte*** BF frequency **0.** Hin hoosifnee (No) **1.** Guyyaatti si'a 1-3(1-3/d) **2.** Guyyaatti si'a 4-6(4-6/d) **3.** Guyyaatti si'a 7-9(7-9/d) **4.** Guyyaatti si'a ≥ 10 (≥10/d) **9.** Hin Bekkuu (Don't know)	**Nyaata dabalata kennamee ala kan biraa yoo da'imichi/ttiin nyaate/tte** **1.** Eyyeen Yes **0.** Lakki No **9.** hinbeeku/deebii kennuu didde	**Kiniina haafee** (lakk YKN **99**. Yoo Diddee) Remaining Capsules (nr or 99. Refusal)	**Nyaataa dabalataa hafee** (lakkoofsa saakeetti nyaata irraa jiru irra marsii) Remaining Food (draw circle around appropriate number)	**Yaada** Remarks	**Hojjettoota hawwaasa community worker**		**Maqaa nama to'attuu supervisor**
							Lakk No°	**Mallattoo Sign**	**Mallattoo Sign**
V1	|__|__|/|__|__|/|__|__|	|__|	|__|	|__|__|	saakkeetti 1 bag 1 1. 2. 3. 4. 5. 9.Diddee saakkeetti 2 bag 2 1. 2. 3. 4. 5. 9.Diddee		|__|__|		
V2	|__|__|/|__|__|/|__|__|	|__|	|__|	|__|__|	saakkeetti 1 bag 1 1. 2. 3. 4. 5. 9.Diddee saakkeetti 2 bag 2 1. 2. 3. 4. 5. 9.Diddee		|__|__|		
V3	|__|__|/|__|__|/|__|__|	|__|	|__|	|__|__|	saakkeetti 1 bag 1 1. 2. 3. 4. 5. 9.Diddee saakkeetti 2 bag 2 1. 2. 3. 4. 5. 9.Diddee		|__|__|		
V4	|__|__|/|__|__|/|__|__|	|__|	|__|	|__|__|	saakkeetti 1 bag 1 1. 2. 3. 4. 5. 9.Diddee saakkeetti 2 bag 2 1. 2. 3. 4. 5. 9.Diddee		|__|__|		

Yoo haatii daa'imaa torbee tokko keessatti deddeebi'uun dhabamtee qorattoota kanaafi gabaasi. Inform researcher if mother could not be reached for 1 full week.

6. Unka Qajeelfama Compliance Form

CHILD ID : «CID»

Nyaatiicha **daa'immaa waggaa 2 gaddii qofaaf akka ta'ee haadha yaadachiisi**. Albuudoota nyaata dabalataa kana keessa jirru daa'imashee akka qaamaa fi sammuun guddatu fi gabbatu gargaara. Yoo nyaatichaa nama birraatiif kenname garuu, daa'imni ishee faayyidaa nyaatiichaa irra hinargatu.qaamaan guddachuu dhiisuufi gabbachuu dadhaba.

Please remind the mother that the food supplement is **only for children under 2 years of age**. The ingredients in the food supplement will help her child grow and develop healthily.

If she gives it to other people, her child will not grow and develop healthily.

Qubee jalqabaa fi mallattoo gageessaa qorannichaa Initials and Signature Researcher……………………………………………...

Mallattoo ragaa kan galchitu Signature Data Entry Clerk………

7. ሂሞግሎቢን፣ሲ-ሪያክቲቭፕሮቲንናየሆድትላትልማስወገጃቅፅHb – CRP – Deworming Form CHILD ID___

D1. የህጻንመለያኮድ: **C______________D2.** የህጻንስም:**D3.** ጾታ:**D4.** የልደትቀን(እ.ኢ..አ): (ቀን/ወር/ዓ.ም.)**D5.** ምንጭ:: **D6.** እድሜበመጀመሪያልኬትጊዜ:**12.4**ወር**D7.** የእናትስም: **D8.** የቤተሰቡኃላፊስም:

D9. የጤናአክስቴንሽንሰራተኛስም: **D10.** ገሬ: **D11.** ዞን: **12.4D12.** ቀበሌ:

	ቀንበኢትዮጵያአቆጣጠር (ቀን/ወር/ዓ.ም.) **Ethiopian date** (dd/mm/yy)	ሲ.ሪ.ፒ* **CRP** (mg/l)	ሄሞግሎቢን (ግ/ሊ) **Hemoglobin** (g/l)	ልዩአስተያየት **Remarks**	ነርስ **Nurse**		ጥራትተቆጣጣሪ **Quality Supervisor**	መረጃአስገቢ **Data Clerk**
					ቁጥር **N°**	ፊርማ **Sign**	ፊርማ **Sign**	ፊርማ **Sign**
D14. በጥናቱመጀመሪያ Baseline	|__|__|/|__|__|/|__|__|	|__|__|__|	|__|__|__|		|___|			
D15. በጥናቱአጋማሽ Mid-term	|__|__|/|__|__|/|__|__|	|__|__|__|	|__|__|__|		|___|			
D16. በጥናቱመጨረሻ Endline	|__|__|/|__|__|/|__|__|	|__|__|__|	|__|__|__|		|___|			

መለኪያዉከ 5 ሚ.ግ./ሊ. በታችካሳየ፤እንደሚከተለዉይመዝገብ: |__|__|__| (If “<5 mg/l” is displayed on device, record)

	ቀንበኢትዮጵያአቆጣጠር (ቀን/ወር/ዓ.ም.) **Ethiopian date** (dd/mm/yy)	የተሰጠሜቤንዳዞልመጠን(ሚ.ሊ.) **Given dose of Mebendazole** (ml)	ልዩአስተያየት **Remarks**	ነርስ **Nurse**		ጥራትተቆጣጣሪ **Quality Supervisor**	መረጃአስገቢ **Data Clerk**
				ቁጥር **N°**	ፊርማ **Sign**	ፊርማ **Sign**	ፊርማ **Sign**
D17. 12 ወርእድሜላይ Age 12 months	|__|__|/|__|__|/|__|__|	|__|__|__|		|__|			
D18. 18 ወርእድሜላይ Age 18 months	|__|__|/|__|__|/|__|__|	|__|__|__|		|__|			

እድሜያቸዉከ12 ወርበታችለሆኑህጻናትአይሰጥም! Not for infants less than 12 months old !

Summary

This PhD work was carried out to address feeding behavior of mother and caregivers, the role of infant and young child feeding practice indicators such as dietary diversity score in predicting complementary food quality and their association with growth in two distinct agricultural seasons in Ethiopia. Moreover, the role of n-3 LC-PUFA in child growth, morbidity and systemic inflammation was evaluated. **Chapter 1** introduces about the problem of undernutrition and feeding behavior and practices and the role of essential fatty acids in the diet of infants and young children globally and in the Ethiopian context, while **chapter 2** describes the study aims and study setting.

Malnutrition remains one of the leading public health problems globally. The high level of undernutrition in East, Central and Western African region more than any region in the world contributes to increased risk of mortality, disease burden and increasing trend of non-communicable diseases such as overweight and obesity, diabetes mellitus etc. Ethiopia is one of the countries in African region having one of the highest rates of stunting, wasting, and underweight among children under the age of five years. The absolute numbers children with undernutrition are raising due to an increase in population growth and slow drops in the prevalence of stunting. According to the famous UNICEF conceptual framework, it is known that determinants of undernutrition/malnutrition operate at different levels and are intricate.

The “first 1000 days”, the time from pregnancy and the first two years of life, is considered the most sensitive period in terms of vulnerability and the potential for the maximum intervention effect. Both caregiver-child interactions during feeding and the practice of different modes of feeding are crucial for optimal child physical growth and development. Appropriate caregiver-child interaction during feeding is widely recommended for the different feeding modes to have an effect on child growth or development; however, there are few studies showing the link. Moreover, provision of appropriate and adequate complementary food has been a challenge in most developing regions and interventions targeting complementary feeding period had no or little effect on child growth, development or morbidity. The diets in these settings lack diversity and are composed mainly of cereals, roots and tubers with cow milk used as a main animal based source of protein and micronutrients. The addition of fats both as energy sources or sources of essential fatty acidshavebeen advocated in the last a decade or so; but the impact of additional supplementation with n-3 fatty acids is not known in developing country setting and the available evidence from the West is equivocal.

Chapter 3 presents the analysis of type of caregiver-child interaction in southern Ethiopia. A total of 764 caregivers provided complete data on feeding styles out of a sample of 826 using a questionnaire adapted from a Caregiver Feeding Style Questionnaire (CFSQ). Multivariate multinomial logistic regression was employed to identify predictors of caregivers' feeding styles. The majority of the caregivers practiced responsive feeding style. Caregivers other than the biological mother favoured a laissez-faire feeding style, while caregivers residing in rural *kebeles (*smallest administrative unit*)* were more responsive. Caregivers who were breast feeding relatively frequently were less responsive. The feeding style questionnaire utilized in this study needs to be validated in the local context and a prospective design with direct observation of caregiver-infant interaction should be used in order to ascertain causal relationship between caregiver feeding behaviour (style) and various child outcomes.

Even if breastfeeding is practiced widely in Ethiopia, early initiation and exclusive breastfeeding for the first six months remains to be significantly improved and practices of timely introduction of adequate, safe and appropriate complementary foods are very low. Moreover, there is a need to have a simple, inexpensive and valid tool to assess of intake of nutrients among children. Chapter 4 presents the analysis of the relationship between simple food group indicator (dietary diversity score, DDS) and dietary quality assessed using the mean of micronutrient density adequacy (MMDA). An interactive 24-hr dietary recall assessment was used to determine nutrient intake from a total of 320 and 312 infants in harvest (HS) and preharvest seasons (PHS) in the catchment of the Gilgel Gibe Field Research Centre of Jimma University, Southwest Ethiopia and a separate intake of food groups was carried out to calculate dietary diversity score. MMDA was calculated from eight nutrients. The validity of DDS to predict nutrient density adequacy was determined using receiver operating characteristic analysis using "low" MMDA (<50%) and "higher" MMDA (≥75%) as cutoffs. We found that DDS was associated with MMDA after controlling for confounding variables in multiple linear regression. Moreover, a DDS of ≤2 food groups best predicted "low" MMDA(<50%) with 84% and 92% sensitivity, 36% and 43% specificity and 47% and 51% correct classification for the HS and PHS respectively. DDS was not able predict "higher" MMDA as very few study children attained MMDA of ≥75%. The findings support the use of DDS to indicate inadequate intake of micronutrients by breastfed infants in different seasons.

Indices of infant and child feeding have been used to predict growth by various authors, however, the results are inconsistent and the effect of agricultural seasons on such relationship is not known. **Chapter 5** presents the evaluation of the association of DDS and composite index of infant and child feeding (infant and child feeding index, ICFI) with growth among infants and young children in repeated cross-sectional and follow up surveys in two distinct agricultural seasons in rural Ethiopia (HS and PHS). A sample of 6-12 month old childrenduring HS (n=320) and PHS (n=312) were taken. In addition, 6-12 month old children from the HS were reassessed 6 months later during PHS. Data on feeding practices and anthropometry were collected in the two seasons. To increase the power, pooled data of 6-12 children from both seasons was analyzed. The mean (±SD) length-for-age z-score (LAZ) of the 6-12 month old children was -0.77(±1.4) and -1.0 (±1.3) in HS and PHS, respectively; while the mean (±SD) of the follow-up children in PHS was -1.0 (±1.3). The median DDS (IQR) was not different between the two seasons for the 6-12 months old children and obviously the follow up children had a slightly higher median of DDS. The DDS in HS was positively associated with LAZ at follow-up (β= 0.16; 95%CI: 0.01, 0.30; *P*=0.03); however, ICFI and DDS were not associated with mean LAZ, weight-for-height z-score and weight-for-age z-score within season. The odds of being stunted was higher for those children with low DDS (DDS≤2) in HS and in the pooled data from the two seasons. Therefore, it was concluded that DDS was a good indicator of stunting during Ethiopian HS and it can be used as a tool to evaluate the association of feeding practice and child growth irrespective of season. However, inclusion of additional dimensions and the use of longitudinal ICFI should be used for future evaluation of the association of this composite index with growth in different seasons.

It was previously reported in Africa that early exposure to unhygienic environment was implicated in the onset of enteric inflammation or enteropathy starting from early age and significantly contributed to linear growth faltering which starts in the first few months of life. **Chapter 6** presents the results of an intervention looking at the effect a long-chain omega-3 polyunsaturated fatty-acids (n-3 LC-PUFAs) supplementation to breast feeding mothers and their infants on morbidity, systemic inflammation and growth of children 6-24 months in rural southwest Ethiopia. A four-arm double-blind randomized controlled trial was carried out by including a total of 360 mother-infant pairs. Infants were 6-12 months old at enrollment. Study arms were both the lactating mother and child receiving the fish oil intervention (MCI); only the lactating mother receiving intervention (MI); only the child receiving intervention (CI) and both mother and child receiving a placebo supplement or control (C). Subjects were followed up for a period of about 12 months. The maternal intervention was provided in the form of air tight gel capsules contained either fish oil (intervention) or corn oil (placebo). The intervention capsules contained 500 mg n-3 LC-PUFAs (215 mg docosahexaenoic acid-DHA + 285 mg eicosapentaenoic acid-EPA). Whereas, the child intervention food supplement was enriched with microencapsulated fish-oil providing a daily dose of 500 mg n-3 LC-PUFAs (169 mg DHA + 331 mg EPA). The intervention supplement was packaged in such a way to prevent oxidation. Multiple micronutrients were added to the food supplement for all arms in the food supplement. The primary study outcome was linear growth assessed by monthly changes in length-for-age z-score. Anthropometric measurements were taken monthly, and hemoglobin and C-reactive protein were measured at baseline and after 6 and 12 months of follow-up. Blood spot samples were collected in the field from a sub-sample of 164 children to analyze for whole blood DHA, EPA, and arachidonic acid (AA) concentrations. Moreover, weekly morbidity surveillance was conducted throughout the study. It was demonstrated that fish-oil supplementation resulted in significant increases in blood n-3 LC-PUFA concentration (P<0.01) and decreases in AA/DHA+EPA ratio (P<0.001) in all intervention arms. No significant intervention effect was found on linear growth, morbidity or systemic inflammation. Compared to the control group, significantly larger monthly changes in weight-for-length z-scores was found in the CI arm [effect size (95% CI): 0.026 (0.004, 0.048) per mo] and the MCI arm [(effect size (95% CI): 0.022 (0.000, 0.043) per mo]. In conclusion, the result of the trial demonstrated that n-3 LC-PUFA supplementation of lactating women and children did not affect linear growth and morbidity of children in a low-income setting. The n-3 LC-PUFA supplementation given directly to children increased relative weight gain but the quality of weight gain has not been verified with body composition measurement. All in all the findings support the existing evidence of inconsistent or lack of effect of n-3 LC-PUFA supplementation on linear growth, morbidity and inflammation.

In sum, this Ph.D. work provided evidence regarding the level of caregiver feeding behavior, the utility of simple food group based indicator of infant and child feeding as an important tool in the evaluation of dietary quality and growth irrespective of agricultural seasons. However, constructing a composite infant and child feeding index using additional components and the use of prospective design was recommended. The findings from our n-3 LC-PUFA intervention study provided comparable evidence of lack effect on linear growth and morbidity like another study in developing country setting and further trial using a larger dose and early initiation of n-3 LC-PUFA supplementation has been recommended for researchers. A child development outcome measured with specific development test battery is also recommended to generate evidence on the effect of n-3 LC-PUFA supplementation on child development in this setting.

Samenvatting

Dit doctoraatswerk werd uitgevoerd om het voedingsgedrag van moeder en zorgverleners te verbeteren. Hierbij werd het belang van voedingspraktijkindicatoren voor baby's en jonge kinderen bekeken. Er werd nagegaan hoe de diversiteit in het dieet zowel kwaliteit van het dieet als groei kon voorspellen in twee verschillende landbouwseizoenen in Ethiopië.

Daarnaast werd de rol van n-3 LC-PUFA bij kindergroei, morbiditeit en systemische inflammatie geëvalueerd. **Hoofdstuk 1** beschrijft het probleem van ondervoeding en voedingsgedrag en de rol van essentiële vetzuren in de voeding van zuigelingen en jonge kinderen wereldwijd en in de Ethiopische context, terwijl **Hoofdstuk 2** de studiedoelstellingen en de onderzoeksopzet beschrijft.

Wereldwijd blijft ondervoeding een van de grootste bedreigingen voor de volksgezondheid. De hoge mate van ondervoeding in Oost-, Centraal- en West-Afrikaanse regio's draagt nergens ter wereld meer bij tot een verhoogd risico op sterfte, ziektelast en een toename van niet-overdraagbare ziektes zoals overgewicht, obesitas, diabetes mellitus, etc. Ethiopië is een van de landen met een hoog percentage van groeiachterstand en ondergewicht bij kinderen onder de vijf jaar. In absolute cijfers neemt het aantal kinderen met ondervoeding toe. Dit is het gevolg van een combinatie van zowel bevolkingsgroei en een langzame afname van de prevalentie van groeiachterstand. Volgens het UNICEF zijn de determinanten van ondervoeding een complex gegeven en is hun werking aanwezig op verschillende niveaus.

De "eerste 1000 dagen", dit is de tijd vanaf de zwangerschap en de eerste twee levensjaren, is meest gevoelige periode. De kwetsbaarheid van kinderen is er het hoogst en het potentieel voor interventies is maximaal. Zowel de interactie tussen zorgverlener en kind tijdens het voeden, als het gebruiken van verschillende voedingsmethoden, zijn essentieel voor de optimale groei en ontwikkeling van het kind. Een voldoende mate van interactie tussen zorgverlener en kind tijdens het voeden, wordt sterk aanbevolen, opdat de verschillende voedingswijzen een effect kunnen hebben op de groei en ontwikkeling van het kind. Er zijn echter weinig studies die deze link aantonen. Beschikken over voldoende en goede complementaire voeding is voor de meeste ontwikkelingsgebieden een hele uitdaging. Interventies die zich richten op een aanvulling van voeding hebben tot nu toe tot weinig effect gehad op de groei, ontwikkeling en ziektecijfers van kinderen. De meeste diëten bestaan voornamelijk uit granen, wortels en knollen met koeienmelk als een voornaamste dierlijke bron van eiwitten en micronutriënten. In het laatste decennium werd er gepleit voor de toevoeging van vetten die zowel als energiebron en bron van essentiële vetzuren dienen. Over de impact van supplementen met n-3 vetzuren is in ontwikkelingsgebieden weinig geweten en ook in het Westen zijn beschikbare gegevens twijfelachtig of onduidelijk.

In **hoofdstuk 3** wordt een analyse van de interactie tussen zorgverlener en kind in Zuid-Ethiopië beschreven. Met behulp van een aangepaste vragenlijst werd de voedingsstijl gemeten bij een steekproef van 826 zorgverleners. Er werd gebruik gemaakt van multivariate multinomiale logistische regressie om de voorspellende factoren van de voedingsstijl van de zorgverleners te identificeren. De meerderheid van de zorgverleners maakten gebruik van een responsieve voedingsstijl. Zorgverleners die niet de biologische moeder waren, gaven de voorkeur aan een laissez-faire voedingsstijl, terwijl zorgverleners uit rurale dorpenvoor een eerder responsieve aanpak opteerden. Zorgverleners die relatief vaak borstvoeding gaven, waren minder responsief. De vragenlijst over voedingsstijlen die in dit onderzoek wordt gebruikt, moet worden verder gevalideerd in de lokale context en een prospectieve studie met directe observatie van de interactie tussen zorgverlener en kind. Op die manier kan het oorzakelijk verband gemeten worden tussen het voedingsgedrag of stijl van de zorgverlener en de effecten voor het kind.

Ook al wordt het geven van borstvoeding op grote schaal toegepast in Ethiopië, moet de vroege start en het uitsluitend geven van borstvoeding in de eerste zes maanden nog aanzienlijk worden verbeterd. Bovendien komen praktijken, die op het juiste tijdstip voldoende, veilige en geschikte complementaire voedingsmiddelen introduceren, amper voor. Daarenboven is er nood aan een eenvoudig, goedkoop en gevalideerd instrument dat de inname van voedingsstoffen bij kinderen kan beoordelen.

Hoofdstuk 4 behandelt de analyse van de relatie tussen de eenvoudige voedselgroep indicator (DDS: Dietary Diversity Score) en de kwaliteit van voeding met behulp van de micronutrient density adequacy (MMDA). Om de inname van nutriënten te bepalen werd gebruik gemaakt van een interactieve 24-uurs bevraging van het dieet bij 320 kinderen in oogst seizoen (OS) en 312 kinderen in pre-oogst seizoen (POS) in het stroomgebied van Gilgel Gibe van het onderzoekscentrum van Jimma University in het Zuidoosten van Ethiopië.

De inname van voedselgroepen werd gemeten om een diversiteitsscore van het dieet te berekenen. De MMDA van 8 verschillende nutriënten werd berekend. De validiteit van de DDS in het voorspellen van de 'nutrient density adequacy' werd nagegaan door middel van receiver operating characteristic analysis met "lagere" MMDA (<50%) en "hogere" MMDA (≥75%) cutoffs. De resultaten toonden een verband aan tussen DDS en MMDA na de controle voor randfactoren in meervoudige lineaire regressie. Een DDS van ≤2 voedselgroepen voorspelde het best een "lagere" MMDA (<50%) met een sensitiviteit van 84% en 92%, een specificiteit van 36% en 43% en een correcte classificatie van 47% en 51% voor respectievelijk de OS en POS. DDS was geen goede voorspeller voor een "hogere" MMDA doordat weinig kinderen binnen de studie een MMDA van ≥75% haalden. De bevindingen bevestigen het gebruik van DDS om aan te geven dat in verschillende seizoenen micronutriënten onvoldoende worden opgenomen door zuigelingen die borstvoeding krijgen.
Verschillende auteurs gebruikten indices betreffende de voeding van baby's en kinderen. De resultaten zijn inconsistent en het effect van landbouwseizoenen op dergelijke relatie is niet bekend.

Hoofdstuk 5 presenteert een evaluatie van het verband tussen DDS en de samengestelde index van de voeding van baby's en kinderen (ICFI, infant and child feeding index) en groei bij zuigelingen en jonge kinderen. Hiervoor werd een herhaalde cross-sectionele en follow-up enquête in twee verschillende landbouwseizoenen op het platteland van Ethiopië (OS en POS) opgezet. Er werd een steekproef genomen van kinderen tussen 6 en 12 maanden oud tijdens OS (n=320) en POS (n=312). Bijkomend werden kinderen tussen de 6 en 12 maanden oud van het OS 6 maanden later opnieuw onderzocht tijdens het POS. De data omtrent voedingspraktijken en antropometrie werd verzameld gedurende de twee seizoenen. Om de sterkte van de studie te maximaliseren werd de gepoolde data van de kinderen tussen 6 en 12 maanden uit beide seizoenen geanalyseerd. De gemiddelde (±SD) lengte-voor-leeftijd z-score (LLZ) voor de 6 tot 12 maanden oude kinderen was 0.77(±1.4) en -1.0 (±1.3) in respectievelijk OS en POS. Het gemiddelde (±SD) van de follow-up kinderen in POS was -1.0 (±1.3). De mediaan van

DDS (IQR) verschilde niet tussen de twee seizoenen voor de 6 tot 12 maanden oude kinderen. Zoals verwacht had de groep follow-up kinderen een iets hogere mediaan van DDS. De DDS in OS kent een positief verband met LLZ binnen de follow-up groep 0.16; 95%CI: 0.01, 0.30; *P*=0.03). De ICFI en DDS hadden echter geen verband met het gemiddelde LLZ, de gewicht-voor-lengte z-score en de gewicht-voor-leeftijd z-score binnen het seizoen. De kinderen met een lage DDS (DDS≤2) in OS en de kinderen uit de gepoolde data hadden een hogere kans op groeiachterstand. Om die reden werd geconcludeerd dat DDS een goede indicator is voor groeiachterstand tijdens het Ethiopisch OS en kan het gebruikt worden om het verband tussen voedingsprakijken en de groei van het kind te evalueren, ongeacht het seizoen. Toekomstige evaluatie van het verband van deze samengestelde index met de groei in verschillende seizoenen moet echter extra dimensies opnemen en gebruik maken van longitudinaal ICFI.

Eerdere onderzoeken in Afrika toonden aan dat een vroegtijdige blootstelling aan een onhygiënische omgeving betrokken was bij het ontstaan van enterische ontsteking of enteropathie vanaf jonge leeftijd, alsook sterk bijdroeg aan het stokken van de lineaire groei gedurende de eerste levensmaanden.

Hoofdstuk 6 behandelt de resultaten van een interventie die keek naar het effect van een 'long-chain omega-3 polyonverzadigd vetzuur (n-3 LC-PUFAs) supplement voor moeders die borstvoeding geven en hun kinderen op morbiditeit, systematische inflammatie en de groei van kinderen tussen 6 en 24 maanden oud in ruraal zuidwest Ethiopië. Een dubbelblinde gerandomiseerde en gecontroleerde studie met 4 groepen werd uitgevoerd bij 360 moeder-kind paren. De kinderen waren tussen 6 en 12 maanden oud bij de aanvang van de studie. In de eerste interventiegroep ontvingen zowel de lacterende moeder als het kind visolie (MKI). In de tweede interventiegroep ontving alleen de moeder het middel (MI). De derde groep kreeg enkel het kind de visolie (KI) en in de vierde groep, de controlegroep, kregen zowel de moeder als het kind een placebo toegediend (C). De proefpersonen werden gedurende 12 maanden gevolgd. De maternale interventie bestond uit luchtdichte gelcapsules die ofwel visolie (interventiegroep) ofwel maïsolie (controlegroep) bevatten. De interventiecapsules bevatten 500 mg n-3 LC-PUFAs (215 mg docosahexaenoic acid-DHA + 285 mg eicosapentaenoic acid-EPA). Bij de kinderinterventie daarentegen, waren de voedingssupplementen verrijkt met micro ingekapselde visolie die een dagelijkse dosis van 500 mg n-3 LC-PUFAs (169 mg DHA + 331 mg EPA) voorzag. Verschillende micronutriënten werden toegevoegd aan de voedingssupplementen van zowel de interventiegroepen als de controlegroepen en om oxidatie te voorkomen. Het primaire onderzoekresultaat was een lineaire groei, die werd beoordeeld aan de hand van maandelijkse veranderingen in de lengte-voor-leeftijd-z-score. Maandelijks werden antropometrische metingen uitgevoerd en het hemoglobine en C-reactief proteïne niveau werden gemeten bij aanvang en na 6 en 12 maanden bij een follow-up. Daarnaast werden bij een sub-steekproef van 164 kinderen bloedvlekstalen afgenomen in het veld om te analyseren op DHA, EPA en arachidic acid (AA) concentraties in het bloed. Bovendien werd doorheen de gehele studie de morbiditeit geobserveerd. Het onderzoek toonde aan dat de visolie supplementen resulteerden in een significante toename in bloed n-3 LC-PUFA-concentraties ($P<0.01$) en afnames in AA/DHA+EPA-ratio ($P<0.001$) in alle onderzochte groepen. Er werd geen significant interventie-effect gevonden op lineaire groei, morbiditeit of systemische inflammatie. In vergelijking met de controlegroep werden er in de KI-interventiegroep significant grotere maandelijkse veranderingen gevonden in de gewicht-voor-lengte z-scores [effectgrootte (95% KI): 0,026 (0.004, 0.048) per maand] alsook in de MKI-interventiegroep [(effect grootte (95% KI): 0.022 (0.000, 0.043) per maand]. Tot slot toonde de studie aan dat het innemen van een n-3 LC-PUFA supplement bij lacterende vrouwen en kinderen geen invloed had op de lineaire groei en morbiditeit van kinderen in een milieu van lage inkomens. Het n-3 LC-PUFA-supplement dat rechtstreeks aan kinderen werd gegeven, verhoogde de relatieve gewichtstoename, maar de kwaliteit van deze gewichtstoename werd niet geverifieerd met metingen van de lichaamssamenstelling. Welbeschouwd ondersteunden deze bevinding het reeds bestaande bewijs dat er een gebrek is aan een effect van n-3 LC-PUFA-suppletie op lineaire groei, morbiditeit en ontsteking.

Samengevat leverde dit doctoraatswerk bewijs met betrekking tot het niveau van het voedingsgedrag van zorgverleners, de bruikbaarheid van een eenvoudige op voedingsgroepen gebaseerde indicator voor baby- en kindervoeding als een belangrijk hulpmiddel bij de evaluatie van voedingskwaliteit en -groei ongeacht de landbouwseizoenen. Het is echter aanbevolen om een samengestelde zuigelingen- en kindervoedingsindex samen te stellen met behulp van extra componenten en het gebruik van een prospectief design. De bevindingen van onze n-3 LC-PUFA-interventiestudie leverden vergelijkbaar bewijs van het gebrek aan effect op de lineaire groei en morbiditeit. Zoals ander onderzoek in de setting van ontwikkelingslanden wordt verder onderzoek aanbevolen met een grotere dosis en een vroegere initiatie van n-3 LC-PUFA-suppletie. Het ontwikkelen of aanpassen van een maatstaf voor kinderontwikkeling, gemeten met een specifieke testbatterij is nodig om het bewijs aan te sterken over het effect van n-3 LC-PUFA-supplementen op kinderontwikkeling in deze setting.

Curriculum vitae of the author

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Mekitie Wondafrash was born in Debre Berehan (Ethiopia) on August 5 and raised in the rural district near the town. After completing high school in 1995, he joined the former Jimma Institute of Health Sciences (currently Jimma University) and studied medicine and graduated with medical doctorate degree in 2001. Shortly after graduation, he joined Jimma University in the former Community Health Program (currently Faculty of Public Health) as assistant lecturer. Two year later, he secured a scholarship from Belgium government (via VLIR-OUS) to follow a graduate program in Food Science and Nutrition at Ghent University in Belgium. Upon returning from Belgium in 2004, he got reinstated at the same department (Department of Population and Family Health). Since then, he has been serving as department head, coordinator of the first graduate program in the history of the Jimma University and as research team leader. Through the institutional collaboration established between Jimma University and Flemish Universities in Belgium, he has been actively involved in a research project entitled “Child Health and Nutrition Project” and served as principal investigator and project leader since July 2007. He has 26 publications in local and international peer reviewed journals. Moreover, he has been involved in teaching learning process of graduate students in nutrition and public health at Jimma University and served as a guest lecturer in various public universities outside of Jimma. Since 2010, he has been working as a Ph.D. fellow at Ghent University under a Ph.D. title of:“*The role of feeding practices, dietary diversity and seasonality and n-3 long-chain polyunsaturated fatty acids in the diet of Ethiopian infants and young children*”.

<u>Selected peer review publications with scientific citation index (A1 or A2)</u>

1. Argaw A., **Wondafrash M,** Kolsteren P, Bouckaert KP, Lachat C, Belachew T, De Meulenaer B, and Huybregts L; effects of n-3 LC-PUFA supplementation through breast milk and complementary food on growth and morbidity of children 6-24 months: a randomized controlled trial in rural Ethiopia (accepted for publication at The at American Journal of Clinical Nutrition)
2. **Wondafrash M,** Huybregts L, Lachat C, and Bouckaert KP, Kolsteren P: Feeding practices and growth among young children during two seasons in rural Ethiopia. BMC Nutrition 2017, 3(1):39.

3. Worku, B. N., Abessa, T. G., **Wondafrash, M**., Vanvuchelen, M., Bruckers, L., Kolsteren, P. and Granitzer, M. (2018); The relationship of undernutrition/psychosocial factors and developmental outcomes of children in extreme poverty in Ethiopia. BMC Pediatrics, 18(1), pp. 45.
4. Worku, B. N., Abessa, T. G., **Wondafrash, M**., Lemmens, J., Valy, J., Bruckers, L., Kolsteren, P. and Granitzer, M. (2018) Effects of home-based play-assisted stimulation on developmental performances of children living in extreme poverty: a randomized single-blind controlled trial. BMC Pediatrics, 18(1), pp. 29.
5. Abessa TG, Worku BN, **Wondafrash M,** Valy J, Lemmens J, Thijs H, Yimer WK, Kolsteren P, Granitzer M. Adaptation and standardization of a Western tool for assessing child development in non-Western low-income context. BMC Public Health 2016; 16(1):1-13. doi: 10.1186/s12889-016-3288-2.
6. James P, Sadler K, **Wondafrash M**, Argaw A, Luo H, Geleta B, et al. (2016) Children with Moderate Acute Malnutrition with No Access to Supplementary Feeding Programmes Experience High Rates of Deterioration and No Improvement: Results from a Prospective Cohort Study in Rural Ethiopia. PLoS ONE 11(4): e0153530. doi:10.1371/ journal.pone.0153530
7. Andarge, E., Nigussie, A. and **Wondafrash, M**. (2017) Factors associated with birth preparedness and complication readiness in Southern Ethiopia: a community based cross-sectional study. BMC pregnancy and childbirth, 17(1), pp. 412.
8. **Wondafrash, M**., Huybregts, L., Lachat, C., Bouckaert, K., & Kolsteren, P. (2016). Dietary diversity predicts dietary quality regardless of season in 6–12-month-old infants in south-west Ethiopia. Public Health Nutrition, 19(14), 2485-2494. doi:10.1017/S1368980016000525
9. Mekonen S, Lachat C, Ambelu A, Steurbaut W, Kolsteren P, Jacxsens L, **Wondafrash M**, Houbraken M, Spanoghe P. Risk of DDT residue in maize consumed by infants as complementary diet in southwest Ethiopia. Science of the Total Environment 2015; 511:454-60.
10. Asfaw M, **Wondafrash M**, Taha M, Dube L. Prevalence of undernutrition and associated factors among children aged between six to fifty nine months in Bule Hora district, South Ethiopia. BMC public health 2015;15(1):1.
11. **Wondafrash M,** Amsalu T, Woldie M. Feeding styles of caregivers of children 6-23 months of age in Derashe special district, Southern Ethiopia. BMC Public Health 2012;12(1):1-8. doi: 10.1186/1471-2458-12-235

12. Tekeste A, **Wondafrash M**, Azene G, Deribe K. Cost effectiveness of community-based and in-patient therapeutic feeding programs to treat severe acute malnutrition in Ethiopia. Cost Effectiveness and Resource Allocation: C/E 2012;10:4-doi: 10.1186/1478-7547-10-4.
13. Addisu F, **Wondafrash M**, Chemali Z, Dejene T, Tesfaye M. Length of stay of psychiatric admissions in a general hospital in Ethiopia: a retrospective study. International journal of mental health systems 2015;9(1):1.
14. Bogale B, **Wondafrash M**, Tilahun T, Girma E. Married women's decision making power on modern contraceptive use in urban and rural southern Ethiopia. BMC Public Health 2011;11(1):1-7. doi: 10.1186/1471-2458-11-342.
15. Deribe K, Woldemichael K, **Wondafrash M**, Haile A, Amberbir A. Disclosure experience and associated factors among HIV positive men and women clinical service users in southwest Ethiopia. BMC Public Health 2008;8:81-. doi: 10.1186/1471-2458-8-81.
16. Yirga, D., Deribe, K., Woldemichael, K., **Wondafrash, M**. and Kassahun, W. (2010) Factors associated with compliance with community directed treatment with ivermectin for onchocerciasis control in Southwestern Ethiopia. Parasites & Vectors, 3(1), pp. 48.
17. Yohannes S, **Wondafrash M,** Abera M, Girma E. Duration and determinants of birth interval among women of child bearing age in Southern Ethiopia. BMC pregnancy and childbirth 2011; 11(1):1.

Papers under review

1. Asfaw Ayalew; **Mekitie Wondafrash,** Hirut Assaye; Diet-related chronic diseases and risk factors in Ethiopia: a systematic review (Under review in BMC Public Health)
2. Teklu Gemechu,; Berhanu Nigussie Worku; **Mekitie Wondafrash**; Tsinuel Girma Nigatu; Johan Valy; Johan Lemmens; Liesbeth Bruckers; Patrick Kolsteren; Marita Granitzer Effect of play-based family-centered psychomotor/psychosocial stimulation on the recovery of severely acutely malnourished under-six children in a low income setting (submitted to PLOS ONE)

Manuscript to be submitted

1. Alemayehu Argaw, Lieven Huybregts, **Mekitie Wondafrash,** Patrick Kolsteren, Tefera Belachew, Berhanu N. Worku, Teklu Gemechu and Kimberley P. Bouckaert ; Effects of N-3 LC-PUFA Supplementation through Lactation and Supplementary Feeding on Development of Children 6-24 Months: a Randomized Controlled Trial in Rural Ethiopia

Oral or Poster presentation

2. **2011 (September): Mekitie Wondafrash,** Patrick Kolsteren; Nutritional status and associated factors among 6-59 months old children in the Gilgel Gibe Field Research Center of Jimma University , Ethiopia : a baseline study for the Child Health and Nutrition Project of JU-IUC Presented on a "A National Community Day" organized by The Institute of Health Science Research, Jimma University, September 2011 (Poster presentation)
3. **2014 (October):** James, P., Sadler, K., **Wondafrash, M.,** Argaw, A., Luo, H., Geleta, B., Kedir, K., Getnet, Y., Belachew, T. and Bahwere;Outcomes of MAM among under five children: a prospective cohort study from a food-secure setting in rural Ethiopia: (presented a part of ongoing OR for the implementation of the National Nutrition Program, organized by EPHI , Adama, Ethiopia) (http://www.ephi.gov.et/images/pictures/MAM-Engine-MW.pdf) (**Oral presentation)**
4. **2016 (June):** James, P., Sadler, K., **Wondafrash, M.,** Argaw, A., Luo, H., Geleta, B., Kedir, K., Getnet, Y., Belachew, T. and Bahwere;Children with Moderate Acute Malnutrition with No Access to Supplementary Feeding Programmes Experience High Rates of Deterioration and No Improvement: Results from a Prospective Cohort Study in Rural Ethiopia (presented at the national research dissemination workshop organized by USAID/ENGINE, Addis Ababa, Ethiopia). (**Oral presentation).**

Trainings

1. **2013 (October):** Introduction to clinical trials : offered by Dr Saghir Bashir from European Food Safety Authority, organized by JU-IUC
2. **2015 (October):** Ethical Aspects of Scientific Research and Communication offered by Prof. Oswald Van Cleemput, (Emeritus professor from Ghent University, Belgium), The training workshop was organized by JU-IUC

Scholarships

1. **2003**: Secured a scholarship from the Belgium (through VLIR-UOS) to attend an international graduate program in Food Science and Nutrition (2003-2004)
2. **2010**: Obtained a scholarship to follow a sandwich Ph.D. at the Department Of Food Safety And Food Quality, Faculty of Bioscience Engineering, and Ghent University as part of Child Health and Nutrition Project of JU-IUC / VLIR-UOS.