COMMUNITY INTERVENTIONS TO IMPROVE IRON AND IODINE STATUS IN MOTHER AND CHILD DYADS IN NORTHERN GHANA

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ABSTRACT

COMMUNITY INTERVENTIONS TO IMPROVE IRON AND IODINE STATUS IN MOTHER AND CHILD DYADS IN NORTHERN GHANA.

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Ghana, especially northern Ghana has high prevalence of micronutrient deficiencies due to diverse reasons which include; high food insecurity prevalence, poor yields, inability to cultivate food during the prolonged dry/lean season (7-8 months/year), unavailability of dietary approaches to screen for micronutrients deficiencies, and limited information on efficacy of native diets to improve nutrients status. Our research aims at contributing to resolve these challenges with our main focus being on trace minerals (iron and iodine). Four hypotheses of this research are: 1) dietary diversity score (DDS) and/or dietary patterns (DP) can predict the risk of iron and iodine deficiencies in women and their toddlers; 2) standardized native iron-rich *Hibiscus Sabdariffa* leaves meal is efficacious in improving iron status of mother-child dyads with time during dry/lean season; 3) weekly household iodized salt supply can improve iodine status of mother-child dyads; and 4) container gardening can be used to cultivate *Hibiscus Sabdariffa (HS)* and cabbage during the dry/lean season, producing adequate amount of *HS* for mother-child dyads consumption and economic gains from cash crop cabbage for purchase of iodized salt and dry fish. In a cross-sectional study (women: n=118; children: n=121), the relationship between DDS (DDS<4 & DDS≥4) and dietary pattern scores (tertiles) and the risk of iron and iodine deficiencies (hypothesis 1) was tested by multivariable logistic regression models. In a quasi-experimental community-based feeding trial for 12 weeks (hypothesis 2), multiple logistic regression was conducted

using GENMOD in comparing the groups' iron status. Median UICs of both groups before and after intervention (hypothesis 3) was compared by chi-square tests. For hypothesis 4, women groups in two communities farmed 40 wooden containers to grow indigenous iron-rich *HS* for consumption and 15 containers for cash crop cabbage in the dry/lean season. Community and participants' characteristics, yields, and consumption were compared between intervention and control communities by descriptive statistics. We found that DDS and DP respectively predicted the risk of iron and iodine deficiencies in women but not in their toddlers after controlling for sociodemographic characteristics; native meal made of *HS* leaves was efficacious in improving iron status of women with time and protected toddlers from stunting during dry/lean season; weekly household iodized salt supply improved mother-child dyads iodine status; and containers can be used to cultivate iron-rich *HS* and cabbage during the dry/lean season, producing adequate amount of *HS* for mother-child dyads consumption and economic gains from cash crop cabbage production for purchase of iodized salt and dry fish, an ingredient of *HS* meals. These findings provide scientific bases to guide recommendations and policy making on programs targeted at reducing the risk of iron and iodine deficiencies in northern Ghana.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

KEY TO ABBREVIATIONS

AOR: Adjusted odds ratio

BND: Builsa north district

CHPS: Community-Based Health Planning and Services

DDS: Dietary diversity score

DP: Dietary pattern

HAZ: Height-for-age z-score

Hb: Hemoglobin

HDDS: Household dietary diversity score

HSM: *Hibiscus Sabdariffa* leaves meal

ID: Iron deficiency

IDA: Iron deficiency anemia

KNWD: Kassena Nankana West

PCA: Principal component analysis

Stfr: Serum transferrin receptor

UIC: Urinary iodine concentration

WHZ: Weight-for-age z-score

Chapter One: Introduction

Iron and iodine deficiencies are of public health concern and specifically detrimental to maternal and child health. Iron deficiency (ID) is the most common form of nutritional deficiency with children and childbearing age women being at the highest risk globally (Centers for Disease Control Prevention, 2002; Gayer and Smith, 2015; Stoltzfus, 2003). In Ghana, the prevalence of ID, iron deficiency anemia (IDA), and anemia are highest among children (ID:78% in northern Ghana in 2012), (IDA:63% in northern Ghana in 2012), and (anemia: 66% in 2014), and women of childbearing age (anemia:42% in 2014) (Abizari et al., 2012; Ewusie et al., 2014; GSS, 2009; 2011; 2014; Zimmermann and Hurrell, 2007) and stunting in children (19%) are high. The three northern regions (Upper East, Upper West and Northern regions) have high prevalence of anemia (74-82%) and stunting rate (14-33%) compared to **southern** regions (Anemia: 54-70% and stunting: 10-22%). (GSS, 2009; 2011; 2014; WFP, 2015). This prevalence increases during the dry/lean season (7-8 months/year) where food insecurity peaks in northern Ghana and agricultural production is very difficult for inhabitants who are mainly peasant farmers.

Iodine deficiency or iodine deficiency disorders (IDD), a collective number of health outcomes resulted by iodine deficiency, is the world's leading cause of preventable cognitive retardation and poor psychomotor development in children (Zimmermann et al., 2008b). Additionally, iodine deficiency is commonly associated with goiter, intellectual impairments (Delange, 1994), growth retardation, neonatal hypothyroidism, and increased pregnancy loss and infant mortality (Haldimann et al., 2015; Pearce et al., 2013). Globally, iodine deficiency is the leading cause of intellectual deficiency (Hollowell et al., 1998; Zimmermann, 2009). About 1.88 billion people have insufficient dietary iodine intakes with most of them being economically disadvantaged groups living in remote areas including Africa (Andersson et al., 2012). Household iodized salt usage survey in 2014 reported that 69% of households in Ghana reported using iodized salt. The three northern regions have highest unmet needs (consume salt with no or with iodine<15 ppm): Northern (84%), Upper East (83%), and Upper West (72%) regions of Ghana. Currently iodine deficiency prevalence among women of reproductive age and children in Ghana is unknown. No data are available on urinary iodine concentration. Data is only available for household usage of salt fortified with iodine at the level recommended by WHO (≥15 ppm).

Globally, approaches to counteract iron and iodine deficiencies have included food fortification, supplementation and increased dietary diversity. The key challenges with the fortification and supplementation approaches in developing countries are limited access and distribution to rural households, cost, and sustainability (Yip and Ramakrishnan, 2002). Particularly the majority of subsistence farmers rely on limited variety and amount of foods they produce locally. Furthermore, researchers have questioned benefits of foods fortified with elemental iron as risks for infections may outweigh the benefits in developing countries with high prevalence of malaria (Sazawal et al., 2006). One alternative approach could be food-to-food fortification without incorporation of elemental iron to maximize the benefits of indigenous iron-rich food sources. Currently limited reports are available on the creative efforts and efficacy of utilizing indigenous iron-rich foods in the regions with high risk of ID.

Iodine comes from diet or supplementation through iodized salt. Dietary sources of iodine however vary from country to country and among regions within a country as near-coastal soils are more enriched with iodine than inland soils. One highly concentrated source of iodine is seafood (Fuge and Johnson, 2015). The alternative source of iodine is iodized salt (Lynch, 2011; Ross et al., 2014). Iodized salt is the most common source of iodine for most African countries including Ghana. Ghana adopted a universal salt iodization policy for over two decades ago, yet iodized salt use and intake is inadequate. WHO recommends >90% households in the country use I-salt fortified at or above 15 ppm iodine (Pearce et al., 2013). Among several reasons, cost is often named as one of the key barriers for rural people (Rasheed et al., 2001). A need to check for iodine deficiency prevalence and the role of iodized salt in improving the iodine status of the population is urgently desired. Additionally, investigation of the usage of alternative iodine sources, such as bouillon cubes, is necessary.

The overarching aim of this doctoral dissertation research was to investigate if 1) dietary diversity score (DDS) and dietary patterns (DP) can predict the risk of iron and iodine deficiencies in women and their toddlers in Upper East region during its long dry/lean season, 2) if consumption of *Hibiscus Sabdariffa* meal thrice a week could protect toddlers from stunting and improve mother-child dyads iron status , 3) if provision of iodized salt to each household weekly could improve mother-child dyads iodine status, and 4) if container garden can produce adequate amount of the iron-rich vegetable (*Hibiscus Sabdariffa*) and cash crops (cabbage) during the dry/lean season to sustain mother-child dyad's consumption and economic gains for purchase of iodine-rich and iron-rich ingredients (iodized salt and dry fish).

1.1 Significance

Subsistence farming is the most common way of life in northern regions in Ghana where the dry/lean season lasts 7-8 months a year and is further exacerbated by climate changes. Food and nutritional insecurity in the northern Ghana is in a dire situation as evidenced by higher prevalence of trace mineral deficiencies and stunting with lower rates of adoption of universal salt iodization, compared to those in southern regions of Ghana. The rates of anemia between the three northern regions and the remaining seven regions of Ghana has been persistently high at alarming rates (GSS, 2009; 2011; 2014; WFP, 2015). The disparity is evidenced by recent rates of anemia at 82.1% (northern Region), 73.8% in Upper East and Upper West Regions vs. a percentage range of 53.7% (Ashanti Region) to 70.2% (central region) in southern Ghana (GSS, 2014). Estimated inadequate intake of iodine by the 2014 Ghana demographic and health survey suggest high prevalence of deficiency in northern Ghana. To reduce the high prevalence and potential detrimental effects of iron and iodine deficiencies, there is a need to assess, screen and intervene by non-invasive dietary intake methods, compared to costly and laborious biomarkers or non-specific clinical examinations. The key challenge with this approach is the lack of food composition tables of indigenous foods in Ghana. This project addresses the persisting problems by looking for ways (via our study aims) to fill the gap in our knowledge and practices pertaining to serious trace mineral deficiencies that result in stunting and cognitive impairment in northern Ghana. Both iron and iodine deficiencies are closely associated with impaired somatic and cognitive development of fetus and children, and birth outcomes of pregnant women.

Several approaches have been used globally to curb iron and iodine deficiencies. The commonly held approach is iron fortification of foods but there have been reports of fortification showing little success in Africa (Abizari et al., 2012; Zimmermann and Hurrell, 2007) as benefits of foods fortified with elemental iron may not outweigh the risks with increased infections in developing countries with high prevalence of malaria. The need to explore the efficacy of indigenous iron-rich foods consumed by rural populations who are often at the highest risk of ID is crucial.

Iodine is an essential component of thyroid hormones and must be obtained from the diet (Jooste and Strydom, 2010). Iodine content in native foods is usually low and varies geographically and across a region (Assey et al., 2006). The variation is due to differences in geological formation, such as flooding, soil erosion, human activities especially densely populated areas and glaciations (Zimmermann et al., 2008a). Ghana implemented a universal salt iodization policy in 1996 (Nyumuah et al., 2012a) as iodized salt is the key source of dietary iodine. As of 2013, Ghana suffers from moderate iodine deficiency (fig 1) (Pearce et al., 2013), this might even be worse in the three northern regions. 2014 demographic household survey indicates that the proportions of households using adequately iodized salt has been low (16-28%). This low usage level suggests a possible high presence of iodine deficiency. A need to check for iodine deficiency prevalence is thus crucial. Research into the role of iodized salt in improving iodine status of the population is urgently needed.

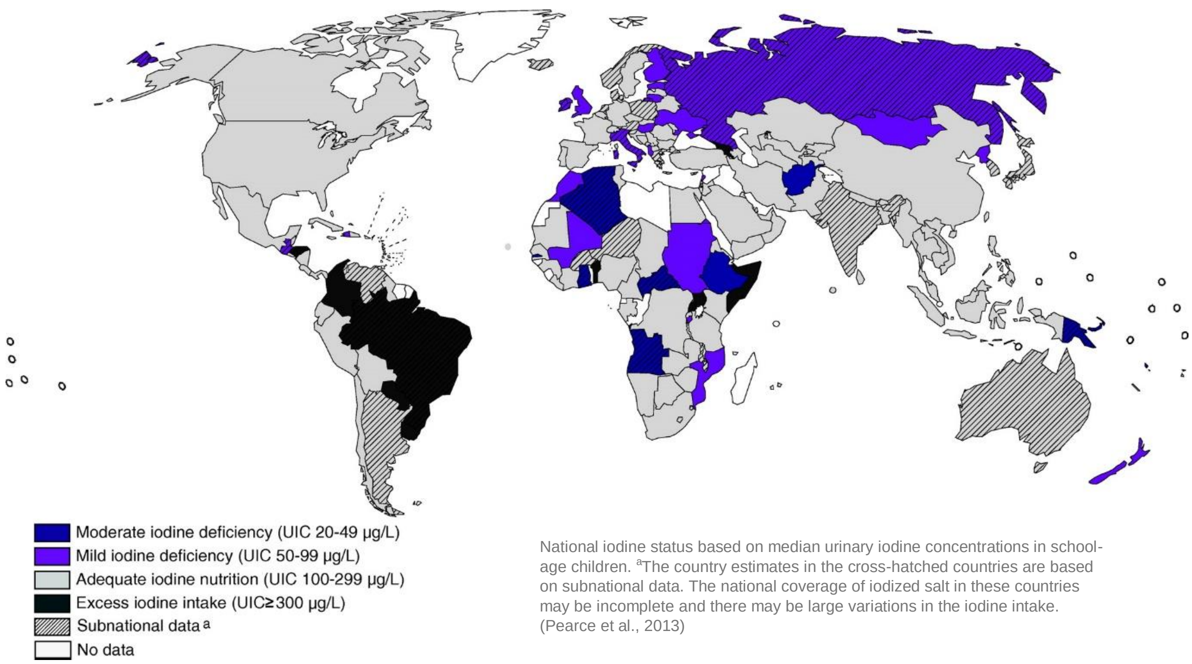

Figure 1. A map of iodine status

Although inadequate iron and iodine in diet is the major cause of anemia and iodine deficiencies, little has been done to improve the accessibility of iodine rich and iron-rich food sources for the vulnerable groups, particularly in the peak of food insecurity, during dry/lean season in most parts of Africa including Ghana. The need to explore alternative and non-conventional approaches such as using containers to cultivate iron-rich *Hibiscus Sabdariffa* and cash crop (cabbage) during the dry/lean season for mother-child dyads consumption and provide economic gains from cash crop cabbage production for purchase of iodized salt and dry fish. The dry fish is an important food ingredient for the “meal” that has been traditional or compatible for taste that indigenous people are familiar with. this can be an extraordinary source of high quality protein that lacks in people’s diet, enhances iron absorption from plant sources (*Hibiscus Sabdariffa*), and becomes an important source of calcium. The results from our research provide scientific bases to guide recommendations and policy making on programs targeted at reducing the risk of iron and iodine deficiencies, particularly in northern Ghana during the dray/lean seasons.

1.2 Innovation

Screening and intervening by non-invasive dietary intake methods, compared to costly and laborious biomarkers or non-specific clinical examinations are needed. The prevalence of iodine and iron deficiencies have been persistent in northern Ghana for years with several approaches being adopted but none has explored assessment. Screening by dietary intake methods may serve as a basis for screening for early detection of iron and iodine deficiencies. Beyond screening, the efficacy of indigenous foods could serve as local and accessible solutions to reduce iron deficiency prevalence. Demonstrating the feasibility of using non-conventional approaches such as using containers to cultivate iron-rich *Hibiscus Sabdariffa* and cabbage during the dry/lean season for mother-child dyads consumption, provide economic gains for purchase of iodized salt and dry fish may be a game changer in the fight against micronutrients deficiencies. The novelty of this approach does not only lie in making iron rich foods accessible but also solves the problem of scarcity of production plots for peasant farmers, and water as cultivation is done in controlled and portable environment using relative less water.

Chapter Two: Literature Review

2.1 Food security and nutrition security

Food security has several definitions. Among them is FAO's definition "when all people, at all times, have physical, social and economic access to sufficient, safe and nutritious food that meets dietary needs and food preferences for an active and healthy life" (Qureshi et al., 2015). Anything short of the definition results in food insecurity. Others simply put food insecurity as the uncertain availability of nutritionally adequate, safe foods for healthy life (Weiser et al., 2013).

Ghana has been reported to have less than 9% of its population being food insecure in 2011-2013 (FAO, 2015), based on caloric availability per person which does not reflect the true meaning of the FAO definition. The most food insecure households are found in northern Ghana: The highest 28% in the Upper East Region, 16% in the Upper West Region and 10% in Northern Region (Table 1). As indicated in Table 1, the five districts with the highest proportion of severely and moderately food insecure households are Wa West (42%), Central Gonja (39%), Talensi-Nabdam (39%), Kassena-Nankana West (35%) and Kassena- Nankana East (33%) (WFP, 2012).

able 1. Severely or moderately food insecure households in northern Ghana and by district in Upper East region

Region	District	Percent of households
Upper East		28.2
	Bawku Municipality	25.2
	Bawku West	6.3
	Bolgatanga Municipality	28.2
	Bongo	20.2
	Builsa*	39
	Garu-Tempane	29.1
	Kassena-Nankana East	33.2
	Kassena-Nankana West	34.9
	Talensi-Nabdam	39.1
Upper West		16.2
	Jirapa	23.8
	Lambussie-Karni	21.3
	Lawra	11.8
	Nawdowli	2.7
	Sissala East	6.8
	Sissala West	2.3
	Wa East	24.5
	Wa Municipal	12.3
	Wa West	42.1
Northern		9.7
	Bole	28.2
	Bunkpurugu-Yunyoo	20.3
	Cherepone*	25.7
	East Gonja	0.9
	Central Gonja	39.2
	Gushegu	22.2
	Karaga	11.0
	Kpandai	6.8
	East Manprusi	14.5
	West Manprusi	5.0
	Nanumba North	0.5
	Nanumba South	0.9
	Saboba	10.9
	Savelugu-Nantong	5.0
	Sawla-Tuna-Kalba	8.2
	Tamale Metropolis	3.6
	Tolon-Kumbungu	14.9
	West Gonja	8.2
	Yendi	9.5
	Zabzugu-Tatale	10.0

* Food consumption data cannot be treated as representative due to small sample size (WFP, 2012)

The causes of these prevalence are complex and are largely due to poverty, agricultural limitations, seasonal challenges and high food prices (Fig 1). In all these, small holder farmers, households headed by women, women and children suffer most. The consequences of food insecurity reflect in high malnutrition prevalence as evidence by the rates of anemia in children (74-82%) and stunting (14.4-33.1%) in northern Ghana (GSS, 2009; 2011; 2015). With the reported food insecurity prevalence of less than 9% based on the access to caloric needs, stable political environment, relative economic growth and improved health system over the years in Ghana (IFPRI, 2014), it is expected that malnutrition prevalence would have been improved.

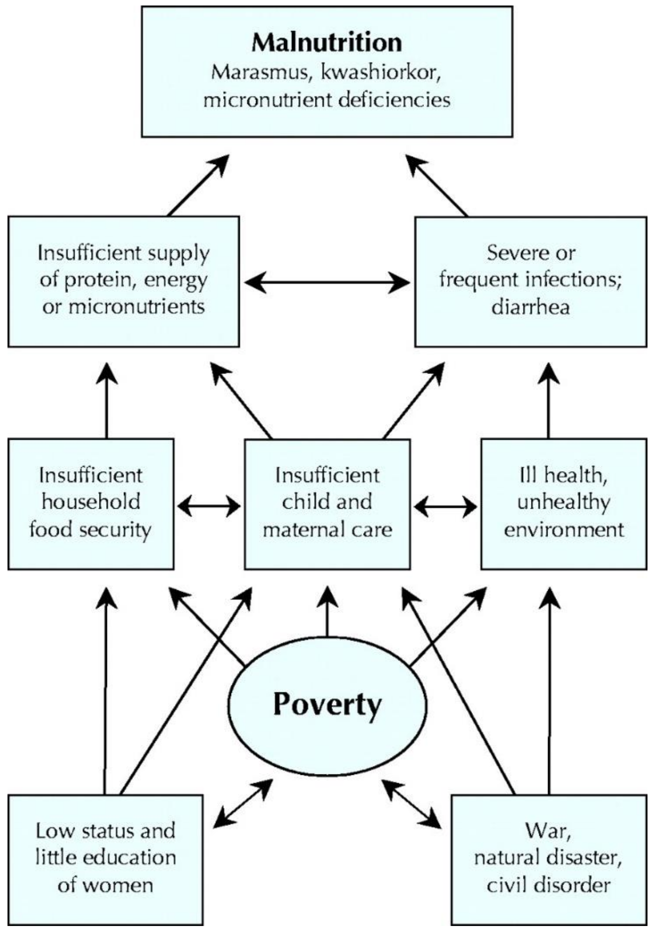

Figure 2. Direct and indirect causes of malnutrition (Müller and Krawinkel, 2005)

Nutrition security differs from food security. Nutrition security reflects ‘food’, ‘health’ and ‘care’ collectively. Thus, nutrition security cannot be achieved in the absence of food security (FAO, 2009). Nutritional security thus requires measurement of functional status of nutrients, other than calories, and is likely underestimated by food security measurements, just like caloric intake underestimates food security.

In sum, food insecurity prevalence is high in northern Ghana and women and children are affected most. Although food security and nutrition security are related, food security measurements cannot be used to imply nutrition security. There is a need to use dietary based approaches and to develop screening tools to specifically measure nutritional status (not caloric intake). Nutrients such as iron, iodine and zinc are critically important in growth and development of children and pregnant women in order to break intergenerational vicious cycle of poverty that results in poor health, growth and cognitive development.

2.2 Micronutrients in human health

Micronutrients are essential vitamins and minerals required in small amounts by the body for proper growth and development. They include vitamins A, B, C and D, calcium, folate, iodine, iron, and zinc (Gernand et al., 2016). They play vital biological roles, and some are considered as problematic nutrients because of wide spread of deficiencies. Key problematic micronutrients in Ghana are iron, iodine and zinc (Gayer and Smith, 2015; Stewart et al., 2013). They play major important biological functions in the development of fetus, children and pregnant women, and multiple deficiencies are common (Kontic-Vucinic et al., 2006). Iron, iodine, and zinc are the most widespread trace mineral deficiencies globally (Gayer and Smith, 2015). Controlling deficiencies in women of childbearing age can break the intergenerational malnutrition cycle (Berti et al., 2014; Micke et al., 2014; Radlowski and Johnson, 2013; Wang et al., 2015) since maternal nutrition has great influence on the birth outcomes of their offspring as well as their own health. It is known that maternal deficiencies in iron, iodine, and zinc among other factors affect intrauterine growth of fetus (Gayer and Smith, 2015; Stewart et al., 2013) and that infants with both symmetric and asymmetric intrauterine growth retardation demonstrate limited catch-up growth which persists into early childhood (Strauss and Dietz, 1997). Nutritional status in childhood affects nutritional and reproductive health in adulthood. This invariably affects health status and productivity of subsequent generations. Studies among women of childbearing age is vital especially with respect to iron, iodine, zinc, and selenium (Malhotra et al., 2014; Radlowski and Johnson, 2013; Schmidt et al., 2014; Valera-Gran et al., 2014).

Iron is one of the problematic nutrients in the health of women and children. Iron in diets exists in two forms: ferrous form (Fe^{2+}) is from animal sources of foods present in heme, and the ferric form (Fe^{3+}) is from plant sources and in non-heme iron. Total human body iron content is estimated to be 3.8 g in men and 2.3 g in women. Most iron in the body is in the form of heme iron. Heme iron is the essential constituent of hemoglobin in blood and myoglobin in muscle for oxygen transport, oxygen storage, electron transport for cytochrome function in aerobic respiration, and signal transduction as a cofactor for nitric oxide synthase and guanylyl cyclase. Iron is essential for immune and cognitive development and functioning. The second largest pool of iron in the body is in its storage form ferritin, a ubiquitous intracellular protein (Ross et al., 2014).

Utilization of iron in food sources by humans and animals is highly influenced by bioavailability. Iron bioavailability is however lowered by such other food substances as oxalic acid, tannins, phytate, polyphenols, carbonate, phosphate, fiber and other metal ions. These bioabsorption inhibitors form non-absorbable complexes with the iron in the intestinal lumen. On the other hand, vitamin C, fructose, citric acid, dietary protein, lysine, histidine, cysteine, and methionine enhances iron absorption (Haider et al., 2013). These bioavailability enhancers form a chelate with ferric iron that remains soluble at the alkaline pH of the duodenum (Lynch and Cook, 1980)

Under normal physiological conditions, low serum ferritin value reflects depleted iron stores, but not necessarily the severity of the depletion as it progresses. Presence of inflammation impacts negatively on iron status as well as its assimilation from the gastrointestinal tract. Infections (notably malaria, HIV disease, and tuberculosis), parasites (tape and hook worms), and inherited disorders of erythropoiesis (such as the thalassemic syndromes and hemoglobinopathies) lower serum ferritin concentration (Humphries et al., 2011; Lynch, 2011). Additionally, in the presence of inflammation, hepcidin inhibits iron absorption which ultimately affects individuals' iron status (del Giudice et al., 2009). In areas where inflammation is prevalent, the concentration of soluble transferrin receptor is usually used to assess iron status as it does not rise in response to inflammation (WHO, 2011).

Deficiencies of iron are associated with morbidity, mortality, preterm birth, low birthweight and inferior cognitive development (Haider et al., 2013). Iron deficiency is most common among women and children due to higher physiological needs and iron losses (menses in women). The burden is heaviest in developing countries largely due to poor diets and infections. Anemia is reported in 52% of pregnant women in non-industrialized and 23% of those in industrialized countries. Children in industrialized and non-industrialized countries are also reported to suffer from anemia (WHO, 2001) at 20% and 39%, respectively. In Ghana, anemia is seen in 66% of children under 5 yrs and 42% of women of 15-49 years of age (GSS, 2014).

Iodine, is essential to living organisms as it is an intramolecular component for the biosynthesis of thyroid hormones. Thyroid hormones control cell growth and differentiation, and increase metabolism of proteins, lipids, and carbohydrates (Haldimann et al., 2015). Iodine Deficiency Disorders (IDD), a collective number of health outcomes, is the world's leading cause of preventable cognitive retardation and poor psychomotor development in children. Additionally, iodine deficiency is commonly associated with goiter, intellectual impairments, growth retardation, neonatal hypothyroidism, and increased pregnancy loss and infant mortality (Haldimann et al., 2015; Pearce et al., 2013).

Globally, iodine deficiency is the world's leading cause of intellectual deficiency (Hollowell et al., 1998). About 1.88 billion people have insufficient dietary iodine intakes with most of them being economically disadvantaged groups living in remote areas (Andersson et al., 2012) including Africa. In Ghana, only 35% of households were reported using adequately iodized salt in the 2014 demographic health survey (GSS, 2014). The rate was much lower in Northern (16%) and Upper East (17%) regions of Ghana compared to other such regions as Western (60%), Accra (55%), Ashanti (46%), Brong Ahafo (35%) Generally, the rate of households using adequately iodized salt is much lower in rural areas (26%) than in urban areas (50%)(GSS, 2014).

Iodine needs are mainly met from diet or supplementation. Deficiency can lead to iodine deficiency disorders (IDD) such as hypothyroidism, metal impairment, goiter, cretinism, etc. Dietary sources of iodine vary from country to country and from one local area to another. Iodine is rich in seafood and in near-coastal soils (Fuge and Johnson, 2015). The rate of deficiency tends to be lower in countries with high consumption of sea products. In some instances, milk could be a rich source of iodine when cows are fed with iodine supplements. The alternative source of iodine is iodized salt (Lynch, 2011; Ross et al., 2014). Iodized salt is the most common source of iodine for the world, and most African countries, including Ghana, adopted universal salt iodization. From the usage level of iodized salt, it could be inferred that 72-84% and 40-87% of households in northern and southern Ghana respectively could be at risk of iodine deficiency (GSS, 2014). This makes the present study of iodine deficiency among the vulnerable groups very timely. Additionally, limited data exist on urinary iodine concentrations (UIC) at the population level. Household iodized salt usage is a proxy for assessing population-based iodine status and not necessarily a true indicator of iodine status at the population level (WHO, 2007).

Deficiencies are generally assessed by measured UICs which is considered cost effective and reliable. Median urinary iodine concentrations between 100-299 µg/l in children and non-pregnant women indicate no iodine deficiency in a population. World Health Organization further states that, not more than 20% of a sampled population UICs should be below 50 µg/l. Cut offs are higher in pregnancy, median UICs between 150- 249 µg/l in a population indicates no iodine deficiency (World Health Organization, 2007).

2.3 Dietary diversity score (DDS) as a measure of diet quality

DDS has been used as an indicator of food quality as well as a measure of food security (Hoddinott, 2002; Kennedy et al., 2009; Ruel, 2003). DDS is determined by the number of foods or food groups consumed by an individual (IDDS) or by any member of the household (HDDS) over a reference time period, which is usually 24 hours (UN, 2008). Food grouping is dependent on the study objectives, such as emphasis on energy-dense foods or micronutrient-rich foods. Usually, the total number of food groups range from 5 to 14, depending on the main characteristic of the diet that the score intends to reflect (UN, 2008).

In Niger and Kenya, dietary diversity was positively associated with anthropometric status of children under five years (Onyango et al., 1998; Tarini et al., 1999). Additionally in Niger, dietary diversity was positively associated with intake of such micronutrients as iron, zinc and vitamin A (Tarini et al., 1999).In Ghana and Malawi however, weaker or no association was found for most nutrients (Ferguson et al., 1993).

Household level food group diversity (HDDS) and energy intake showed consistently positive association in a multi-country study (Hoddinott, 2002). In Venezuela, household food consumption of 12 or more food items explained 90.7% of the variance in predicting total energy intake (Lorenzana and Sanjur, 1999). A similar result has also been reported in another report (Lorenzana and Mercado, 2002). These studies are context-specific, and thus applications of the results should be carefully designed for each population based on their specific dietary patterns.

Individual dietary diversity measures (IDDS) obtained from 24-hour recalls or food frequency questionnaires showed positive correlation with energy intake in several studies (Kennedy et al., 2007; Moursi et al., 2008). In Mali, Torheim and colleagues (Torheim et al., 2004) tested two dietary indicators in association with energy intake: one based on the number of unique foods consumed by the individual (food variety score - FVS) and the other based on the number of food groups consumed (dietary diversity score – DDS). They found both FVS and DDS to correlate positively with energy intake, with coefficients of 0.38 and 0.29, respectively (Torheim et al., 2004). Also in Vietnam, women with high FVS and DDS had significantly higher mean energy intakes than those with lower scores (Ogle et al., 2001). Dietary diversity indicators could be useful to assess the adequacy of energy intake at population levels.

Research reports that shows dietary diversity score as an indicator for diet quality and micronutrients intake in developing countries are summarized in **Table 2.** All the studies in table 2 generally measure the quality of diet that is consumed. In these studies, a variety of dietary diversity measures were used based on different food and food group classification systems, different numbers of foods or food groups, and varying reference period lengths (Ruel, 2003b). Cutoff points of DDS to define varying levels of dietary quality have to be defined in the context where they are used, taking into account local food systems and dietary patterns (Ruel, 2003c). In using dietary diversity to predict diet quality and/or disease outcomes, use of diverse classification of dietary diversity is recommended. Interestingly however none of these studies predicted functional biomarkers which are the ultimate measures of nutrient status in validating an individual's nutritional status.

In calculating dietary diversity scores, cut off points or units of portion sizes to be used in computation should also be carefully considered as appropriate to various locations and cultural practices. In such developed countries as United States and Europe, computation of DDS begins with clear definition of inclusion and exclusion criteria (Ruel, 2002). Ruel (2002) reported an error in the inclusion of fish in food group diversity score computation of preschoolers in northern Ghana where an apparent high fish intake was not a true reflection of fish intake in terms of quantity. Further investigation identified the consumption as minute amounts of fish powder added to porridges as a condiment. Similarly, inaccurate DDS resulted when very small amounts in the form of condensed sweetened milk added to hot beverages by young children in Accra was counted towards intake of dairy food group. Prior knowledge of dietary patterns in populations of interest is vital for determining foods susceptible to this type of errors (Ruel, 2002).

Table 2.Dietary diversity score as a measure of diet quality

Author	DDS description	Subject Description	Selected nutrients	Conclusion
Korkalo et al., 2016[1]	9 groups – 24hr recall 9 groups (>15g cutoff) – 24hr recall 9 groups – 7 days recall	Cross-sectional study of 227 adolescent girls (14-17 yrs) in Mozambique	Iron, zinc, folate and vit A	DDS is associated with serum zinc in hunger season only
Kennedy et al., 2007[2]	10 groups (no cutoff) – 24hr recall 10 groups (>1g & >10g cut off) – 24hr recall	Cross-sectional study of 3164 children 24–71 months in Philippine	Energy, protein, fat, calcium, iron, vit A, vit C, thiamin, riboflavin, niacin, vit B-6, vit B-12, folate, and zinc	Both DDS 1g and DDS 10g were predictors of adequate micronutrient intake (p<0.05)
Moursi et al., 2008[3]	8 groups (no cutoff) – 24hr recall 8 groups (>1g & >10g cut off) – 24hr recall	Cross-sectional study of 3164 children 6 -23 months in Madagascar	Energy, protein, calcium, iron, vitamin A, vitamin C, thiamin, riboflavin, vitamin B-6, vitamin B-12, and zinc	DDS useful proxy of micronutrient density of foods consumed by breastfed (BF) and non-BF infants
Arimond et al., 2010[4]	6, 9, 13, or 21 groups (>1g & >15g cut off) – 24hr recall	Cross-sectional study of 2078, 2024, 2086, 2083, 1211 women inBurkina Faso, Mali, Mozambique, Bangladesh, and the Philippines, respectively	Vit A, thiamin, riboflavin, niacin, vit B-6, folate, vit B-12, vit C, calcium, iron, and zinc.	Simple food group diversity indicators hold promise as proxy indicators of micronutrient adequacy

FVS: Food variety score. DDS: Dietary diversity score.

Title of papers

[1] Associations of dietary diversity scores and micronutrient status in adolescent Mozambican girls
[2] Dietary Diversity Score Is a Useful Indicator of Micronutrient Intake in Non-Breast-Feeding Filipino Children
[3] Dietary Diversity Is a Good Predictor of the Micronutrient Density of the Diet of 6- to 23-Month-Old Children in Madagascar
[4] Simple Food Group Diversity Indicators Predict Micronutrient Adequacy of Women's Diets in 5 Diverse, Resource-Poor Settings

Table 2 (cont'd)

Author	DDS description	Subject Description	Selected nutrients	Conclusion
Steyn et al., 2006[5]	9 groups (no cutoff) – 24hr recall 45 food items (no cutoff) – 24hr recall	Cross-sectional study of 2200 children (1-8 yrs) in South Africa	Vit A, B1, B2,B-6, B-12 and C, niacin, , folate, calcium, iron and zinc	FVS or DDS are used as a simple and quick indicator of the micronutrient adequacy.
Mirmiran et al., 2004[6]	23 groups (1/2 serving cut off) – 24hr recall	Cross-sectional study of 304 adolescents (10-18 yrs.) in Teheran	Vit A, B1, B2 and C calcium, iron, zinc, phosphorus, magnesium, protein, potassium, fat, carbohydrate	DDS is an appropriate method to evaluate nutrient intake adequacy in adolescents
Bukania et al., 2014[7]	9 groups (no cutoff) – 24hr recall	Cross-sectional study of 277 women (15-49 yrs) and children (6-36 mn) pair in Kenya	Macronutrients - protein and carbohydrates	DDS is highly correlated with caloric and protein adequacy. Differences in agroecological zones may not affect DDS and nutritional status of farmer households. DDS may underestimate food insecurity in semiarid settings.

FVS: Food variety score. DDS: Dietary diversity score.

Title of papers

[5] Food variety and dietary diversity scores in children: are they good indicators of dietary adequacy?
[6] Dietary diversity score in adolescents - a good indicator of the nutritional adequacy of diets: Tehran lipid and glucose study
[7] Food Insecurity and Not Dietary Diversity Is a Predictor of Nutrition Status in Children within Semiarid Agro-Ecological Zones in Eastern Kenya

2.4 Dietary approaches to improve trace mineral status

Globally, trace mineral deficiencies are of great concern with the most widespread deficiencies being iron, zinc, and iodine (Gayer and Smith, 2015). Several approaches have been adopted to counteract these deficiencies including food fortification, supplementation and dietary diversity approaches. The key challenges with the former two approaches are limited sustainability due to sociocultural acceptability, affordability and availability throughout the rural and urban areas in developing countries (Yip and Ramakrishnan, 2002) where transportation and population density vary widely within natural ecological system (Linard et al., 2012). Dietary approaches such as food-to-food fortification without incorporation of elemental iron can be promising to improve trace minerals intake (Cercamondi et al., 2014) especially in developing countries by overcoming the limitations of other approaches. The fundamental challenge of food-to-food fortification is establishing the efficacy of food-to-food fortification in improving trace mineral status. Little data exist that were based on indigenous foods in Sub-Saharan African countries including Ghana.

Below (table 3) gives detailed summary of various studies that used dietary approaches to improve micronutrient status. Predictably, these studies relied heavily on fortification or supplementation. However, food-to-food fortification without incorporation of elemental iron approach is rarely used though it may be a good alternative in real world situations. Food-to-food fortification on its own is not without challenges, it comes with the challenge of having to balance dietary iron content and iron bioavailability. Plant based meals have low iron bioavailability whereas in the fortification of food with elemental iron (≥12 mg fe in all the under listed studies), bioavailability of iron is very high. To enhance iron bioavailability in food-to-food fortification, there is a need to have a mixture of plant and animal based food or a mixture of plant and bioavailability enhancers (Vitamin C, fructose, citric acid, dietary protein, lysine, histidine etc) as demonstrated in the studies below (table 3).

Table 3. Dietary approach interventions to improve nutrients intake

Author	Title	Subject Description	Selected nutrients	Conclusion
Egbi et al., 2015[1]	3 groups fed with 3 meals: Group 1: cowpea-based food + 3% fish powder + 33 mg vit C/100 mL Group 2: cowpea-based food + 33 mg vit C/100 mL Group 3: cowpea-based food + placebo	6-month nutrition intervention study among 162 children (6-12 yrs) in Ghana	Iron	Cowpea- based food containing 3% fish meal and served with vit C–rich drink (group 1) improved hemoglobin concentration and minimized the prevalence of anemia
Tetens et al., 2007[2]	2 groups received meat or vegetable to take home (biweekly). Iron contents similar in both groups. Compliance measured by quantity of foods returned. Group 1: meat (animal based diet) Group 2: vegetables (plant based diet)	A stratified random treatment of 57 women (19-39 yrs) for 10-20 wks in Denmark	Iron	Serum ferritin remained unchanged in women on the meat-based diet but decline for women on the vegetable-based diet. Concluded that It's important to ensure a balance between dietary iron content and iron bioavailability for the maintenance of blood indicators of iron stores in women with initially low iron status
Parker et al., 2015[3]	2 groups given dry rations: Group1: Fortified rice (iron, zinc, vit B1 & folic acid) Group2: Rice + Beans (vit A & iodized salt)	Cluster-randomized feeding trial in 994 children (7-11 yrs) for 7 months in Burundi	Iron	No significant difference in Hb concentration between the intervention (group 1) and control (group 2)
Christofides et al., 2006[4]	Multi-micronutrient sprinkles treatment for 3 groups (Nutrients: Fe, Zn, folic acid, vit A & D) Group1: 12.5 mg of iron Group2: 20 mg of iron Group2: 30 mg of iron	A randomized clinical trial for 8 weeks of 133 anemic children (6-18 moths) in Ghana	Iron	Low doses of sprinkles (12.5 mg) may be effective in reducing anemia children

Titles of papers

[1]Impact of Cowpea-Based Food Containing Fish Meal Served With Vitamin C–Rich Drink on Iron Stores and Hemoglobin Concentrations in Ghanaian Schoolchildren in a Malaria Endemic Area

[2] The impact of a meat- versus a vegetable-based diet on iron status in women of childbearing age with small iron stores

[3] A Blinded, Cluster-Randomized, Placebo-Controlled School FeedingTrial in Burundi Using Rice Fortified With Iron, Zinc,Thiamine, and Folic Acid

[4] Multi-micronutrient Sprinkles including a low dose of iron provided as microencapsulated ferrous fumarate improves haematologic indices in anaemic children: a randomized clinical trial.

Table 3 (cont'd)

Author	Title	Subject Description	Selected nutrients	Conclusion
Cercamondi et al., 2014[4]	3 test meals 1. Maize-based t ô + amaranth sauce, 2. Maize-based t ô + iron-improved amaranth sauce (increased quantities) 3. Maize-based t ô + iron-improved jute sauce (increased quantities)	A randomized crossover design with multiple meals was used with each woman (18-40 yrs., n=18) serving as her own control for 23 days in Burkina Faso	Iron	A food-to-food fortification approach based on an increase in leafy vegetables quantity does not provide additional bioavailable iron, presumably due to the high phenolic compound concentration of the leaves tested. No difference between groups.
Abizari et al., 2012[6]	2 groups fed with 2 meals: Group1: Fortified cowpea-based food with NaFeEDTA Group2: Unfortified cowpea-based food	Randomized, double-blind, controlled trial. 241 children (2 -12 years) feed with tubani (3 d/wk) in Ghana	Iron	consumption of fortified cowpea meal significantly improved both functional (transferrin receptor) and storage (ferritin) iron status
Zhao et al., 1999[7]	3 groups, salt given at household level: Group1: 25 ppm iodize salt every 2 months Group2: purchased iodize salt every 2 months Group3: 4 capsules of iodized oil (100 mg iodine each) at baseline + purchased iodize salt every 2 months	Randomized clinical trial involving 145 children "8-10 yrs" for 6,9,12,& 18 months in China	Iodine	The median urinary Iodine improved at the 6-month follow-up. Children with initially a low to moderate level of iodine deficiency were not iodine deficient on all indicators after 18 months of study.

Titles of papers

[4] A Higher Proportion of Iron-Rich Leafy Vegetables in a Typical Burkinabe Maize Meal Does Not Increase the Amount of Iron Absorbed in Young Women

[6] Whole Cowpea Meal Fortified with NaFeEDTA Reduces Iron Deficiency among Ghanaian School Children in a Malaria Endemic Area

[7] Randomized clinical trial comparing different iodine interventions in school children

2.5 Dietary diversity score and dietary pattern as predictors or indicators of disease outcomes

In previous studies, DDS reflects micronutrient adequacy of the diet and is associated with better child growth as well as health outcome (Amugsi et al., 2014; Nguyen et al., 2013). In computing DDS, Nguyen et al (2013) used seven food groups from a 24-hr recall for subjects in Bangladesh, Vietnam, and Ethiopia while Amugsi et al (2014) used 16 food groups in Ghana. A study in Cambodia indicates that consumption of a diverse diet (FVS, 9 food items–DHS dataset) among children (12-59 mo) 12-19 mo was associated with a reduction in rate of stunting (Darapheak et al., 2013). Similarly, in Ghana, DDS (16 food groups, 24 hr recall, DHS) was inversely associated with wasting among rural children with coefficient of -0.11, but not urban children, after controlling for child, maternal, and household characteristics (Amugsi et al., 2014).

In Kenyan HIV patients, DDS (9 food groups, 24hr recall) was associated with health-related quality of life (mental, physical and general health scores). DDS was thus considered as part of comprehensive interventions designed to mitigate psychosocial consequences of HIV (Palermo et al., 2013). This study however failed to include one's household characteristics as an immediate environment that might also contribute to disease outcome despite the fact that DDS is known to be associated with one's household characteristics.

Dietary pattern (DP) on the other hand is multiple dietary components operationalized as a single exposure in relation to an outcome (nutrient adequacy, health status, lifestyle and demographic variables) (Kant, 2004). It is defined as the quantities, proportions, variety or combinations of different foods and beverages in diets, and the frequency with which they are habitually consumed (Kant, 2004).

DP has been suggested as an approach to predict the association between diet and development of diseases (Hoffmann et al., 2004) or health conditions associated with dietary habits. The rationale of the suggestion is that foods are consumed in combination making it difficult to separate the effects of single foods on the development of diseases in observational studies (Hoffmann et al., 2004; National Research Council, 1989).

Dietary patterns have been derived by various approaches to validate their associations with development of diseases (Fung et al., 2001; Hauta-alus et al., 2017; Hu et al., 2000; Panagiotakos et al., 2006; Shin et al., 2015). Commonly dietary patterns have been derived by reduced rank regression (Shin et al., 2015), dietary intake by index analysis (Tobias et al., 2012), factor analysis (Zhang et al., 2006) or individual nutrient intakes (Bao et al., 2014). In Ghana, few studies have investigated dietary patterns and DP association with disease (Frank et al., 2014; Jannasch et al., 2017) or nutrient deficiency. To our best knowledge, to date no Ghanaian diets have been investigated for their associations with the biomarkers of the problematic nutrients, for instance iron and iodine.

2.6 Household food production to improve nutritional status

Agricultural production influences the quality of diets of smallholder farming households through consumption of subsistence food crops and animals directly at the household level, or through the sale of agricultural goods for household incomes which affects food purchases and consumption (Jones et al., 2014). Other pathways that affect the nutritional status of individuals within households are through (1) household income spent to purchase healthcare for individuals, (2) a woman's time and workload that affect her energy expenditure, health and capacity to feed and care for young children, and (3) a woman's control of household income and expenditure. These are affected in part by her ownership of farm output, the kind of income generated from that output, the kinds of purchases made with the income and the allocation of resources within households (Jones et al., 2014).

The fundamental model of household agriculture production in improving family nutritional status stems from the basis that agriculture production leads to increased availability of nutritious foods which improves household's consumption patterns. These consumption patterns then lead to improved health and nutritional status which in turn improve family wellbeing and the agricultural production. This cycle is based on the condition that the consumed food is of quality (diverse diet) and the body is able to utilize the foods. It is on this basis that a proposition is made to explore alternative, non-traditional routes to producing nutrient-rich vegetables during the dry season in Ghana.

Chapter Three: Dietary approaches to mitigate iron and iodine deficiencies of mother-child dyads during dry/lean season in northern Ghana

3.1 Abstract

Iron and iodine deficiencies are among the most common form of nutritional deficiencies globally. Iron and iodine deficiencies lead to detrimental effects on fetus, infant, and maternal health. Although the causes are multifaceted, attention has mainly been on fortification, supplementation, clinical examination, and treatment though nutrients deficiencies can be prevented through screening and intervention with dietary approaches in developing countries. We aimed to investigate i) if dietary indicators or identify dietary indicators (dietary diversity score - DDS, dietary patterns - DP) that predict the risk of iron and iodine deficiencies in women (15-49 yr, n=118) and their toddlers (6-23 mo, n=121), and ii) the relationship between mother-toddler health indicators in a cross sectional-study during lean/dry season in northern Ghana. We found that risk of iron and iodine deficiencies can be predicted by DDS and DP. DDS<4 have higher odds of iron deficiency (AOR: 3.21; 95% CI: 1.02-10.09; p=0.0466) and iodine deficiency (AOR: 0.11; 95% CI: 0.02-0.76; p=0.0360). Of two dietary patterns: DP 1 ("rural elites diet") and DP 2 ("indigenous diet") emerged, *rural elites diet* significantly predicted iron deficiency risk (AOR: 8.71, 95% CI: 1.79-42.31; p=0.0265 and AOR: 8.65, 95% CI: 1.76-42.39; p=0.0287) while *indigenous diet* predicted iodine deficiency risk (AOR: 9.41; 95% CI: 1.29-68.43; p=0.0400 and AOR: 11.41; 95% CI: 1.36-95.97; p=0.0406) in women respectively but not in toddlers. The malaria status of mothers and their toddlers significantly correlated (r=0.20, p=0.0315) while iron status (stfr, Hb,

anemia, and IDA) and iodine status (UIC) of mothers and toddlers had insignificant correlation.

3.2 Introduction

Iron and iodine deficiencies are detrimental to maternal health and lead to fetal and infant growth retardation, stunting and impaired cognitive development (Bath et al., 2013; Gayer and Smith, 2015; Stewart et al., 2013). Globally, deficiencies of such micronutrients as iron and iodine are common in women of reproductive age and children due to increased needs and vulnerability of such groups (Centers for Disease Control Prevention, 2002; Gayer and Smith, 2015; Stewart et al., 2013; Stoltzfus, 2003). In Ghana, iron deficiency, iron deficiency anemia, iodine deficiency, and anemia prevalence are highest among women of reproductive age and children (Abizari et al., 2012; Ewusie et al., 2014; GSS, 2009; 2011; 2014; Zimmermann and Hurrell, 2007). The rate of anemia and malnutrition prevalence is highest in northern Ghana which is made up of Upper East region, Upper West region and Northern region, compared to the remaining seven regions of Ghana (GSS, 2009; 2011; 2014; WFP, 2015). The disparity is evidenced by the rate of anemia at 82.1% in Northern region, 73.8% in Upper East and Upper West Regions versus 53.7% in Ashanti region (GSS, 2014). Iodine deficiency is estimated by household use of iodized salt in countries where iodized salt is the main source of iodine for which Ghana is no exception. In 2014, about 38.5% of households reported using adequately iodized salt. This percentage of household iodized salt use dwindles from Western region (60.3%), Accra (54.9%), Ashanti (46%), Brong Ahafo (35%) to Northern (16%) and Upper East (18%) regions in Ghana. Urban areas are more likely to use adequately iodized salt (50%) than rural areas (26%) (Ghana Statistical Service et al., 2015). This suggest high prevalence of inadequate iodine intake may exist in Northern and the Upper regions of Ghana. Recent data shows prevalence of iodine deficiency as 36% and 29% for women (15-49yrs) and children (6-24mo) respectively (Kubuga, unpublished).

Among multifaceted causes of such micronutrient deficiencies as iron and iodine, dietary intake plays the key role (Andersson et al., 2012; Humphries et al., 2011; Lynch, 2011) thus micronutrient deficiencies can be effectively assessed, screened and intervened by non-invasive dietary intake approaches, compared to costly and laborious biomarkers or non-specific clinical examinations. Currently Ghana has no dietary assessment tools that are validated by biomarkers. Most importantly, due to lack of food composition tables of indigenous foods, the possibility of using the interplay of mother-child health indicators (iron, iodine, and malaria) to reduce deficiencies has not been fully explored. Knowing the status of either a mother or a child may inform or predict the status of the other thus contribute to prevention/treatment. The aim of this study was to use dietary approaches to predict iron and iodine status and assess the relationship between mother-child dyad health indicators. Studies among women of child bearing age are vital with respect to malaria, iron, and iodine indicators due to the biological roles iron and iodine play in the life cycle (Malhotra et al., 2014; Radlowski and Johnson, 2013; Schmidt et al., 2014; Valera-Gran et al., 2014).

3.3 Subjects and methods

This cross-sectional study is drawn from a community-based feeding trial (detailed methods published elsewhere). Data were collected from women 15-49 yrs, and their toddlers (6-23 months) sampled from five communities in two districts in the

Upper East region (Kassena Nankana West and Builsa North Districts) between May-June 2016.

Dyads were identified using community-based birth registers at Community-Based Health Planning and Services (CHPS) compounds kept by community health nurses/volunteers. Announcements were then made through the community chiefs/leaders in the respective communities for all women with children younger than 5 years of age to meet at their respective community health centers. Research team briefed the women and the community leaders on the study. Names that were shortlisted from the birth registers and other sources were read out. Dyads were contacted individually to check on their willingness to be part of the study through the community health volunteers and community health nurses. Verbal consent was sought from spouses of women who were willing to be part of the study. A total of 120 dyads were drawn from the two districts. Two dyads dropped out from one district before data collection.

3.3.1 Measurements and data collection

Malaria testing: Rapid malaria diagnostic cassettes (Lot: 05CDB050DA. Standard Diagnostic, Inc. Republic of Korea.) were used to test for malaria status for all research participants. Malaria status was assessed based on the presence of Histidine-Rich Protein-2 (HRP-II) in whole blood. HRP-II is known to be specific to *Plasmodium falciparum*, which causes more than 90% of malaria cases in Ghana (Asante et al., 2011). Sensitivity and specificity of cassettes were 95% and 99.5% respectively. Malaria status was tested as it could be a confounder in predicting iron status. All participants who tested positive for malaria were referred to their respective CHPS compound for treatment

Anthropometry: Participant weights and heights were measured according to standard procedures (Cogill, 2003). Electronic scale (Serial number 5874030154862, Model 874 1321009. Seca gmbh & co kg, 22089 Hamburg, Germany.) was used to measure weights to the nearest 0.1 kg. Scale was calibrated using a known weight on days when measurements were taken.

Questionnaire/interview: Questionnaire on food intake frequency and 24hr food intake recall was administered for all participants. Information on sociodemographic characteristics was also taken.

Biochemical measurements: 5 ml of whole blood was drawn into silica-coated serum separator vacutainers (Lot: 20140618, Anhui Kangning Industrial (Group) Co. Ltd, Tianchang City, China) and held at ambient temperature both on the field and during transportation. Serum was separated using a centrifuge (Hettich) at 500 × *g* for 5 min at room temperature. Separated serum was aliquoted, kept frozen at −18°C (Thermo Fisher Scientific) at NHRC, and then transported on dry ice to MDS Lancet laboratories- South Africa for serum transferrin receptor analysis. Hemoglobin levels were checked in the field using HB 201 analyzer (componay, location) according to prescribed procedures. Anemic individuals were referred for treatment. Content of iodine (o ppm, <15 ppm, and ≥15 ppm) in household salt samples provided by women were tested using validated Rapid Test Kits (Batch No: M 050, MBI KITS International, India) according to prescribed procedures. Change of color that indicates iodine concentration was compared with the RTK charts and recorded.

3.4 Statistical analysis

Data analysis has been done using SAS 9.4 (SAS institute). Characteristics of participants were described using frequency distributions. Dietary diversity score (0-9) was calculated from 24 hr intake according to the Food and Agriculture Organization (FAO). A score of 1 was assigned for consumption of a food group and a score of 0 for non-consumption for the food group. The nine food groups per FAO's classification are starch staples; dark green leafy vegetables; other vitamin A rich fruits and vegetables; other fruits and vegetables; organ meat; meat and fish; eggs; legumes; nuts and seeds; milk and milk products.

Dietary patterns of the women were identified by subjecting food items to factor analysis using principal component analysis (PCA) as the extraction procedure. Principal components are linear combinations of the input variables and explain the variation in the data. Each component describes a dietary pattern and the linear combination gives room for the calculation of a component score for each woman/child. In determining the number of factors for retention, multiple methods were used (parallel analysis, visual scree test and literature search). Varimax rotation was used to help in the interpretation of components. Factor adequacy was established *a priori*, pattern coefficients ≥ 0.40 were considered salient and practically significant. Complex loadings which were salient on more than one factor were rejected in the interest of parsimony and to be in consonance with simple structure. Singletons were also rejected. Factors with at least 3 salient pattern coefficients, internal consistency reliability ≥0.70 were considered adequate. Internal reliability was checked by using factors loadings at Cochran's alpha level ≥0.60.

Multivariable logistic regression models were used to examine the relationship between DDS (DDS<4 & DDS≥4)/dietary pattern tertiles and the risk of iron and iodine deficiencies. Independent variables of interest were iron status (ID: stfr>4.40 ug/l for women, MDS Lancet lab reference; Anemia: Hb<12 g/dl; IDA: concurrent presence of ID and anemia) and iodine deficiency (UIC<50 ug/l). Pearson correlation and kappa analysis were done for mother-child dyads inter and intra relationships of iodine and iron status.

E*thics:* Research procedures were in accordance with the Michigan State University's Institutional Review Board and Navrongo Health Research Centre (NHRC) Institutional Review Board (IRB) in Ghana.

3.5 Results

Sociodemographic characteristics and health indicators (weight, height, serum transferrin receptor level, hemoglobin level, iodine status, malaria status, anemia and iron status) of child-bearing age women and their children are summarized in Table 4. Summary of DDS distribution among mother-child dyads in table 5 indicates a score of 0-6 and 0-5 out of a total score of 9 for women and children respectively. In table 6, we compared dyads whose DDS was less than four to those that had DDS of four or more in non-adjusted (model 1) and adjusted model (model 2). The adjusted model shows significant risk of iron deficiency (AOR: 3.205; 95% CI: 1.02-10.09; p-value: 0.0466) and iodine deficiency (AOR: 0.11; 95% CI: 0.02-0.76; p-value: 0.0360 and AOR: 6.66; 95% CI:1.35-32.81; p-value:0.0199).

Acknowledging the effect of malaria, we adjusted for the stated covariates plus malaria (model 1) and without malaria (model 2). Significant (p=0.0466) and marginal significant (p=0.0526) risk of iron deficiency in women with DDS<4 in model 1 and model 2 respectively. Details of results not shown here. In a similar comparison for iodine deficiency, mothers with DDS<4 were at a higher risk of iodine deficiency (AOR:5.32, p=0.0215) as compared to those with DDS≥4. Poor participation in household's purchasing decision making and large household size (>4 vs. ≤4 adults) increased the risk of having iodine deficiency (UIC<50ug/l). DDS<4 showed no significant risk of neither iron nor iodine deficiency in children (results not shown).

Two and one dietary pattern(s) were identified (table 7) by factor analysis with 17 food items or groups among women and their toddlers respectively. These patterns were named according to food group factor loadings: “rural elites diet” and “indigenous diet”. The “rural elites diet” pattern was characterized by a high intake of tea, sugar, milk creamers, and fish condiments while the indigenous diet was characterized by dark green vegetables, grains, legumes, and wild fruits. The children had one dietary pattern (indigenous diet).

We compared the risk of iron and iodine deficiency across women dietary patterns in three models (Table 8): Crude comparison (model 1), adjusted for household wealth index, BMI, household iodized salt usage, woman's age, number of adults and children in household (model 2), and model 3: Model 2 + malaria. Models 2 and 3 indicated significant higher risk of iron deficiency (AOR: 8.71, 95% CI: 1.79-42.31; p=0.0265 and AOR: 8.65, 95% CI: 1.76-42.39; p=0.0287) in “rural elites diet” and iodine deficiency (AOR: 9.41; 95% CI: 1.29-68.43; p=0.0400 and AOR: 11.41; 95% CI: 1.36-95.97; p=0.0406) in “indigenous diet”. Women in the middle tertile for “rural elites diet” had significantly greater odds of being iron deficient. Risk of iodine deficiency was significantly higher in the lower tertile for “indigenous diet”. There is no significant risk relationship between children dietary pattern tertiles and children iron and iodine status.

Correlation and kappa analysis of mother and child health indicators was done, results tables not shown here. Significant relationships are highlighted here. Mother stfr inversely correlates with mother Hb (r=-0.47, p=<0.0001), IDA (r=-0.62, p=<0.0001), and anemia (r=-0.20, p=0.0324), which signifies that as stfr declines (improved iron status), Hb increases and the proportion of mothers with anemia and IDA declines. Mothers Hb positively correlates with mothers IDA (r=0.20, p=0.0316), and anemia (r=0.76, p=0.0324), which signifies improvement in Hb improves proportion of mothers with adequate iron and Hb status (Hb≥12 g/dl). Mothers UIC positively correlates with mothers’ iodine deficiency (r=0.57, p=<0.0001) signifying improvement in UIC improves proportion of mothers with adequate iodine (UIC≥50ug/l). There was disagreement/poor agreement (k≤0.0) in intra comparison of iron and iodine indicators. This may suggest absence of coexistence of iron and iodine deficiencies. Disagreement between IDA and anemia may suggest that greater proportion of mothers with anemia may not necessarily be caused by iron deficiency as evidenced by the large percentage difference between IDA and anemia.

On the part of children, similar observation as in their mothers was made. stfr inversely correlates with Hb (r=-0.34, p=0.0002), IDA (r=-0.29, p=<0.0011), and anemia (r=-0.20, p=0.0324), which signifies that as stfr declines (improve iron status), Hb increases, proportion of children with anemia and IDA declines. Hb positively correlates with anemia (r=0.60, p=<0.0001), which signifies increase in Hb improves proportion of toddlers with adequate Hb status. Similarly, as urinary iodine concentration increases, proportion of children with UIC≥50ug/l also increases ($r=0.52$, $p=<0.0001$). disagreement/poor agreement ($k\leq0.0$) in intra comparison of iron and iodine indicators was also observed in children.

Mother-child dyads biomarker inter relationship shows an inverse and positive relationship between mother stfr vs. child Hb ($r=-0.24$, $p=<0.0106$), and mother stfr vs. child IDA ($r=0.21$, $p=<0.0207$) respectively. As mother stfr declines (improved iron status), child Hb increases (improve Hb levels). As stfr increases, proportion of children with adequate iron status increases showing similar relationship as in the intra mother-child biomarker relationship. Mothers IDA vs. child stfr shows an inverse correlation ($r=-0.18$, $p=0.0485$) suggesting that as the proportion of mothers with adequate iron status declines, children stfr increases (poor iron status). Mother iodine status was inversely related with child anemia status ($r=-0.26$, $p=<0.0352$), this may signify high prevalence of IDA as compared to iodine deficiency in children. Mother and child malaria status was positively correlated ($r=0.20$, $p=<0.0315$) suggesting either mother or child malaria status may be used as a modest proxy for the other. The kappa value ($k=0.20$) further suggest the fair agreement of this proposed proxy. Only malaria status of mother-child dyads significantly correlates and gives a modest kappa agreement suggesting either mother or child malaria status may be used as a crude proxy for the other.

Table 4. Demographic characteristics and health indicators of child-bearing age women (15-49 yrs) and their children (6-23mo) during dry/lean season in northern Ghana.

Variable		n	Mean ± SD
Women age (yrs)		118	26.4±6.0
Women height (cm)		118	161.0±6.1
Women weight (kg)		118	57.2±8.6
Women Hb (g/dl)		118	11.8±1.3
Women sTfr (ug/l)		117	4.9±2.4
Household head's age		118	44.8±16.1
Number of household Adults		118	4.1±2.5
HAZ		121	0.01±1.4
WHZ		121	-1.1±1.3
children Hb (g/dl)		121	9.2±1.5
Children sTfr (ug/l)		120	9.5±5.2
Children age (months)		121	13.3±5.2
# Siblings in household		121	1.1±1.6
# children < 5 yrs		118	1.1±1.2
		n	%
Occupation of women	Farmer	82	69
	Handy works	10	8
	Housewife	6	5
	Trader	20	17
Household head sex	Male	110	93.2
	Female	8	6.8
Wealth index	Lower	39	33.0
	Middle	40	34.0
	Upper	39	33.0
Decision making	Low	20	17.0
	Average	61	51.7
	Good	37	31.3
Household iodized salt**	Non-users	105	89.7
	Users	12	10.3
Iodine level in household salt	<15ppm	112	95.7
	≥15ppm	5	4.3
Ethnicity	Builsa	55	47
	Kassena	60	51
	Nakani/Fulani	3	3
Religion	Christian	91	77
	Muslim	9	8
	*Traditionalist	18	15

*Traditionalist: Practitioners of African Traditional Religion; Decision making: women participation in household decision making. Hb: Hemoglobin, Stfr: Serum transferrin receptor, UIC: Urinary iodine concentration. Median UIC<100 ug/l indicates iodine deficiency in a population. **Non-users of iodized salt: iodine content of household salt is 0 ppm.

Table 4 (cont'd)

Variable	Status	n	%
women UIC (ug/l)	deficient	24	36.4
	normal	42	63.6
women ID-stFr(ug/l)	deficient	56	47.9
	normal	61	52.1
women IDA	anemic	31	26.5
	normal	86	73.5
Women general anemia–Hb(g/dl)	anemic	62	52.5
	normal	56	47.5
Women malaria	Positive	12	10.2
	Negative	106	89.8
Children UIC (ug/l)	deficient	19	28.8
	normal	47	71.2
Children ID-stFr(ug/l)	deficient	117	97.5
	normal	3	2.5
Children IDA	anemic	106	88.3
	normal	14	11.7
Children general anemia-Hb (g/dl)	anemic	110	90.9
	normal	11	9.1
Children malaria	Positive	110	91.7
	Negative	10	8.3
Stunting (HAZ)	Stunted	10	8.3
	Normal	111	89.7
		n	Median
Women median UIC (ug/l)		66	62.2
Children median UIC (ug/l)		66	124.7

*Traditionalist: Practitioners of African Traditional Religion; Decision making: women participation in household decision making. Hb: Hemoglobin, Stfr: Serum transferrin receptor, UIC: Urinary iodine concentration, ID: iron deficiency (women - stfr>4.40ug/l, children - stfr>2.85ug/l), Anemia (women - Hb<12g/dl, children - Hb<11g/dl); Iodine deficiency defined (UIC <50ug/l); IDA: Concurrent presence of anemia and IDA. Median UIC<100 ug/l indicates iodine deficiency in a population.

Table 5. Dietary diversity score distribution

Dietary diversity score (DDS)	Women DDS9 n (%)	Women DDS9G n(%)	Children DDS9 n (%)	Children DDS9G n(%)
0	1 (0.9)	1 (0.9)	29 (24.0)	29 (0.9)
1	1 (0.9)	1 (0.9)	7 (5.8)	7 (0.9)
2	2 (1.7)	13 (11.0)	2 (1.7)	13 (11.0)
3	33 (28.0)	49 (41.5)	28 (23.1)	34 (41.5)
4	33 (28.0)	53 (44.9)	22 (18.2)	38 (44.9)
5	45 (38.1)	1 (0.9)	33 (27.3)	-
6	3 (2.5)	-		
Mean±SD	4±1	3±0.8	2.9±1.9	2.4±1.6

DDS: Dietary diversity score. DDS9: Score based on FAO 9 food group classification for women, DDS9G: Fish, milk & milk products excluded in scores calculation per FAO recommendations on food group inclusion based on quantities. DDS 0 represent an individual none consumption of any of the food groups in previous 24hrs (this is not unusual in the lean season). DDS 0 for children meant they only had breastmilk in the previous 24hrs.

FAO food groups: Starch staples; dark green leafy vegetables; Other vitamin A rich fruits and vegetables; Other fruits and vegetables; Organ Meat; meat and fish; eggs; Legumes; nuts and seeds; milk and milk products.

Table 6. The risk of anemia, iron deficiency, and iodine deficiency by adequacy of dietary diversity score (24 hr recall) among women of child bearing age

Dietary diversity score (DDS)	Iron deficiency (stfr>4.4ug/l)		Iodine deficiency (UIC<50ug/l)	
	Model 1 OR (95% CI)	Model 2 OR (95% CI)	Model 1 OR (95% CI)	Model 2 OR (95% CI)
DDS9G<4	0.72 (0.35-1.50)	3.21 (1.02-10.09)*	0.41 (0.14-1.20)	0.11 (0.02-0.76)*
DDS9G≥4	Ref	Ref	Ref	Ref
P-value	0.3793	0.0466	0.1034	0.0360
DDS9<4	0.96 (0.44-2.08)	1.27 (0.39-4.11)	0.42 (0.15-1.21)	6.66 (1.35-32.81)*
DDS9≥4	Ref	Ref	Ref	Ref
P-value	0.9079	0.6959	0.1072	0.0199

*P<0.05, OR: Odds ratio, Hb: Hemoglobin, stfr: Serum transferrin receptor, DDS9: Score based on FAO 9 food group classification for women, DDS9G: Fish, milk & milk products excluded in scores calculation per FAO recommendations on food group inclusion based on quantities. ^Dropped BMI &, household iodized salt usage had counts<5. **Model 1:** Crude association between anemia (Hb<12.g/dl, n=118; stfr>4.4ug/l, n=117), iodine deficiency (UIC<50ug/l) and Dietary Diversity score. **Model 2:** Adjusted for household wealth index, BMI, household iodized salt usage, woman's age, number of adults and children in household, involvement in household decision making

Table 7.Rotated Factor Pattern (24 hr Dietary Recall) among child bearing age women (15-49yrs) and their toddlers

Food item	**Women**		**Toddlers#**
	Factor 1 (“Rural Elites diet”)	**Factor 2** (“indigenous diet”)	**Factor 1** (“indigenous diet”)
Dark green vegetables	-17	45*	64*
Grains	-4	76*	84*
Legumes	-8	81*	91*
Eggs	-4	13	11
Fat and oils	30	-13	46*
Powdered fish	42*	38	78*
Other liquids	3	9	16
Mango and pawpaw	23	-12	11
Any meat	0	0	0
Milk	73*	7	0
Organ meat	0	0	0
Other diary	0	0	0
Other fruits and vegetables	31	44*	73*
Roots	-16	8	9
Sugar	56*	6	-8
Tea	78*	3	0
Yellow vegetables	-16	-4	0
Variance explained	1.97	1.84	3.37

Printed values are multiplied by 100 and rounded to the nearest integer. Values n >40 are flagged by an ''. Factor pattern not rotated as factor is only one#*

Table 8. The risk of anemia and iodine deficiency across dietary pattern scores tertiles (24hr recall) among women of child bearing age

	Iron deficiency (stfr>4.4ug/l)			Iodine deficiency (UIC<50ug/l)		
	Model 1	Model 2	Model 3	Model 1	Model 2	Model 3
			Rural Elites diet			
Tertile	OR (95% CI)	OR (95% CI)	OR (95% CI)	OR (95% CI)	OR (95% CI)	OR (95% CI)
Tertile 1	0.86 (0.35-2.09)	2.33 (0.56-9.66)	2.47 (0.58-10.50)	0.86 (0.26-2.79)	1.33 (0.24-7.38)	1.31 (0.24-7.33)
Tertile 2	0.78 (0.32-1.88)	8.71 (1.79-42.31)*	8.65 (1.76-42.39)*	0.69 (0.20-2.43)	0.50 (0.08-3.13)	0.50 (0.08-3.14)
Tertile 3	Ref	Ref	Ref	Ref	Ref	Ref
P-trend	0.8533	**0.0265**	**0.0287**	0.848	0.5685	0.5873
			Indigenous diet			
Tertile 1	1.17 (0.49-2.78)	2.10 (0.52-8.49)	2.54 (0.58-11.09)	3.15 (0.92-10.84)	9.41 (1.29-68.43)*	11.41 (1.36-95.97)*
Tertile 2	1.85 (0.75-4.61)	6.09 (1.34-27.69)*	6.08 (1.30-28.38)*	0.89 (022-3.53)	0.99 (0.13-7.29)	1.00 (0.13-7.62)
Tertile 3	Ref	Ref	Ref	Ref	Ref	Ref
P-trend	0.3968	0.0636	0.0722	0.0823	**0.0400**	**0.0406**

*P<0.05, OR: Odds ratio, Hb: Hemoglobin, stfr: Serum transferrin receptor. Model 1: Crude association between anemia (Hb<12.g/dl, n=118; stfr>4.4ug/l, n=117). Model 3: Adjusted for household wealth index, BMI, household iodized salt usage, woman's age, number of adults and children in household. Model 3: Model 2 + Malaria.

3.6 Discussion

Currently, research is predominantly focused on micronutrients, not approaches that take into account overall dietary intake (dietary patterns or food groups) in relation to the risk of iron and iodine deficiencies in developing countries. However, meals consumed habitually consist of diverse foods containing complex combinations of nutrients that may be interactive, synergistic or antagonistic (National Research Council, 1989). Studying dietary approaches may have important public health implications because overall patterns of dietary intake might be simpler than nutrients or food components for the public to translate into diets (National Research Council, 1989). Additionally, knowing the relationships between mother-child dyads health indicators may contribute to early detection and treatment of either mother or child or both. Our study aimed to fill the gap in knowledge of: i) the role of DDS and or dietary patterns to predict the risk of iron and iodine deficiencies in women and their toddlers, ii) the relationship between mother-toddler health indicators (malaria, iron, and iodine indicators).

In this cross-sectional study, we examine the risk of iron and iodine deficiencies in relation to DDS and DP. DDS<4 had higher odds of iron and iodine deficiency, dietary patterns significantly predicted the risk of iron and iodine deficiency in women but not in toddlers. The scoring system in DDS9 which included 'fish' and 'milk and milk products" identified iodine deficiency risk only while DDS9G identified iron and iodine deficiency risk. As "fish" and "milk and milk products" are only consumed as condiments or added in minute amounts and viewed as 'prestigious foods', this may be an indication that

DDS9 points more to social structure of the rich and poor in our research setting, the rich have the financial muscle to purchase iodized salt.

Our findings on DDS are contrary to the findings of a similar study in Mozambique that found the inability of DDS to predict micronutrients (iron: haemoglobin & serum ferritin, and folate) status of women except for zinc which was only detected in the hunger season (Korkalo et al., 2016). Our findings was also contrary to a Cambodian study which found non-significant association of dietary diversity status and haemoglobin levels (McDonald et al., 2015). In Low- and Middle-income countries DDS is often used to predict the quality of diet (Arimond et al., 2010; Kennedy et al., 2009; Kennedy et al., 2007; Korkalo et al., 2016; Moursi et al., 2008) with little known about whether or not DDS can predict levels of biochemical indicators of micronutrient status. Though further research is needed, this makes our results relevant to screening for micronutrient deficiencies in rural northern Ghana. It was also interesting to note that women who lived in households that had a higher number of adults (>4) and had poor participation in household purchasing decision making had higher odds of iodine deficiency. The non-significant findings in toddlers may be due to kids relying largely on breastmilk as their source of nutrients and consumed less and monotonous complementary feeds.

Two dietary patterns were identified for women (DP 1 and DP 2) and one dietary pattern (DP 2) for toddlers. DP 1 (“rural elites diet”) is characterized by a high intake of tea, sugar, milk creamers, and fish condiments; and DP 2 (“indigenous diet”) characterized by dark green vegetables, grains, legumes, and wild fruits. The children had only “indigenous diet”. The two dietary patterns point to social structure, the rich consuming refined sugars and the poor consuming staple vegetables and grains. Children dietary pattern is largely being explained by the nature of complementary foods being based on cereal grains in the research setting. The risk of iron deficiency associated with the “rural elites diet” is largely due to consumption of added sugars, tea, and non-consumption of iron rich foods (dark green leafy vegetables and legumes) in the research settings. The non-significant association of DP 1 to iodine deficiency risk points partially to households’ ability to purchase iodized salt in rural communities. Iodized salt is one of the key sources of iodine in the research settings, the strong association of the “indigenous diet” and iodine deficiency may be a better predictor of iodine status in rural settings as we see an inverse association between household iodized salt use and iodine deficiency in our results.

Our results further indicated that as stfr decreases (improved iron status), Hb increases (improves) in both mother and child biomarkers intra and inter relationships. Additionally, nearly all the anemia in toddlers may be attributed to iron deficiency. Our findings on toddlers’ anemia is contrary to a study conducted in northern Ghana among school children. Mother-Child dyads’ IDA and anemia inversely inter correlates but not statistically significantly suggesting higher prevalence of deficiencies in toddlers (Table 2). This inverse relationship is further confirmed by the disagreement/poor agreement shown by our kappa analysis.

The strengths of our study were the use of DDS/dietary patterns to predict iron and iodine status using the biochemical indicators of the micronutrients. The study serves as a basis for screening for early detection of iron and iodine deficiencies. To the best of our knowledge our study is the first to use dietary diversity score and dietary patterns to predict the biochemical indicators of the iron and iodine in our research settings. This study could thus serve as the basis for further research into the development of dietary screening tools for micronutrients in Ghana.

We acknowledge that our study was a cross-sectional study as such a cause-effect relationship cannot be proven. Although the dietary pattern approach helps capturing the complexity of diet, the complexity poses researchers' bias or subjective interpretation when dietary patterns are labelled or named. The application of this results may be limited to settings similar to northern Ghana.

3.7 Conclusion

We found that the risk of iron and iodine deficiencies could be predicted or screened by DDS and DP. DDS<4 indicated higher odds of iron and iodine deficiency compared to DDS≥4. Two dietary patterns: DP 1 ("rural elites diet") and DP 2 ("indigenous diet") emerged, *rural elites diet* significantly predicted iron deficiency risk while *indigenous diet* predicted iodine deficiency risk. Dietary patterns prove to be a better predictor of iodine status in rural settings than use of iodized salt by households. We also found that anemia detected in toddlers was likely due to dietary iron deficiency while mothers' anemia is not. Mothers' and their toddlers' malaria status significantly correlated while iron status and iodine status of mothers and toddlers had insignificant correlation. our results suggest mother's malaria status may be used as a crude proxy for the child or vice versa. These findings are hoped to provide scientific bases to guide recommendations and policy making on programs targeted at reducing the risk of iron and iodine deficiencies.

Chapter Four: Hibiscus Sabdariffa meal improves iron status of child-bearing age women with time and prevents stunting in their toddlers in northern Ghana

4.1 Abstract

Globally, Iron deficiency (ID) is the most common form of nutritional deficiency, particularly in young children and childbearing age women. ID can affect maternal health and such birth outcomes as stunting and impaired cognitive development. In an effort to prevent ID in populations, fortification of selected foods with iron has been widely practiced and resulted in undesired such complications as increasing infections in developing countries with high prevalence of malaria. We investigated efficacy of an alternative food-to-food fortification utilizing indigenous iron-rich food sources. Child-bearing age women (15-49 yrs, intervention: n=58; control: n=60) and their children (6-24 mo,) consumed *Hibiscus Sabdariffa* leaves meals (HSM, 1.71mg Fe/100g meal) three times a week for 12 weeks during the dry/lean season in northern Ghana. We found that feeding the HSM (1.9 kg/day) improves iron status of women of child-bearing age with time (β=-0.32; 95% CI: -0.52, -0.07; p=0.011), and protects stunting among children during the dry/lean season (X^2=5.08; p=0.0243), the period with the worst food and nutrition insecurity. Compared with the control group, the number of stunted children declined in intervention group.

4.2 Introduction

Worldwide, iron deficiency (ID) is the most common form of nutritional deficiency with children and women of childbearing age being at the highest risk (Centers for Disease Control Prevention, 2002; Gayer and Smith, 2015; Stoltzfus, 2003). In Ghana, the prevalence of anemia is highest among children (anemia: 66% in 2014,) and women of childbearing age (anemia:42% in 2014) (Abizari et al., 2012; Ewusie et al., 2014; GSS, 2009; 2011; 2014; Zimmermann and Hurrell, 2007) along with high prevalence of stunting in children (19% in 2014). Within Ghana, the three northern regions (Upper East, Upper West and Northern regions) had higher prevalence of anemia (74-82%), iron deficiency anemia (IDA, 63%), iron deficiency (78%) and stunting rate (14.4-33.1%) compared to southern regions (54-70% anemia and 10.4-22.0% stunting) in 2014 (GSS, 2009; 2011; 2014; WFP, 2015). (GSS, 2014).

Consequences of IDA include poor maternal health and poor fetal growth (Caulfield et al., 2006; Strauss and Dietz, 1997), and birth outcomes such as growth retardation, stunting and impaired cognitive development (Soemantri et al., 1985). Globally, several approaches to counteract iron and other trace minerals deficiencies included malaria control, fortification of food, supplementation and dietary diversity approaches. Several challenges associated with the fortification of food and supplementation of elemental iron in developing countries include limited access by rural households, cost, and sustainability of the products (Yip and Ramakrishnan, 2002). With complications of malaria such as parasitaemias and inflammation, World Health Organization recommended the use of serum concentration of soluble transferrin receptor (stfr) in testing for iron status as stfr is a known sensitive marker of iron status which is less affected by infection and inflammation as compared to serum ferritin (World Health Organization, 2001).

In Ghana, numerous costly supplementation and food fortification programs have been adopted to counteract iron and other micronutrients deficiencies. Ghana has implemented iron and folic acid supplementation for pregnant women, national vitamin A supplementation for children under 5 yrs and pregnant and lactating women; universal salt iodization policy (David, 2003; Nyumuah et al., 2012b; Saaka, 2012). However, to the best of our knowledge, efficacies of these programs at the national level has not yet been reported in the midst of high prevalence of anemia.

In developing countries in Africa, ID accompanies nutritional deficiencies of many other micronutrients and proteins due to monotonous, plant-based, staple food intake with limited nutrient-rich animal source of foods. Sustainable, food-based, practical, and cost-effective, long-term, alternative solutions to prevent ID at the population level in Africa (Abizari et al., 2012; Zimmermann and Hurrell, 2007) are yet to be explored. Little data on food composition and or iron content exist for most indigenous foods in sub-Saharan Africa for which Ghana is no exception. The aim of this study was therefore to investigate the efficacy of native leaves meal (HSM) containing dry fish in improving iron status of reproductive age women (all are lactating women) and protects toddlers from stunting through a community-based feeding intervention during dry/lean season.

4.3 Subjects and methods

4.3.1 Study design

This community-based 12-week feeding trial was planned in a quasi-experimental design with its primary focus on improving iron status of dyads, determined by hemoglobin and serum concentration of soluble transferrin receptor (sTfR).

4.3.2 Study site

This study was carried out in two districts. The districts are in the Upper East Region in Ghana (Fig 1): Kassena Nankana West (KNWD) and Builsa North (BND) districts. The capitals of these districts are about 25 km away from each other and linked with a single paved road via the Kassena Nankana Municipality and about 42 km away from the regional capital Bolgatanga. The study area has a typical savannah woodland vegetation characterized by short, scattered, drought-resistant trees and grass. The study area has two main seasons: a dry season (October – April/May) characterized by high temperatures and the rainy season (May/June–September). People in this area are mostly subsistence farmers. The study area is malaria endemic with peaks at the end of the rainy season when 45% of households are food insecure (Osarfo et al., 2016). These districts were among the top five food insecure districts in the region. The selected sites for this study were two communities (Sakaa and Chania) in KNWD and three communities (Chuchuliga-yipaala, Azoayeri, and Awulansa) in BND. The study sites were selected based on the inclusion criterion of having water source (functional borehole) throughout the dry season from our preliminary inquiries, existing women groups, and access to Community-Based Health Planning and Services (CHPS) compounds, sizeable number of mother and young children (6-23 months) dyads for good sampling frames and community health nurses who were willing to work with researchers between May and August 2016. Researchers' previous experience with those communities facilitated the community entry process.

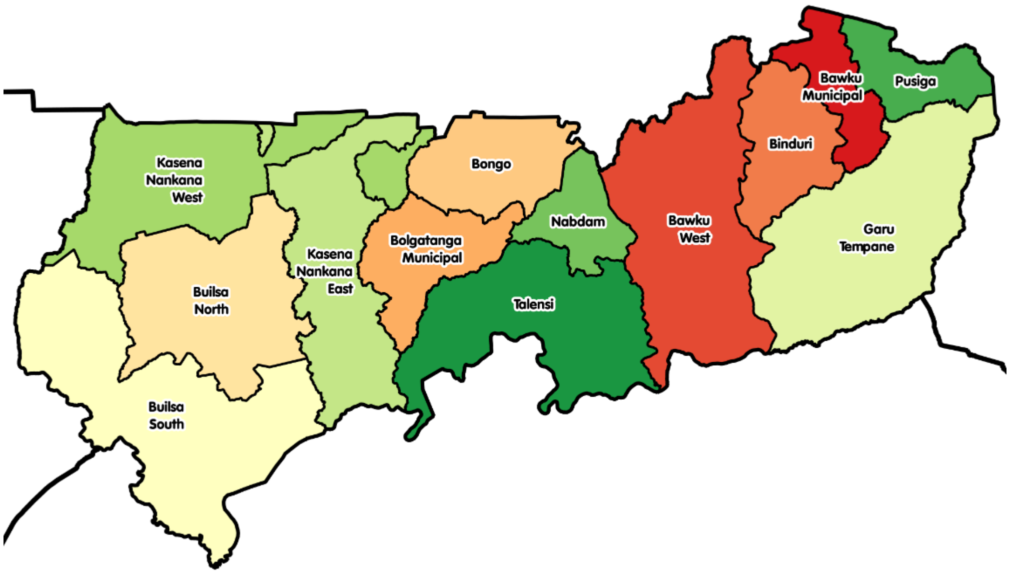

Figure 3. A map of Upper East region showing its districts

4.3.3 Study subjects and selection

The study subjects, women 15-49 y and their children 6-23 months were recruited in May 2016. The dyads were drawn from selected districts using community-based birth registers at CHPS compounds kept by community health nurses stationed in these communities or by community health volunteers. The health volunteers are community members who are often not paid. Announcements through the community chiefs/leaders were then made in the respective communities for all women with children under 5 years of age to meet at their respective community health centers.

Research team briefed the women and the community leaders on the study. Names that were shortlisted from the birth registers were read out. All dyads including those who were not in register but obtained from the health volunteers who knew almost everybody in their catchment areas were contacted and checked on their willingness to participate in the study through the community health volunteers and community health nurses. Finally, verbal consent was sought from spouses of the women who were willing to be part of the study.

A total of 120 dyads (60 mother/child dyads each for intervention and control group) were drawn from the two districts and agreed to participate in the study. Optimal design software (version 3.01) was used to calculate the minimal sample size of 100 dyads (50 dyads each for intervention and control groups) with a power of 80%, significance level of 5%, coefficient of determination of 65%, minimum detectable effect of 0.33 of diet on hemoglobin change, and 20% attrition rate.

We had three sets of twins, one in intervention group and two in the control group in the study. We had 7% attrition rate in intervention group; two dyads relocated before baseline data collection and two relocated in the 6th week. All relocations were either to join a spouse or the entire nuclear family were migrating for farming purposes.

4.3.4 Intervention

Feeding trial

The participating dyads in intervention communities were invited to consume veo soup/meal three times a week and provided weekly supply of iodized salt (450 g) for the household usage.

Veo soup modified: The veo soup/meal is a local Ghanaian soup/meal mainly made of *Hibiscus Sabdarifa* leaves. It is a soup when prepared a bit watery and consumed with 'tou zaafi' (millet or corn based cooked paste). It is also a meal when prepared thick and eaten by itself. The *Hibiscus Sabdarifa* leaves meal used in the present study was made of 18 kg leaves, 8 kg groundnut, 1.1 kg dawadawa (fermented African locust beans), 3 kg dried fish plus 0.045 kg iodized salt, cooked with about 23 L (23 kg) water to yield 52.5 kg veo meal. The meal was modified in this study by addition of dried fish. Dried fish or meat is a commonly added ingredient for people of high socioeconomic status especially in the cities.

The *Hibiscus Sabdarifa* leaves meals were prepared by the women themselves to build trust and avoid any forms of suspicion. The researcher provided all ingredients and supervised all cooking and feeding activities. In each community, groups of ten women took turns to share the cooking activities, washing of bowls, and making water available for cooking. Three women in each community were trained on food weighing/measurement to conform to our standardized recipe. It took 45-60 minutes to cook a batch of the meal. It was then served (1.5 kg/woman) to all women and their children (0.5 kg/child) separately. The women and children were given two separate bowls, so that the researcher could monitor each person's consumption. All dyads were encouraged to consume to their satisfaction by requesting additional servings. The meal intake was measured by the researcher (CK) and trained women by the differences between the quantities served minus leftovers. The women and children consumed on average 1.9 kg and 0.4 kg, respectively.

The food composition analysis of the final veo meal products was carried out by Great Lakes Scientific Inc. (Stevensville, MI, USA. Web: http://www.glslab.com/). Our standardized veo meal (per 100 g) contained 1.7 mg iron, 6.6 mg protein, 4.6 mg fat, 82.6% water, and 2.7% ash.

4.3.5 Measurements and data collection

Questionnaire/interview: a questionnaire on food intake frequency and 24hr food intake recall was administered at baseline and at the end point for all participants. At baseline, information on sociodemographic characteristics was also taken.

Malaria status was screened at baseline, midpoint, and at end point for all research participants by Rapid Malaria Diagnostic cassettes (Lot: 05CDB050DA. Standard Diagnostic, Inc. Republic of Korea.). Malaria screening is based on the presence of Histidine-Rich Protein-2 (HRP-II) in whole blood. HRP-II is known to be specific to *Plasmodium falciparum*, which causes more than 90% of malaria cases in Ghana (Asante et al., 2011). Sensitivity and specificity of cassettes were 95% and 99.5% respectively. All participants who were screened positive for malaria were referred to their respective CHPS compounds for treatment. In most instances, the community health nurses who were in charge of compounds and also part of our research team provided medicine to children on site and women upon presenting their health insurance IDs. Anemic individuals were also referred for treatment.

Anthropometry: Participants' weights and heights were measured at baseline, midpoint, and at end point according to standard procedures (Cogill, 2003). Electronic scale (Serial number 5874030154862, Model 874 1321009. Seca gmbh & co kg, 22089 Hamburg, Germany.) was used to measure weights to the nearest 0.1 kg.

Biochemical measurements: At baseline and endpoint, 5 ml whole blood was withdrawn into silica-coated serum separator vacutainers (Lot: 20140618, Anhui Kangning Industrial (Group) Co. Ltd, Tianchang City, China) and held at ambient temperature until and during transportation. Serum was separated using a centrifuge (Hettich) at 500 ×*g* for 5 min at room temperature. Separated serum was aliquoted, kept frozen at −18°C (Thermo Fisher Scientific) at NHRC, and then transported on dry to MDS Lancet laboratories in South Africa for serum transferrin receptor analysis. Hemoglobin levels were measured in the field using HB 201 analyzer according to prescribed procedures. At mid-point, only hemoglobin levels were measured.

Nutrient measurement: Iron concentrations of *veo meal, hibiscus leaves, dawadawa, Amani, and groundnut* were obtained by removing the organic content of samples in a high temperature muffle furnace. The resulting minerals were diluted in acid and absorbance were read by Atomic Absorption Spectrometry (3110 Perkin Elmer Atomic Absorption Spectrometer with a Hollow Cathode Calcium/Magnesium Lamp at 285.5 nm). Analyses were carried out by standard procedures iron (AOAC 985.35), protein (AOAC 928.08II), fat (AOAC 925.12), moisture (AOAC 950.46A), and ash (AOAC 923.03). Analyses were done by Great Lakes Scientific Inc. Stevensville, MI, USA.

4.3.6 Statistical analysis

Data analyses were carried out by SAS 9.4 (SAS institute). Characteristics of participants were described using frequency distributions. Comparison of participants' characteristics between intervention and control groups was done using Chi-square statistics and Student t-test for categorical and continuous variables, respectively.

Multiple logistic regression was done using GENMOD. GENMOD is capable of handling data when the normality assumption may not be reasonable. When the responses are discrete and correlated, generalized estimating equations (GEEs) provide a practical method with reasonable statistical efficiency to analyze such data using GENMOD.

Dependent variables of interest were linear growth (Stunting: HAZ <-0.2) of children, and iron status (ID: stfr>4.40 Lab reference; Anemia: Hb<12g/dl; IDA: concurrent presence of anemia and ID). Intra group hemoglobin levels were compared by PROC MIXED.

Statistical analysis was done in twofold: 1) analysis using non-anemic (Hb≥12 g/dl) participants at baseline and analysis with anemic (Hb<12 g/dl) plus non-anemic (Hb≥12 g/dl) participants combined, 2) analysis using iron sufficient (stfr≤4.4 ug/l) individuals at baseline and analysis with IS (stfr≤4.4 ug/l) plus ID (stfr>4.4 ug/l) participants combined. This became necessary as anemia and iron deficiency were not matching, anemia under estimated the prevalence of iron deficiency. Emphasis is on the later analysis as our focus is on the potential of meal to improve iron status. Iron status measured by anemia using hemoglobin levels did not truly reflect iron deficiency, most of the anemia could be due to other causes. It is worth noting that for iron status using stfr, we had 58 samples at baseline in intervention but got results for 57 samples as one of the samples was not adequate for such analysis. At endpoint we got 53 results which included endpoint results for the one missing at baseline, this explains the discrepancies in tables 2 and 3. The combined number of drop outs and missing data (12%) is less than the anticipated attrition rate of 20%. Based on the 12%, the final data analysis is not expected to be affected. Additionally, participants with missing variables were not significantly different from the other participants.

E*thics:* Research procedures were in accordance with the Michigan State University's Institutional Review Board and Navrongo Health Research Centre (NHRC) Institutional Review Board (IRB) in Ghana

4.4 Results

Sociodemographic characteristics of childbearing age women and their children in both districts is summarized in Table 10. There were no significant differences between the two districts in all measured variables (participation in household decision making; wealth index; household head's sex; living with animals in same compound; number of adults and children under five years in household; number of child's siblings; age of woman, child, and household head), except for women's occupation, religion, ethnicity, and quantity of iodine in household salt. All the women who reported other forms of occupations were also engaged in farming in the wet season. Mother and child health indicators (weight, height, serum transferrin receptor level, hemoglobin level, iodine status, malaria status, anemia and iron status) in Table 11 do not vary by groups except children anemia status, iodine status, and height. On child nutritional status, there was a significant difference between groups for Z-scores for HAZ but not WHZ. Interestingly, when HAZ was categorized into stunted and non-stunted children, there was no differences between the groups. Anemia, ID, and IDA prevalence are not significantly different between intervention and control groups across time except for anemia and IDA in children at baseline. There was significant difference between groups for stunting at endpoint (p=0.0243)

In comparing the prevalence of anemia and iron deficiency at endpoint among mothers who were iron deficient (ID) and iron sufficient (IS) at baseline in table 12, there was no significant differences between groups. Intervention however had fewer iron deficient/anemia (n=1, 3.7% / n=11,40.7%) as compared to control group (n=4,14.3% / n=13,43.3%) becoming iron deficient/anemic at endpoint though not statistically significant but biologically relevant. The differences at endpoint among initially anemic individuals (baseline) could be influenced by a decision to accept/seek treatment after referral at baseline.

Endpoint comparison of iron status shows no significant differences between groups in comparing individuals who were iron deficient (ID) and iron sufficient (IS) at baseline (data not shown). Interestingly, intervention group indicates a protective direction for iron deficiency (beta=-1.28, 95% CI: -5.49-2.93) though not statistically significant (p=0.552). Probably only direction can be shown with the small sample size, no power to indicate differences in groups as data has been split we thus combined the two groups in table 13.

Table 13 indicates endpoint comparison of iron status (combined individuals who were IS and ID at baseline). Intervention group appears to be protective of iron deficiency with time (beta=-0.32, p=0.011) after adjusting for household wealth index, marital status, sex of household head, mother's age, child's age, malaria status, participation in household decision making, number of children and adults in household. This could suggest that we needed a longer duration to enable us establish differences between groups. Households with male heads are at a higher risk of iron deficiency (coefficient =1.66, p-value=0.045) as compared to households with female heads.

On Hemoglobin (Table 14), there were no significant differences between group means. Changes within groups were however observed. Intervention group had some significant variations (T3-T1: mean Δ=0.24g/dl, p-value=0.0987; T3-T2: mean Δ=-0.06g/dl, p-value=0.0442; and T2-T1: mean Δ=0.06g/dl, p-value=0.6712) but not in the control group (T3-T1: mean Δ=0.05g/dl, p-value=0.7065; T3-T2: mean Δ=0.11g/dl, p-value=0.6479; and T2-T1: mean Δ=-0.06g/dl, p-value=0.0987). After adjusting for covariates (household wealth index, marital status, sex of household head, mother's age, child's age, malaria status, time*group interaction, empowerment, number of children and adults in household), T3-T1 (p=0.0793) and T3-T2 (0.0824) became marginally significant in intervention group. Changes within control group remain insignificant after adjustment. Women and children in the intervention consumed on average 1.9 kg and 0.4 kg, respectively of HSM per day.

Table 9. Demographic characteristics of childbearing age women (15-49 yrs) and their children (6-23 mo) in intervention and control groups at baseline

Variable		Intervention		Control		
		n	Mean ± SD	N	Mean ± SD	P-value
Women age (yrs)		58	26.7±6.7	60	26.1±5.4	0.598
Children age (months)		59	14.1±5.1	62	12.6±5.2	0.119
Household head's age		58	47.0±16.9	60	42.7±15.1	0.149
Number of household adults		58	4.6±3.3	60	3.7±1.4	0.051
# Siblings in household		59	1.4±1.6	62	1.9±1.5	0.092
# children < 5 yrs		58	1.0±1.2	60	1.1±1.2	0.708
Variable		n	%	n	%	X^2(p-value)
Occupation of women	Farmer	31	53.5	50	83.3	15.3 (0.002)
	Handy works	5	8.6	5	8.3	
	Housewife	6	10.3	2	3.3	
	Trader	16	27.6	3	5.0	
Household head sex	Male	54	93.1	56	93.3	0.00 (0.960)
	Female	4	6.9	4	3.4	
Wealth index	Lower	19	32.8	20	33.3	0.02 (0.991)
	Middle	20	34.5	20	33.3	
	Upper	19	32.7	20	33.3	
Decision making	Low	11	20.0	9	15.0	1.2 (0.545)
	Average	27	46.6	34	56.7	
	Above	20	34.5	17	28.3	
Household iodize salt	Non-users	46	80.7	59	98.3	9.8 (0.002)
	Users	11	19.3	1	1.7	
Iodine level in household salt	<15ppm	52	91.2	60	100.0	5.5 (0.019)
	≥15ppm	5	8.8	0	0.0	
Ethnicity of women	Builsa	2	3.4	56	90.3	92.3 (<.000)
	Kassena	55	93.2	5	8.1	
	Nakani/Fulani	2	3.4	1	1.6	
Religion	Christian	34	57.6	60	96.8	27.0 (<.000)
	Muslim	9	15.3	0	0.0	
	Traditionalist	16	27.1	2	3.2	
Animals live in household	Yes	51	87.9	58	96.7	3.2 (0.074)
	No	7	12.1	2	3.3	

Add superscripts in the table Traditionalist: Practitioners of African Traditional Religion; Decision making: the degree in which women participate in household decision making. Wealth index: Household cumulative living standard measured by using household assets

Table 10. health indicators of child bearing age women (15-49 yrs) and their children (6-23 mo) in intervention and control groups across time

Variable	Intervention		Control		
Baseline	N	Mean ± SD	N	Mean ± SD	p-value
Women height (cm)	58	160.3±6.6	60	161.7±5.6	0.217
Women weight (kg)	58	57.1±9.7	60	57.3±7.6	0.888
Women Hb (g/dl)	58	11.8±1.3	60	11.8±1.2	0.775
Women sTfr (ug/l)	57	5.1±3.1	60	4.7±1.6	0.320
HAZ	59	0.3±1.3	62	-0.3±1.3	0.009
WHZ	59	-1.2±1.3	62	-0.9±1.3	0.294
children Hb (g/dl)	59	9.4±1.7	62	9.0±1.3	0.171
Children sTfr (ug/l)	58	9.1±5.1	62	9.7±5.3	0.528
	Status	N(%)	N(%)	X^2	p-value
women ID (stFr)	deficient	28 (49.1)	28 (46.7)	0.07	0.790
	normal	29(50.9)	32(53.3)		
women IDA	anemic	45(79.0)	41(68.3)	1.69	0.194
	normal	12(21.0)	19(31.7)		
women General anemia(Hb)	anemic	27(46.5)	35(58.3)	1.64	0.200
	normal	31(53.5)	25(41.7)		
Women malaria	Positive	7(12.1)	5(8.3)	0.50	0.502
	Negative	51(87.9)	55(91.7)		
children ID (stFr)	anemic	56(96.6)	61(98.4)	0.40	0.520
	normal	2(3.5)	1(1.6)		
children IDA	anemic	47(81.0)	59(95.2)	5.80	0.016
	normal	11(20.0)	3(4.8)		
children general anemia (Hb)	anemic	50(84.8)	60(96.8)	5.29	0.0214
	normal	9(15.2)	2(3.2)		
Children malaria	Positive	4(6.8)	6(9.8)	0.40	0.545
	Negative	55(93.2)	55(90.2)		
Stunting (HAZ)	Stunted	3(5.1)	7(11.3)	1.50	0.215
	normal	56(94.9)	55(88.7)		
Week 6					
women general anemia (Hb)	anemic	27(51.9)	26(44.8)	0.55	0.457
	normal	25(48.1)	32(55.2)		
women malaria	Positive	20(39.2)	18(31.0)	0.80	0.371
	Negative	31(60.8)	40(69.0)		

iron deficiency (ID) defined by sTfr >4.40 ug/l and >2.85ug/l in women and children respectively, general anemia defined by Hb <12 g/dl and Hb < 11g/dl in women and children respectively, iron deficiency anemia (IDA) defined by concurrent presence of general anemia and ID, Positive Malaria: have malaria. HAZ: height-for-age z-score. WHZ: weight-for-age z-score

Table 10 (cont'd)

Variable		Intervention		Control	
	N	Mean ± SD	N	Mean ± SD	p-value
Week 12					
women ID (stFr)	deficient normal	18(44.0) 35(66.0)	20(35.7) 36(64.3)	0.04	0.848
women IDA	anemic normal	11(20.2) 42(79.3)	13(23.2) 43(76.8)	0.10	0.757
Women general anemia (Hb)	anemic normal	24(44.4) 30(55.6)	30(51.7) 28(48.3)	0.59	0.441
women malaria	Positive Negative	11(20.0) 44(80.0)	13(22.4) 45(77.6)	0.10	0.754
Week 6					
children general anemia (Hb)	anemic normal	49(94.2) 3(5.8)	58(96.7) 2(3.3)	0.40	0.534
children malaria	Positive Negative	20(38.5) 32(61.5)	7(11.7) 53(88.3)	10.93	0.001
Week 12					
children ID (stFr)	deficient normal	58(100) 0(0.0)	56(96.6) 2(3.4)	2.04	0.154
children IDA (stFr)	anemic normal	42(72.4) 16(27.6)	46(79.3) 12(20.7)	0.75	0.386
children Anemia (Hb)	anemic normal	43(78.2) 12(21.8)	49(42.6) 11(9.6)	0.20	0.641
children malaria	Positive Negative	18(32.7) 37(67.3)	11(18.3) 49(81.7)	3.15	0.076
Stunting (HAZ)	stunted normal	2(3.64) 53(96.36)	10(16.39) 51(83.61)	5.08	**0.0243**

iron deficiency (ID) defined by sTfr >4.40 ug/l and >2.85ug/l in women and children respectively, general anemia defined by Hb <12 g/dl and Hb < 11g/dl in women and children respectively, iron deficiency anemia (IDA) defined by concurrent presence of general anemia and ID, Positive Malaria: have malaria. HAZ: height-for-age z-score. WHZ: weight-for-age z-score

Table 11. Endpoint comparison of iron deficiency prevalence among mothers in intervention and control groups

Parameter		Baseline iron deficient				Baseline iron sufficient			
		Intervention group	Control group	X	P-value	Intervention group	Control group	X	P-value
stfr	>4.40ug/l	17(68.0)	16(57.1)	0.67	0.416	1(3.7)	4(14.3)	1.86	0.172
	≤4.40ug/l	8(32.0)	12(42.9)			26(96.3)	24(85.7)		
Hb	<12g/dl	13(50.0)	17(60.7)	0.63	0.429	11(40.7)	13(43.3)	0.04	0.843
	≥12g/dl	13(50.0)	11(39.3)			16(59.3)	17(56.7)		

T1: Baseline, T2: Midpoint, T3: Endpoint, Hb: hemoglobin, Iron deficiency (ID): stFr >4.4ug/l, Anemia: Hb<12g/dl

Table 12. Endpoint comparison of iron deficiency prevalence among intervention and control groups

Parameter	Estimate	SE	95% CI		P-value
Intervention Group	0.17	0.52	-0.85	1.19	0.749
Control group			Ref		
Male household head	1.66	0.82	0.04	3.27	0.045
Female household head			Ref		
Malaria absent	0.15	0.39	-0.61	0.90	0.701
Malaria present			Ref		
Lower wealth index	0.52	0.47	-0.40	1.44	0.269
Middle wealth index	0.45	0.41	-0.34	1.25	0.265
Upper wealth index			Ref		
Married	-0.23	0.93	-2.04	1.59	0.806
Single	-1.22	1.13	-3.43	1.00	0.282
Divorce/separated			Ref		
Low decision making	-0.66	0.54	-1.73	0.41	0.227
Average decision making	-0.79	0.46	-1.69	0.10	0.081
Above average decision making			Ref		
Time and intervention group interaction effect	**-0.32**	**0.13**	**-0.57**	**-0.07**	**0.011**
Time and control group interaction effect	-0.25	0.15	-0.56	0.05	0.097
BMI	0.37	0.33	-0.27	1.01	0.256
Women age (yrs)	-0.02	0.03	-0.09	0.04	0.448
Children age(months)	-0.05	0.04	-0.12	0.01	0.121
Number of household Adults	-0.09	0.11	-0.29	0.12	0.426
# children < 5 yrs in household	-0.04	0.18	-0.38	0.30	0.819

Adjusted: Household wealth index, Marital status, Sex of household head, Mother's age, Child's age, malaria status, time*group interaction, women participation in household decision making, number of children and adults in household.

Table 13. Hemoglobin and transferrin receptor levels of child-bearing age women and the effects of consumption on indicators across study period

			Non anemic and anemic-referred mothers			
Variable/Time	Intervention			Control		
	Mean	P-trend	Adj P-value	Mean	P-trend	Adj P-value
Hb T1	11.84			11.77		
T2	11.78	0.1032		11.87	0.7036	
T3	12.08			11.82		
Group Hb	11.96			12.05		0.6897
	Mean±SE	P-value	Adj P -value	Mean±SE	P-value	Adj P -value
T1-T3 Hb	-0.24±0.15	0.0987	**0.0793**	-0.05±0.13	0.7065	0.5673
T1-T2 Hb	0.06±0.15	0.6712	0.9544	-0.11±0.13	0.4034	0.3473
T2-T3 Hb	-0.30±0.12	**0.0442**	0.0824	0.06±0.13	0.6479	0.6923
			Non anemic mothers at baseline			
T1-T3 Hb	-0.01±0.21	0.7886	0.9657	0.12±0.20	0.6338	0.5498
T1-T2 Hb	0.22±0.23	0.1476	0.3415	0.13±0.21	0.6903	0.5404
T2-T3 Hb	-0.23±0.22	0.2336	0.2920	-0.01±0.20	0.9333	0.9707
	Mean	P-trend		Mean	P-trend	
T1 Hb	12.44			12.68		
T2 Hb	12.22	0.5145		12.55	0.7854	
T3 Hb	12.45			12.56		
Group Hb	12.37			12.60		0.9833

T1: Baseline, T2: Midpoint, T3: Endpoint. HAZ: Height-for-age Z-score, WAZ: Weight-for-age Z-score, WHZ: Weight-for-height Z-score. Adjusted: Household wealth index, marital status, sex of household head, mother's age, Child's age, malaria status, women participation in household decision making, number of children and adults in household.

Note: The women and children in intervention consumed on average 1.9 kg and 0.4 kg, respectively of hibiscus meal per day.

4.5 Discussion

Iron fortification of foods has long been held to contribute to reducing ID and IDA prevalence, it is said to be sustainable, practical, and cost effective in the long term at the population level, but it has shown little success in Africa (Abizari et al., 2012; Zimmermann and Hurrell, 2007). Additionally, fortification may not reach people at greatest risk for nutritional ID who depend on subsistence farming and have little access to processed food items (Lynch, 2011).

This suggests that fortification might not be a universal solution, it would have to work alongside other approaches depending on the settings. Dietary approaches such as food-to-food fortification can be a promising approach to improve trace mineral intake especially in situations of low dietary trace mineral intake and bioavailability from monotonous diets based on a small number of staple plant foods (Cercamondi et al., 2014) as is the case in most developing countries. The fundamental challenge is often the efficacy of these food-to-food fortification to improve iron status. Our study sought to test the potential of indigenous meal to improve iron status in mother-child dyad and linear growth in children.

The results of our study suggest that indigenous *Hibiscus Sabdariffa meal* improves iron status of women of child-bearing age with time and could be protective of stunting among children during the dry/lean season. Lean season in developing countries is widely known to affect food and nutrition security (Gelli et al., 2017), mitigating the effect of nutrient deficiencies in this period is thus very timely and crucial. Our findings could be one of the needed solutions to the high prevalence of iron deficiency and anemia prevalence most especially in the dry season in northern Ghana.

This study is among the very few studies showing the potential of a food-based approach to improve iron status without using biosynthetically incorporated iron. A modest increase in iron status (stfr) was observed in a food study trial using animal-source food in rural Vietnam (Hall et al., 2017). In another fold, cowpea-based food containing fish meal and served with a vitamin C–rich drink improved hemoglobin concentration but not ferritin levels (Egbi et al., 2015). Consumption of heme iron-rich foods (goat meat and liver) and enhancers (orange juice and other fruit juice) of non-heme absorption decreased anemia and iron deficiency anemia but not ferritin levels in Burkinabe preschoolers (Sanou et al., 2010). These findings point to the difficulty involved in testing the efficacy food-to-food fortification, interestingly our study indicates significance with time suggesting that a longer duration is required to give clear cut difference between groups. Time and intervention group interaction effect shows statistically significant (beta=-32, p=0.011) differences between groups. This further points to the role of time and the need for a longer duration for the study. Similarly, a biologically relevant result was obtained as the intervention group had fewer becoming iron deficient (n=1, 3.7%) as compared to those the control group (n=4,14.3%) had becoming iron deficient at study endpoint, though all participants were iron sufficient at baseline.

Similar observations were made in hemoglobin levels. The velocity of change was only prominent in the intervention group, the key change being between the midpoint to the endpoint (p=0.0442, Adjusted p=0.0824). This suggest a need for a longer duration for the trial to clearly elucidate the impact of the diet (*Hibiscus Sabdariffa* meal).

At baseline almost all the children were anemic and iron deficient and were referred for treatment as such the effect of the meal could not be established among the children. Analysis among IS children is not feasible due the small sample size. Similar comparison as in the case of the women cannot be done as well. Ideally, they should have been treated and rolled on to the program but constraints beyond our control did not support this approach. On the other hand, stunting levels were not significantly different between groups at baseline but went up by 5.1% and decreased by 1.4% in control and intervention groups respectively. Growth in this case may not be linked to the iron content but the diet in totality as a review on interventions using energy, protein, zinc, iron, copper, iodine and vitamin A showed no clear and or consistent evidence that supplementation with any of the nutrients singly benefited linear growth. The conflicting results suggest the strong probability that growth is limited by multiple and simultaneous deficiencies in many populations (Allen, 1994).

The strengths of our study were the use of control and intervention groups to elucidate the effect of the intervention. The use of indigenous staple foods makes the findings relevant and practical to impacting nutritional status of vulnerable populations. Furthermore, to the best of our knowledge, our study is the first of its kind in the research settings to use indigenous meal only without using biosynthetically incorporated nutrients to improve iron and nutritional status. This study is not without limitations/challenges. We recognized that not measuring inflammatory markers could have influenced our results, to minimize the impact of infections and inflammation, we used transferrin receptor as recommended by WHO. We suggest measurement of various markers of iron and inflammation be done in future studies. The timing for our study was short and might have impacted on our results, we suggest six to twelve months' study in future research works. Not doing a bioavailability study on our diet is another limitation that needs to be addressed in subsequent studies. Complete treatment of all individuals with anemia especially in the case of the children before rolling them into the program would give the full effects of the prescribed intervention.

4.6 Conclusion

We demonstrated the potential of indigenous *Hibiscus Sabdariffa* meal to improve iron status of women of childbearing age with time, and its protectiveness against stunting among children in a period where prevalence of food and nutrition insecurity are highest. Our findings may be one of the needed solutions to tackling iron deficiency and stunting among rural populations by policy makers, health and nutrition educators as well as all stake holders in the health sector in Ghana. Though our findings look promising, we recognize the need for further research on the efficacy of the meal/soup to improve iron status and linear growth.

Chapter five: Iodine status of reproductive age women and their children in northern Ghana improved through household iodized salt and weekly indigenous meal supply

5.1 Abstract

Iodine deficiency (ID) during pregnancy results in pregnancy losses, intrauterine growth retardation, and lower IQ in the offspring. Even after two decades of universal salt iodization (USI) implementation, the efficacy of USI has not been reported in high risk groups in vulnerable regions in Ghana. We aimed to improve ID status in childbearing age women (all are lactating women) and their children in northern Ghana, a geographically and socioeconomically vulnerable region. We provided weekly household iodized salt supply and community-based feeding of native *Hibiscus Sabdariffa* leaves meal (HSM) prepared with iodized salt to women and their children in intervention (n=60) vs. control group (n=60). At baseline, ID was prevalent in women (36%) and their children (29%). For women, both median UIC values for intervention (57.4 ug/l) and control group (65.1 ug/l) were below the recommended UIC value of 100 ug/l with no significant differences between the two groups (p=0.2778). At the endpoint, median UIC for the intervention group (123.6 ug/l) was significantly higher (p=0.008) than the control group (59.7 ug/l). Our results suggest that iodized salt is an effective channel for the improvement of iodine status of economically disadvantaged groups living in remote areas in vulnerable locations due to geographical proximity to coastal lands. Our results further suggest that decreased urinary iodine levels (median UIC) among lactating mothers does not necessarily imply lower iodine status for their breastfed children. The results may point to a non-improvement in ID (Median UIC of 50-99 ug/l) in the past decade for Ghana. There is a need to revisit, assess, and to

ascertain the bottlenecks and challenges preventing populations from attaining the intended benefits of the universal salt iodization policy adopted by Ghana.

5.2 Introduction

Iodine is an essential trace mineral for living organisms as an intramolecular component for the biosynthesis of thyroid hormones. Thyroid hormones control cell growth and differentiation, increase proteins, lipids, and carbohydrates metabolism (Haldimann et al., 2015). Iodine deficiency disorders, a collective number of health outcomes, are the world's leading causes of preventable cognitive retardation and poor psychomotor development in children. ID is commonly associated with goiter, intellectual impairments (Delange, 1994), growth retardation, neonatal hypothyroidism, and increased pregnancy loss and infant mortality (Haldimann et al., 2015; Pearce et al., 2013).

Iodine needs are mainly met from diet or supplementation. Major dietary sources of iodine vary from country to country dependent on soil content and food sources (Lee et al., 2016a; Lee et al., 2016b). Those major sources of dietary iodine in developed countries such as dairy products, eggs, seafoods and fortified foods are not readily available or consumed in adequate amounts in developing countries. Furthermore, feed for cows and chicken in developed countries are fortified with iodine. The most important alternative source of iodine in developing countries, particularly in locations far from the ocean, is iodized salt (Lynch, 2011; Ross et al., 2014). Iodized salt is the most common source of iodine for most African countries for which Ghana is no exception.

Globally, iodine deficiency is the leading cause of intellectual deficiency (Hollowell et al., 1998; Zimmermann, 2009). About 1.88 billion people have insufficient dietary iodine intakes with most of them living in economically disadvantaged areas (Andersson et al., 2012) including Africa. In an effort to cope with the high magnitude of global burden of iodine deficiency, World Health Organization (WHO)/United Nations Children's Fund (UNICEF)/International Council for Control of Iodine Deficiency Disorders (ICCIDD) recommended universal salt iodization (USI). USI is seen as a safe and cost-effective strategy to mitigate iodine deficiency as salt is consumed by all individuals and in all countries. WHO also recommend routine and regular monitor of a population's iodine status (Lee et al., 2016a; World Health Organization, 2007) by urinary iodine concentration (UIC). Since over 90% of ingested dietary iodine is excreted in the urine, the median UIC from spot urine samples is used in assessing iodine status of the group, but not that of an individual (Hurrell, 1997; Lee et al., 2016a; World Health Organization, 2007).

Iodine deficiency has recently been re-emerging as a public health concern, particularly in some industrialized countries such as Australia and the United Kingdom (UK), which have not adopted USI and hitherto were regarded as iodine-sufficient countries (Bath et al., 2008a; b; Lee et al., 2016a; Li et al., 2006; Vanderpump, 2012; Vanderpump et al., 2011). In developing countries, the burden of ID is further exacerbated (Andersson et al., 2012; Pearce et al., 2013). Currently in Ghana, ID prevalence among women of reproductive age is unknown as there is no data available on UIC. Per the 2014 national demograhic health survey, household iodized salt usage indicated that about 38.5% of households at the national level used iodized salt fortified at adequate concentration (≥15 ppm). However, the rate is much lower in inland Northern (16%) and Upper East (18%) regions compared to those in Western (60.3%) region and such urban areas as Accra (54.9%). In general, households in rural areas use adequately iodized salt at a much lower rate (26%) than those in the urban areas (50%) (Ghana Statistical Service et al., 2015).

Lactating women and their toddlers have increased iodine needs and are vulnerable to iodine deficiency disorders (Joint and Organization, 2005; Secretariat et al., 2007). It is recommended that dietary intakes of iodine for toddlers and lactating women in areas of iodine deficiency should be 15 ug/kg/day and 3.5 ug/kg/day respectively compared to 4 ug/kg/day and 2 ug/kg/day for a 7-12 years old and adolescent/adult respectively (Joint and Organization, 2005). Mother's iodine status is very crucial source of meeting their infants' iodine status via breast milk (Secretariat et al., 2007).

This study had two aims: 1) to investigate the prevalence of iodine deficiency in geographically and regionally vulnerable women of reproductive age in the Upper East region in Ghana and 2) to investigate if the household iodized salt supply along with community-based feeding native *Hibiscus Sabdariffa* leaves meal (HSM) can improve iodine status of child-bearing age women and their children (6-24mo) during dry/lean season.

5.3 Subjects and methods

5.3.1 Study design

The design of this study was a quasi-experimental, community-based, 12-week feeding trial with the primary focus of this paper being iodine status of dyads, determined by urinary iodine concentration (UIC).

5.3.2 Study site

This study was carried out in two districts. The districts are from the Upper East Region in Ghana (Fig 4 and Fig 5): Kassena Nankana West (KNWD) and Builsa North (BND) districts (details of districts published elsewhere). The study sites were narrowed down to two communities (Sakaa and Chania) in KNWD and three communities (Chuchuliga-yipaala, Azoayeri, and Awulansa) in BND.

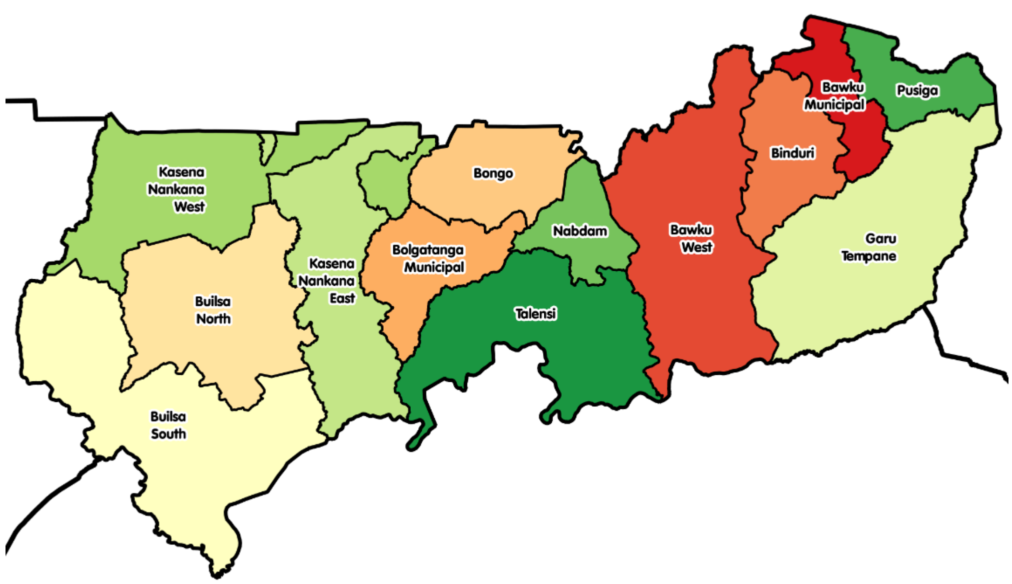

Figure 4. A map of Upper East region showing its districts

5.3.3 Study subjects and selection

We recruited women who were between 15-49 yrs, have a child (6 – 23 months), a member of an existing community-based women group or willing to join one in their respective communities, willing to participate and available throughout our study period (May – September, 2016). These women were part of a feeding trial study as well as a dry season container garden project.

Dyads were drawn from selected districts using community-based birth registers at community health centers kept by community health nurses stationed in these communities or by community health volunteers. Health volunteers are community members who are often not paid. Announcements (through the community chiefs/leaders) were then made in the respective communities for all women with children under 5 years of age to meet at their respective community health centers.

Research team briefed the women and the community leaders on the study. Names that were shortlisted from the birth registers were read out. Dyads were contacted individually (including dyads whose names were not in register but were obtained from the health volunteers who knew almost everybody in their catchment areas) to check on their willingness to be part of the study through the community health volunteers and community health nurses. Finally, verbal consent was sought from spouses of the women who were willing to be part of the study.

A total of 120 dyads (Intervention - 60 mother/child dyads; control - 60 mother/child dyads) were drawn from the two districts agreed to participate in the study (Fig 2). Dyads were assigned at the community levels to minimize contamination of the information at the individual level. During the study, four dyads relocated from intervention group; two before baseline data collection and two relocated in the 6^{th} week. There was no replacement made in these instances. All relocations were either to join a spouse or the entire nuclear family were migrating for farming purposes.

5.3.4 Intervention

The participating dyads in intervention communities were provided weekly supply of iodized salt for household use, and invited to consume veo soup/meal three times a week. The veo soup/meal is a local Ghanaian soup/meal mainly made of *hibiscus sabdariffa* leaves. Recipe for the meal was 18 kg of leaves / 52.5 kg of veo meal (wet 18 kg), 8 kg of groundnut / 52.5 kg of veo meal, 1.1 kg of dawadawa / 52.5 kg of veo meal, 3 kg of dried fish / 52.5 kg of veo meal, 0.045 kg of iodized salt / 52.5 kg of veo meal. The main source of iodine is the iodized salt in the meal. The meal was used to ensure compliance in consumption of iodized salt. The women and children were given two separate bowls, allowing them to eat and feed the children as much as they wanted. The women and children consumed on average 1.9 kg/day and 0.4 kg/day respectively of the meal, translating into 1.6 g/day and 0.3 g/day of iodized salt for women and children respectively. This excludes home consumption.

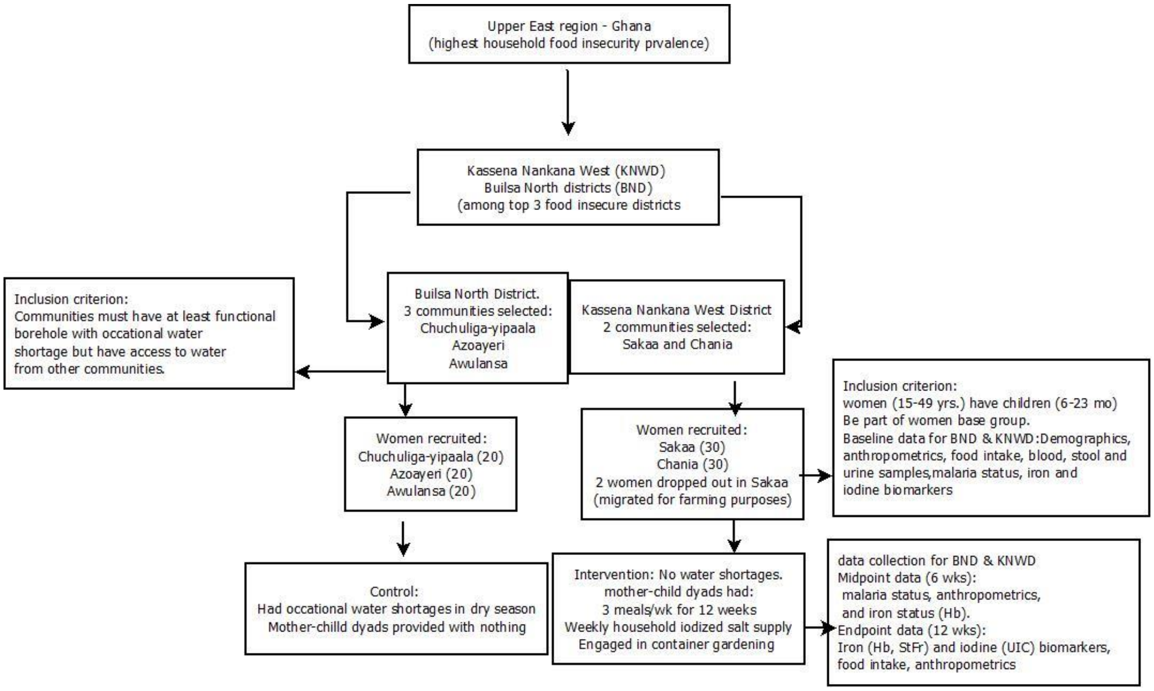

Figure 5. Study design

5.3.5 Measurements and data collection

Questionnaire/interview: At baseline, information on sociodemographic characteristics and food intake was collected. Household iodized salt samples provided by participants were tested at baseline with rapid test kit for the presence of iodine. Spot urine samples of participants were taken in labeled plastic bottles at baseline (0 wks) and at endpoint (12 wks). Investigations were carried out following the rules of the Declaration of Helsinki of 1975. Ethical approval was obtained from Michigan State University’s Ethical Review Board (IRB) as well as Navrongo Health Research Center’s IRB in Ghana.

Anthropometry: Participants weights and heights were measured at baseline, midpoint, and at end point according to standard procedures (Cogill, 2003). Electronic scale (UNIscale; Seca) was used to measure weights to the nearest 0.1 kg. Scale was calibrated using a known weight on days' measurements were taken

Iodine status assessment: WHO provides two criteria for assessing iodine status in populations: use of median UIC of spot urine samples and proportion of the total healthy population with UIC below and above 50 ug/L. The median UIC of <100ug/L in lactating women and children <2yrs IC indicates inadequate iodine intake with no other categories of iodine intake defined (World Health Organization, 2007). The total healthy population with UIC <50 ug/L is more than 20% indicates inadequate iodine intake of the population investigated. We assessed the iodine status of the study population using median UICs and also by the prevalence of <50 ug/L UIC. UIC was analyzed using the Sandell-Kolthoff (acid-digestion) reaction (Hedayati et al., 2011; Hedayati et al., 2007) and results were expressed as micrograms of iodine per liter.

Covariates

The study subjects were reproductive age women (15-49 yrs) with their children (6-23 mo). Sociodemographic and behavioral variables included household usage of iodized salt, age of woman (yrs), height of woman (cm), education of women, occupation of women, ethnicity, religion, malaria status, anemia status, household head sex, household head's age, wealth index, number of household adults, age of children, and number of children < 5 yrs.

5.3.6 Statistical Analysis

Data analyses were conducted using SAS (version 9.4, SAS Institute Inc., Cary, NC). Characteristics of participants were described using frequency distributions. Comparison of participants' characteristics between groups was done using chi-square statistics and student t-test for categorical and continuous variables respectively. We estimated and compared median UICs of both groups. Additionally, prevalence for <50 ug/L UIC between intervention and control groups were estimated to assess iodine status in relation to treatment.

5.4 Results

The subjects of this study were 118 women (15-49 yrs) and their children (6-23 mo) dyads. We had missing UIC values due to samples not having sufficient volume for analysis or containers being empty upon arrival at the Noguchi Iodine Laboratory-Ghana, so we checked to see if these missing values were evenly distributed in both groups using the χ^2– test (Baseline: χ^2=1.60, P = 0.2057; Endpoint: χ2=1.14, P=0.7130) for women's data. We further checked to see whether individuals with missing UIC had different characteristics (wealth index, education, and hemoglobin levels) as compared to those without missing data. Wealth index and education were of particular interest as they are key determinants of iodized salt use in other studies. Sociodemographic characteristics, anemia, UIC, and malaria status were obtained from the participants as indicated in Table 15 and Table 16. At the baseline, the intervention group did not differ significantly from control group, except for occupation (p=0.0007), religion (p<0.0001), ethnicity (p<0.0001) of women, household iodized salt usage (p=0.002), iodine level in household salt (p=0.019), anemia in children (p=0.021), and UIC<50 ug/l in children (p=0.021).

The majority of households (81% of intervention and 98% of control group) did not use iodized salt (0 ppm). Upon adequate consumption of iodine (salt iodine content≥50 ug/l), 8.8% and 0.0% of households consumed salt with adequate iodine amounts in intervention and control groups respectively. In our study population, 44.8% and 29.7% of the women had UIC< 50ug/l in intervention and control groups, respectively (Table 16). Among the children, 15.6% and 41.2% had UIC<50 ug/l in intervention and control groups, respectively. There was no significant difference in the proportion of women with UIC<50 ug/l between groups at baseline (p=0.2057). At the endpoint, there was a significant difference (p=0.0073) in the proportion of women with UIC UIC<50 ug/l between groups (Table 17). In children, there was a significant difference in the proportion of those with UIC<50 ug/l between groups at baseline (p=0.0219) and no significant difference at endpoint (0.0670).

At baseline for women, both median UIC values for intervention (57.4 ug/l) and control group (65.1 ug/l) were below the recommended UIC value of 100 ug/l with no significant differences between the two groups (p=0.2778). At the endpoint, median UIC for the intervention group (123.6 ug/l) was significantly higher (p=0.008) than the control group (59.7 ug/l). Similarly, in children, there was no significant difference between the median UICs of the two groups (p=0.1425) at baseline but there was a significant difference at the endpoint (p=0.009). Both groups had median UIC values higher than their mothers. However, median UIC values in both the control and intervention groups experienced an increment with time. Correlation between UIC of women and their children was not statistically significant at baseline (r=0.25, p=0.123) or endpoint (r=0.21, p=0.159) (data not shown).

Table 14. Demographic characteristics of child-bearing age women (15-49 yrs) and their children (6-23 mo) in intervention and control groups at baseline

Variable		Intervention		Control		
		n	Mean ± SD	n	Mean ± SD	P-value
Women age (yrs)		58	26.7±6.7	60	26.1±5.4	0.598
Children age (months)		59	14.1±5.1	62	12.6±5.2	0.119
Household head's age		58	47.0±16.9	60	42.7±15.1	0.149
Number of household adults		58	4.6±3.3	60	3.7±1.4	0.051
# Siblings in household		59	1.4±1.6	62	1.9±1.5	0.092
# children < 5 yrs		58	1.0±1.2	60	1.1±1.2	0.708
Variable		n	%	n	%	X^2(p-value)
Occupation of women	Farmer	31	53.5	50	83.3	15.3 (0.002)
	Handy works	5	8.6	5	8.3	
	Housewife	6	10.3	2	3.3	
	Trader	16	27.6	3	5.0	
Household head sex	Male	54	93.1	56	93.3	0.00 (0.960)
	Female	4	6.9	4	3.4	
Wealth index	Lower	19	32.8	20	33.3	0.02 (0.991)
	Middle	20	34.5	20	33.3	
	Upper	19	32.7	20	33.3	
Decision making	Low	11	20.0	9	15.0	1.2 (0.545)
	Average	27	46.6	34	56.7	
	Above	20	34.5	17	28.3	
Household iodize salt	Non-users	46	80.7	59	98.3	9.8 (0.002)
	Users	11	19.3	1	1.7	
Iodine level in household salt	<15ppm	52	91.2	60	100.0	5.5 (0.019)
	≥15ppm	5	8.8	0	0.0	
Ethnicity of women	Builsa	2	3.4	56	90.3	92.3 (<.000)
	Kassena	55	93.2	5	8.1	
	Nakani/Fulani	2	3.4	1	1.6	
Religion	Christian	34	57.6	60	96.8	27.0 (<.000)
	Muslim	9	15.3	0	0.0	
	Traditionalist	16	27.1	2	3.2	

Traditionalist: Practitioners of African Traditional Religion; Decision making: the degree in which women participate in household decision making. Wealth index: Household cumulative living standard measured by using household assets

Table 15. Baseline health indicators of child bearing age women (15-49 yrs) and their children (6-23 mo) in intervention and control groups

		Intervention			Control			
Variable		n	Mean± SD	Median	n	Mean± SD	Median	P-value
WOMEN								
height (cm)		58	160.3±6.6		60	161.7±5.6		0.217
weight (kg)		58	57.1±9.7		60	57.3±7.6		0.888
BMI		58	22.2±3.6		60	21.9±2.3		0.537
Hb (g/dl)		58	11.8±1.3		60	11.8±1.2		0.775
sTfr (ug/l)		57	5.1±3.1		60	4.7±1.6		0.320
UIC (Median-ug/l)		29		57.4	37		65.1	
CHILDREN								
HAZ		59	0.3±1.3		62	-0.3±1.3		0.009
WHZ		59	-1.2±1.3		62	-0.9±1.3		0.294
Hb (g/dl)		59	9.4±1.7		62	9.0±1.3		0.171
sTfr (ug/l)		58	9.1±5.1		62	9.7±5.3		0.528
UIC (Median-ug/l)		32		150.2	34		90.4	
		n	%		n	%		X^2 p-value
WOMEN								
UIC (ug/l)	<50	13	44.8		11	29.7		1.6
	≥50	16	55.2		26	70.3		0.205
Malaria infection	Negative	51	87.9		55	91.7		0.5
	Positive	7	12.1		5	8.3		0.502
sTfr (ug/l)	≤4.40	29	50.9		32	53.3		0.1
	>4.40	28	49.1		28	46.7		0.790
Hb (g/dl))	≥12	31	53.5		25	41.7		1.6
	<12	27	46.5		35	58.3		0.200
CHILDREN								
UIC (ug/l)	<50	5	15.6		14	41.2		5.2
	≥50	27	84.4		20	58.8		0.021
malaria infection	Negative	55	93.2		55	90.2		0.4
	Positive	4	6.8		6	9.8		0.545
(sTfr)	≤2.85ug/l	2	3.5		1	1.6		0.4
	>2.85ug/l	56	96.6		61	98.4		0.520
Anemia (Hb)	≥11g/dl	9	15.2		2	3.2		5.3
	<11g/dl	50	84.8		60	96.8		0.021
Stunting (HAZ)	>-2SD	3	5.08		7	11.29		1.54
	<-2SD	56	94.92		55	88.71		0.2153

Hb: Hemoglobin, Stfr: Serum transferrin receptor, UIC: Urinary iodine concentration; Anemia and iron deficiency defined by Hb <12g/dl and stfr <4.4ug/l respectively in women and Hb<11g/dl and stfr>2.85ug/l, respectively in toddlers; Iodine deficiency defined by UIC <50ug/l. HAZ: height-for-age z-score; WHZ: weight-for-age z-score;

Table 16. Median UIC and distribution of UIC of childbearing age women (15-49yrs) and their children (6-23mo) in both intervention and control groups across time

Time line	Status	Intervention n	Intervention %	Intervention Median	Intervention P-value*	Control n	Control %	Control Median	Control P-value*	χ^2	+P-Value
Women											
Baseline	UIC ug/l	29	43.9	57.4	<0.0001	37	56.1	65.1	0.172		0.085
Endpoint		49	48.0	123.6		53	52.0	59.7			0.001
Baseline	UIC<50 ug/l	13	44.8		<0.0001	11	29.7		0.842	1.6	0.206
	UIC≥50 ug/l	16	55.2			26	70.3				
Endpoint	UIC<50 ug/l	5	10.2			17	32.1			7.2	0.007
	UIC≥50 ug/l	44	89.8			36	67.9				
Children											
Baseline	UIC ug/l	32	48.5	150.2	0.0084	34	51.5	90.4	0.379		0.1425
Endpoint		29	54.7	198.0		24	45.3	104.1			0.009
Baseline	UIC<50 ug/l	5	15.6		0.2708	14	41.2		0.161	5.2	0.0219
	UIC≥50 ug/l	27	84.4			20	58.8				
Endpoint	UIC<50 ug/l	2	6.9			6	25.0			3.4	0.067
	UIC≥50 ug/l	27	93.1			18	75				

UIC: Urinary iodine concentration, χ^2: Chi – square test , UIC<50ug/l: *inadequate iodine status,* *P-value: within group comparison for chi-square test, +P-value: between groups comparison for chi-square test

5.5 Discussion

To the best of our knowledge, this is the first study that assessed iodine contents of household salt supply, utilized an indigenous Ghanaian meal as a vehicle for salt intake compliance to improving iodine status of childbearing age women (all were lactating women) and their children (6-24mo) in northern Ghana. Several studies have focused on the prevalence of iodine deficiencies (Cappuccio, 2014; Hess et al., 2016; Simpong et al., 2016).

The high proportion of household at baseline consuming non-iodized salt in intervention (81%) and control (98%) groups is alarming and are far higher than national levels (66%). Nearly all households in intervention (91%) and control (100%) groups consume inadequately iodized salt signifying inadequate iodine intake. It is not surprising that the women and children had severe iodine deficiency. On the flip side, it is possible iodine in salt might have been lost during storage or households may have alternative sources of iodine such as iodine fortified bouillon cubes (Abizari et al., 2016). This may raise a question as to whether iodized salt is still a key source of iodine for households in the inter lands. For this and other reasons, we carried out our intervention study.

We found that the weekly household iodized salt supply (450 g) and consumption of iodized salt (48.9 g/wk/adult and 10.2 g/wk/child) using HSM as a compliance channel significantly improved the iodine status of women in the intervention group. There was a significant increase in proportion of participants with UIC ≥50 ug/l in intervention group while a decline was observed in the control group (p=0.021). Median UIC for the intervention group was also significantly higher than the control group. Our findings indicate that decreased urinary iodine levels among lactating mothers does not necessarily imply lower iodine status for their breastfed children considering the observations in median UIC of the children. Though women had non-optimal iodine status, iodine status of their children was within or near optimal levels. A possible biological explanation for this observation could be due to compensatory mechanism in the mammary glands to provide iodine enriched milk for children (Nazeri et al., 2017). Though children had higher iodine status, it appears the compensatory mechanism in the mammary glands strives to keep milk iodine status within or near optimal levels (Secretariat et al., 2007).

Our findings suggest that iodized salt is a key source of iodine in the research settings. Additionally, it is an effective channel for the improvement of iodine status of economically disadvantaged groups living in remote areas in vulnerable locations due to geographical distance from coastal lands (Andersson et al., 2012). Our findings are in consonance with other clinical trial studies (Zhao et al., 1999) that show a positive improvement of median UIC by iodized salt use in children. A recent study published by Nazeri and colleagues (Nazeri et al., 2017) reported beneficial effect of iodine fortified foods in improving the median UIC of lactating women. Another study in northern Ghana (Abizari et al., 2016) reports the beneficial effect of iodine fortified bouillon cubes. The study further suggests that the bouillon cubes were the major source of iodine intake and that iodine adequacy was largely due to the bouillon cubes but not iodized salt. Their finding is contrary to what is observed in our control group. If bouillon cubes were a major source of iodine, decline in median UIC might have been unlikely.

According to our results, iodine deficiency in the region is within the classification for Ghana being among countries with mild iodine deficiency (Median UIC of 50 – 99 ug/l) for the period of 1993 – 2006 (De Benoist et al., 2008; World Health Organization, 2017). While this observation is worrying, improvement in median UIC is expected as a result of Ghana passing legislation to implement the universal salt iodization policy in 1996 (Nyumuah et al., 2012a). As there has been no improvement for a decade, this could be an indication of the ineffectiveness of policies and programs addressing iodine intake and iodine adequacy in the population for the last decade. This assertion is further corroborated by the high proportion of households (82%) not using iodized salt in the region. We suggest that activities and programs aligned with universal salt iodization should be revisited and assessed to ascertain the bottlenecks and challenges preventing the populations from attaining the intended benefits of the universal salt iodization adopted by Ghana.

This study is not without limitations and strengths. The strengths of our study were mainly: the inclusion of a control group for comparison, urinary iodine concentration measurements taken at baseline and endpoint. Using an indigenous meal to ensure compliance of iodine intake is the first of its kind in the research settings and further strengthen our findings. We recognized that not being able to estimate dietary iodine intakes could be a drawback to this study. However, iodized salt is apparently the main source of dietary iodine in northern Ghana. Our results are limited to population level interpretation as we recognized the limitation of a single spot UIC (König et al., 2011) for an individual’s iodine status.

5.6 Conclusion

To the best of our knowledge, this is the first study to assess the iodine status of women of reproductive age and their children in northern Ghana (baseline) and that supplying households with iodized salt and providing dyads with an indigenous Ghanaian meal containing iodized salt can improve their iodine status. We found a high prevalence of severe iodine deficiency (>20%). Our results suggest that iodized salt is an effective channel for the improvement of iodine status of economically disadvantaged groups living in remote areas in vulnerable locations due to geographical distance from coastal lands. Our findings further suggest that decreased urinary iodine levels (median UIC) among lactating mothers does not necessarily imply lower iodine status for their breastfed children. Further research is needed to ascertain the bottlenecks and challenges preventing populations from attaining the intended benefits of the universal salt iodization policy adopted by Ghana over two decades ago. Efficacy of USI has to be monitored because many factors such as cost, inadequate level of fortification, or loss of iodine during storage can alter the expected outcome in prevention of iodine deficiency in public.

Chapter Six: Container gardening to combat micronutrients deficiencies in mothers and young children during dry/lean season in northern Ghana

6.1 Abstract

Food insecurity is prevalent in northern Ghana, where the dry season stretches for 7-8 months (Oct-May) a year and the majority of inhabitants are subsistence farmers. We aimed to address inadequate dietary sources of iron and iodine in the dry/lean season by mother-child dyads by developing unconventional food production systems. Women groups with 6-24 mo old children (n=58) in two communities farmed 40 wooden containers for growing indigenous iron-rich (19.30 mg /100 g) for consumption and 15 containers for cash crop cabbage. produced from two harvest cycles/dry season and cash crops were adequate for three weekly community meals that improved iron and iodine status of the participating mother-child dyads. Future research is needed to expand the project to involve all households in the communities

6.2 Introduction

Food insecurity is a global challenge and its effects are felt across all continents (Ball, 2015; Headey, 2013). In 2016, global estimates indicate that 10.9% people are undernourished with uneven distributions from less than 5.0% in developed countries, 12.9% in developing countries to 20.0% on the Africa continent (Ball, 2015; Food and Agriculture Organization of the United Nations, 2015; Headey, 2013). Most food insecure people suffer from deficiencies of protein and essential micronutrients, such as iodine, vitamin A and iron (Smith et al., 2000). One of the most common nutritional deficiencies is anemia, with a global prevalence of 24.8%, which leads to detrimental impacts on health, social and economic development (Lopez et al., 2016; Modell and Darlison, 2008; Stevens et al., 2013; World Health Organization, 2001; 2015). About half of anemia is due to iron deficiency (Lopez et al., 2016; Stevens et al., 2013; World Health Organization, 2001). Women of reproductive age (30.2%) and young children (0-5 yrs) have the highest prevalence of anemia (Organization, 2008). In preschool-age children, the prevalence is 47.4% with the highest prevalence reported in Africa (67.6%) and South-East Asia (65.5%) (Stevens et al., 2013). Deficiencies of micronutrients (iron and iodine) affect maternal and child mortality, physical performance (Lopez et al., 2016), fetal growth (Caulfield et al., 2006; Strauss and Dietz, 1997), birth outcomes, stunting and impair cognitive development (Soemantri et al., 1985).

In African continent, the prevalence of anemia varies among countries (World Health Organization, 2015). In Ghana, a 2014 national health survey indicates that anemia was prevalent with 66% in children <5 yrs and 42% in women of 15-49 yrs (Ewusie et al., 2014; Ghana Statistical Service et al., 2015). The prevalence of anemia differed greatly (Ewusie et al., 2014; Ghana Statistical Service, 2012; Ghana Statistical Service et al., 2015; GSS, 2009) among different regions of Ghana: 82.1% in Northern region vs. 53.7% in Ashanti region in southern Ghana (GSS, 2014). Currently, no national/regional data are available on the prevalence of iodine deficiencies assessed by biomarkers. Iodized salt use at the household level, which is the proxy for iodine status, has been low with 61% at the national level (Ghana Statistical Service et al., 2015) and 58% in northern region in 2014.

The root causes of anemia are inadequate intakes of dietary iron, poor bioavailability of dietary iron, deficiency of other micronutrients (folate, or vitamin B12) and/or diseases/infections (thalassemia, malaria, HIV infection, hookworm infection and schistosomiasis) (Stevens et al., 2013). Although inadequate iron in the diet is the major cause of anemia, little has been done in improving the accessibility of iron-rich food sources for the vulnerable groups, particularly in the peak of food insecurity, during dry/lean season in most parts of Africa including Ghana. The major source of iron intake in Africa is plant-based (Zimmermann et al., 2005) (dark green vegetables such as hibiscus, beans, peas etc). Seasonality affects availability and consumption of iron-rich dark green vegetables as they are readily available in the wet season (Smith and Eyzaguirre, 2007), but not in the dry season .

In Ghana, the gravest impact is felt in northern Ghana during the dry/lean season (Oct- May) peaking in June/July (WFP, 2012). Inhabitants generally consume monotonous diets consisting of staple cereals and limited choices and amounts of vegetables with little or no animal products. The seasonal effects decrease the quantity and frequency of consumption, which is a key regional source of iron. The consequential impact of seasonality on food availability and intake of iron is most prominent in reproductive-age women and their young children. The major causes of nutritional insecurity in the dry/lean season stem from a number of reasons. Northern Ghana has a typical savannah woodland vegetation characterized by short, scattered drought-resistant trees and grass with a 7-8 mo dry season (Oct–May) and a 4-5 mo rainy season (June–September). It is extremely difficult to cultivate crops in the dry season as few households have access to the irrigation sites for dry season crop cultivation. Most inhabitants rely largely on the unpredictable, short-lived rainfall for cultivation.

The goal of this study was to develop innovative and unconventional food production systems that could be implemented during the dry season in places where land with irrigation systems is not available to produce food sources rich in micronutrients. It is important to explore alternative, nontraditional routes (Masset et al., 2012; van den Bold et al., 2015) to produce nutrient-rich vegetables during the dry season in northern Ghana to address access to quality food. We specifically aimed to investigate 1) the feasibility of producing widely consumed iron-rich vegetable, *Hibiscus Sabdariffa*, during the dry/lean season using containers, 2) feasibility of producing adequate amount of for three weekly meals by the mother-child dyads during the dry/lean season and 3) economic gains from cash crop cabbage production for purchase of iodized salt, dry fish and sustenance of container garden activities. Container gardening has not been a conventional route for household food production in northern Ghana. In this study, specifically containers were chosen to overcome the constraints experienced by women groups in subsistence farming communities during the dry/lean season with limited lands/plots, water and resources.

6.3 Materials and methods

6.3.1 Study location, design, and subjects

This community-based trial sought to evaluate the feasibility of container gardening during the dry/lean season to produce iron-rich (19.30 mg /100 g) for consumption, and market-demanded cabbage for economic gains to purchase iodized salt, dry fish and garden supplies.

We selected to study in the Upper East region (Fig 6) which has the highest rate of food insecure households (GSS, 2009; Nyantakyi-Frimpong, 2013; WFP, 2012) and highest anemia prevalence (Ewusie et al., 2014; Ghana Statistical Service, 2012; Ghana Statistical Service et al., 2015; GSS, 2009) in Ghana. Kassena Nankana West district in the Upper East region was among the top five food insecure districts in the region. The communities selected had accessibility to water sources (presence of functional borehole) during the dry season. We recruited women (15-49 yrs) with young children (6-23 mo of age) who belong to or were willing to join a community-based women's group. They participated in the community garden project between May-Sept 2016, which is the end of the dry season to the beginning of rainy season. These women also participated in a community feeding trial for three weekly lunch meals made from *Hibiscus Sabdariffa* over a period of 12 weeks.

A women group of each community (28 women in Sakaa and 30 women in Chania) received 20 wooden containers (L: 90cm, W: 40cm, H: 35cm) made by local artisans for production and 7-8 wooden containers (L: 90cm, W: 40cm, H: 40cm) for cabbage production. The containers were assembled in the respective communities' health centers and fenced. The common community location was chosen to (1) reach out to many women and help them to learn from each other, (2) help women market their wares cooperatively to increase their ability to become food-sufficient, (3) provide an opportunity for nutrition and health education sessions, and (4) market the approach to the entire community to facilitate easy adaptation when the approach is finally scaled up at household level for all households in the various communities.

The women group in each community also received a motorbike (tricycle) to help them carry vegetables for sale and to sustain container gardening. The women groups were also encouraged to use the motorbikes as means of public transportation to generate income for the group. We also guided the women groups on marketing skills, including recruitment of clients for their produce before production and harvest in the absence of storage facilities for the produce. We asked them to reach out to potential clients and secure their orders a week prior to harvesting cabbages from containers to minimize the loss and maximize the economic gains.

Figure 6. Map of Ghana indicating upper East region in dark yellow

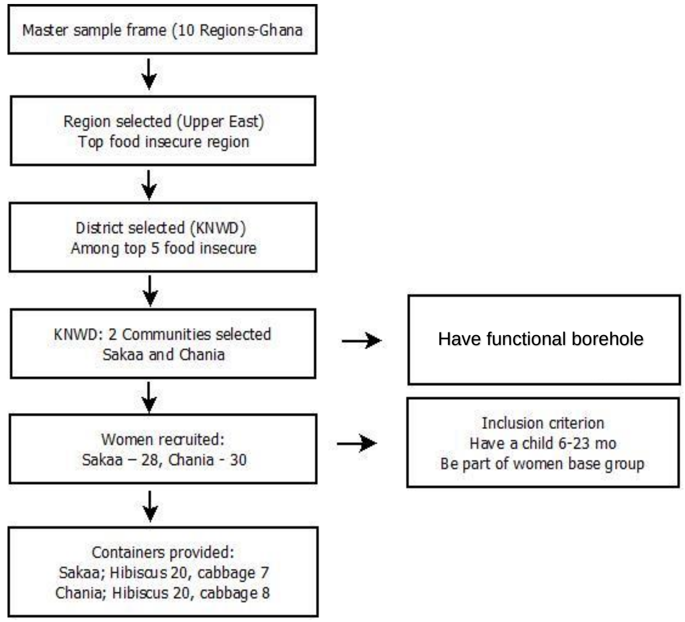

Figure 7. Project design

6.3.2 Cabbage and production

Cabbage seeds were nursed by an experienced community-based gardener three weeks before engaging the women group from each community. The two community groups of women started the gardening on same day by filling containers with a mixture of soil and household-made compost under the guidance of the gardener, sowing seeds and transplanting cabbage seedlings (Figs 8). All containers were then mulched with straw to prevent water loss and keep cool temperature within containers. Pieces of wood were placed on top of straws to prevent wind from carrying mulch away. The bottom of containers was sealed to prevent excessive water loses. Mulch was removed in the later stages of the project as the rains set in.

was harvested four weeks (Fig 8) after sowing and the second sowing was carried out on the same day of the first harvesting. The second harvest took place four weeks later. Upon each harvest, leaves were plugged and weighed at the respective community. Cabbages were harvested between 14th and 15th weeks in response to orders received from recruited clients. The number of cabbage rolls were counted and sold. Labor cost was obtained from crude calculation using prevailing hourly wage (¢10/8hrs/day) for farm activities in the research areas.

6.3.3 Statistical Analysis

Data analyses were conducted using SAS (version 9.4, SAS Institute Inc., Cary, NC). We examined community and participants' characteristics using simple descriptive statistics.

Sowing of and transplanting cabbage seedlings

Sowing of and transplanting cabbage seedlings

Second cycle ready for harvesting in four wks.

Cabbage rolling up with second hibiscus planting session in its second week

Figure 8. Hibiscus Sabdariffa and cabbage production

6.4 Results

Table 17.Characteristics of communities and participants

Variable		N	%
Occupation of women	Farmer	31	53.4
	Handy works*	5	8.6
	House wife*	5	8.6
	Trader/Farmer*	17	29.3
Households' usage of iodize salt	Non-users	47	81.0
	Users	11	19.0
Iodine levels in salt	<15 ppm	53	91.4
	≥15 ppm	5	8.6
		N	Mean ± SD
Number of Adults/household		58	4.6±3.2
Number of children < 5 yrs/household		58	1.1±1.2

* Handy works comprises of hair dressers and seamstress, *Women who reported other forms of occupation also engaged in farming during the wet season.* <15 ppm indicates inadequate level of iodine in household salt.

Table 18. Vegetables yields from container gardening

Vegetable Community		Container (n)	*Yield 1 (kg)	Yield 2 (kg)	Total Yield (kg)
Hibiscus Sabdariffa	Sakaa	20	48	45.5	93.5
	Chania	20	34	33.3	67.3
			**Yield (rolls)	Income (¢)	Total income (¢)
Cabbage	Sakaa	7	75	375	665
	Chania	8	58	290	

*Harvested in the first 4 weeks and ^subsequent four weeks, ** Harvested in 14-15 weeks

Sociodemographic characteristics of women in each community is summarized in Table 18. All the women were engaged in farming in the wet season, though reported other forms of occupations. Only five households (less than 10%) of the households studied used iodized salt with adequate level of iodine suggested by WHO. Table 19 shows leaves (HSL) harvested in Sakaa community for the first and second harvesting session (46.7 kg and 45.5 kg, respectively) and in Chania community (33.6 kg and 33.3

kg, respectively). These yields are averaged to 2.0 kg/container/cycle. Cabbage rolls (75 and 58 heads, respectively) were harvested with a total seasonal market value of ¢375.00 and ¢290.00 in the Sakaa and Chania communities, respectively, with a mean of ¢44.3/container/cycle. In this project, the prices of cabbages sold had dipped as they were sold at the beginning of rainy season

6.5 Discussion

To the best of our knowledge, this is the first study to investigate the feasibility of producing iron-rich vegetables using container gardens in northern Ghana. Our aim was to address the challenges during the dry/lean season in northern Ghana with high rates of food insecurity, poor access to micronutrient-rich foods, limited irrigation lands and water. Most published studies focused on household production, yields and production from irrigated lands to meet food and nutritional needs of households in rural areas (Fox et al., 2005; Roncoli et al., 2001; Tontisirin et al., 2002). Our results demonstrated that producing iron-rich in the dry season in containers is feasible for households or communities with limited land and water. This could be one of the solutions to food insecurity and the shortcomings of conventional irrigation systems during dry season in northern Ghana. The major challenge for rural farmers in adopting this approach might be the initial cost (¢65/container, with life span of 4yrs), this eventually becomes cheaper in the long-term. The cost of containers could also be lowered by modifying container structures in the future.

Recently, efforts have been channeled towards smallholder farmer to ensure food security by expanding smallholder irrigation. This route may however be linked with environmental damage, such as groundwater depletion and wetlands destruction

(Burney et al., 2013; Rodell et al., 2009). Aside from container gardens, a solution to these challenges could be harvesting rain water (Fox et al., 2005) or recycling household waste water to use during dry season. We plan to employ these alternative approaches in the next phase of our research.

Our project further demonstrated that a container garden is capable of producing adequate amount of leaves to meet the mother-child dyad's consumption levels needed to improve iron status and, eventually, curtail anemia prevalence based on our feeding trial studies (unpublished). Mean mother-child dyad's meal intake was 2.3 kg which translates into 0.78 kg of leaves. With a mean yield of 2.01 kg HSL/cycle, one container yield is capable of providing approximately three meals/week for a dyad. On this basis, a dyad needs a minimum of four containers to have fresh HSL throughout the dry season when a strategic approach of sowing seeds in a systematic and rotational order is employed. Sowing should start in the first container in week one, then in the second container in week two. It should continue in that order to the fourth container. By close of week four, the first container would have been ready for harvesting since harvesting is done in every four weeks. For continuity, sowing should be done on the same day after harvesting. At close of week five, the second container would be ready for harvesting, sowing and harvesting should thus proceed in that pattern till the rains set in. With a mean time of 45 minutes' labor/week, this approach would generally make iron-rich staple available throughout the year for dietary iron especially in the dry/lean season. It is recognized that a household is not merely made up of mother-child dyad; the number of containers could be increased to factor in household size. When the number of containers are increased, the unit cost for production may be decreased but with a challenge of storage space for the containers during rainy season. We plan to design collapsible containers for easy packing and storage during rainy season.

Generating income for sustaining the project as well as acquiring micronutrient-rich foods (dry fish and iodized salt) from container gardens was well established. The mean cost of mother-child dyad's weekly dry fish/salt intake and weekly dyad's household iodized salt supply was ¢7.06 from our feeding trial data (unpublished). With an average income of ¢44.3 per container, the cost for fish and iodized salt could be met while saving to sustain garden activities. Our findings are in an agreement with those reported by Sinyolo et al (Sinyolo et al., 2014). The authors indicated that access to irrigation enabled farmers to practice double cropping and to grow such crops as cabbages and potatoes for high dividends (Sinyolo et al., 2014). This project demonstrates a great potential of filling the gap by providing iodized salt to the majority of households (91.4%), which do not used adequately iodized salt with reasons being largely attributed to cost. In our study, half of the income is expected to be used for food items and the other half reserved as savings to take care of purchasing of seeds (¢5) and fixing broken containers. Labor cost (¢0.94/community/week) was not budgeted in this study, as the women would not have been engaged in labor market activities without the garden activities. Additionally, leaves harvested for household consumption compensates women's time in the garden.

Income margin accrued from cabbage production could be made larger by increasing the number containers per woman and also if cabbages are sold at the peak price. The containers are expected to last for at least four years, as they are used only in the dry season and stored during the rainy season. In the event that containers are no longer usable, they are simply recycled as compost for the growing vegetables. An obvious and legitimate question with this approach is how to ensure that money meant for food is used as such. As an inclusion criterion in this study, women had to belong to or were willing to join existing community-based women’s groups in their respective communities. Purchases can thus be done as a group, which can help to support and monitor group members.

All the women included in our project were engaged in farming in the wet season. This is in consonance with other research works in which most women in Africa are engaged in some forms of agricultural labor (Bryson, 1981; Flora, 1985; McGowan, 2011). Although some of the women in our study reported other forms of occupations, such as trader and handy works, these trades occurred mainly on market days, signifying noninterference with our project. Building on women’s experiences in the wet season farming, acceptance of our novel approach in the communities was not a challenge; however, what seems interesting was the skeptism of the cabbage and to grow in the containers. The end result was that of amusement and excitement among women participants. By providing women involved in the project with the tools they need to grow more and better food and market their wares cooperatively, we increased their ability to become self-sufficient with foods. The project relies on giving women the tools they need without requiring any payback, since microcredit has been a sustainable means to empower the poor. The economic model relies on saving part of the proceeds from food sales to financially support the project to make it sustainable over time. We take cognizance that income may not be only used for nutritional inputs as such guidance through appropriate nutrition education and policy would be necessary in large scale implementation.

The strengths of our study were introduction of innovative approach for production and making micronutrient-rich foods available in the peak of food insecurity period, helping women work in groups to learn from each other, aiding women to market their produce cooperatively to increase their ability to become food sufficient, providing an opportunity for nutrition and health education sessions, and engaging the entire community via project positioning. This study is not without limitations/challenges, however. The key challenges range from timing of the planting, soil quality and productivity. We were not able to capture the full spectrum of these factors during the full dry season. We could not draw conclusions if variations in weather across the dry season period could have affected yields. Containers were designed to use as little amount of water as possible. However, in rainy season, water accumulated in containers can affect the second cycle yields of *Hibiscus Sabdariffa*. We also learned that ashes added to containers in Chania community resulted in reduced yields of cabbage that thrive in acidic soil for optimal growth.

6.6 Conclusion

We demonstrated feasibility of using container gardens for women groups to produce indigenous iron-rich vegetable, *Hibiscus Sabdariffa*, during the dry/lean season for consumption, and economic gains from cabbage production. This innovative approach challenges food and nutritional insecurity in the dry/lean season. Our approach allows women groups with limited irrigated lands and water in subsistence farming communities to produce vegetables during the dry season to improve nutritional insecurity. No single intervention package is expected to tackle food and nutrition insecurity in a comprehensive manner. Our intervention could be improved by trials including all households in entire communities, and incorporation of community poultry raising or animal husbandry in a similar fashion to ensure access to quality animal source food and economic gains, hopefully led by women groups in communities.

Chapter Seven: Conclusion

7.1 Conclusion

Food security does not necessarily lead to nutrition security though is needed to ensure nutrition security. As such, nutrition policies directed at ensuring nutrition security or improved trace mineral status should not solely target increasing food production and/or food availability but be guided by or incorporate appropriate nutrition education. We found that food consumption (measured by DDS and DP) was associated with the risk of iron and iodine deficiencies in childbearing age women but not in their toddlers after controlling for sociodemographic characteristics during the dry/lean season in northern Ghana. We further found that consuming thrice a week improved meals prepared with native *Hibiscus Sabdariffa* leaves over 12 weeks was efficacious in improving iron status of women and reducing stunting in toddlers during dry/lean season. Weekly supply of household iodized salt also improved iodine status of mother-child dyads. Finally, we demonstrated that container garden approach can produce adequate amount of iron-rich *Hibiscus Sabdariffa* for mother-child dyads consumption and cabbage as a cash crop to purchase ingredients for preparation of the meals, i.e., iodized salt and dry fish.

7.2 Implications

Very importantly we demonstrated: efficacy of food-to-food fortification to improve iron status, DDS predicting iron and iodine status when the minimum amount was considered to be counted, universal salt iodization program has not yet been fully implemented in northern Ghana in terms of its universal availability, the level of iodine in salt, and monitoring of iodine status at the community levels, and container garden can be implemented during dry/lean season.

The use of DDS and DP to predict iron and iodine biomarkers could serve as basis for non-invasive screening for early detection of iron and iodine deficiencies. Additionally, our study could serve as the basis for further research into the development of dietary screening tools for micronutrients in Ghana.

Our results that meals prepared with indigenous *Hibiscus Sabdariffa* leaves in improving iron status of women of child-bearing age and in reducing stunting among toddlers during the dry/lean season points to a need to orient policies and programs to center on local environment and local solutions. It further implies the need for further research on the efficacy of indigenous foods on micronutrients deficiencies. The impact of weekly household iodized salt supply in improving iodine status points to the assertion that iodized salt is the main source of dietary iodine in northern Ghana, a need to make iodized salt accessible to households coupled with nutrition education could lead to the needed and desired public health outcomes.

The introduction of innovative approach for production and making micronutrient-rich foods available in the peak of long food insecurity period shows another dimension of tackling nutrition insecurity. Helping women work in groups to learn from each other, aiding women to market their produce cooperatively to increase their ability to become food sufficient, providing an opportunity for nutrition and health education sessions, and engaging the entire community via project positioning points to a need to reorient policies and programs targeted at reducing iron and iodine deficiencies.

7.3 Recommendations for future research

All those major implications can be introduced and modified until all can be sustained and maternal and child health is improved. A bioavailability study needs to be planned. Other indigenous crops and produce that are rich in problematic nutrients should be explored.

The persistent and alarmingly high rates of micronutrient deficiencies in northern Ghana is detrimental to maternal and child health. Several approaches have been used in the past by Ghana Health Service, non-governmental, and multinational organizations, and yet the problems persist. Through our current study, we uncovered that DDS and DP can be a non-invasive screening tool to predict the biochemical indicators of iron and iodine during long dry/lean season in northern Ghana. Further research is needed to factor in the quantities of various foods consumed.

Our findings on the efficacy of hibiscus sabdariffa meal in improving iron status and linear growth of toddlers over 12 wks intervention is promising. We however recognize the need for further research on the efficacy of the *hibiscus sabdariffa* meal to improve iron status of nutritionally vulnerable mother and child dyads, and linear growth of young children in much longer duration and in some larger communities. Measurement of various markers of iron, inflammation, and parasites (hook/tape worm) infections should be included in future studies to prevent confounding effects. We further recommend bioavailability studies on the *hibiscus sabdariffa* meal to elucidate the exact role and impact of the meal in improving iron status.

In making iron and iodine rich foods available during periods of food insecurity, we recommend that the container garden concept be repeated to capture the full spectrum of the dry season. Additionally, the impact of variations in weather across the dry season period on yield should be assessed.

APPENDIX

Michigan State University
Department of Food Science and Human Nutrition

Community interventions to improve iron and iodine status in mother and child dyads in northern Ghana

Questionnaire ID………………

Background information

1. Name of interviewer……………… 2. Date …………… 3. Community ………… 4. Mother's ID………………

5. Age of mother (yrs) ……………. 6. Weight of Mother (kg)…………... 7. Height of mother (m) …………

8. Mother Hb (g/l) ……… sFe (ug/l) ……….sTfR (nmol/l) ..…… 9. Religion………… 10. Ethnicity……………….

9. Education: a. none ☐ b. Primary ☐ c. Secondary ☐ d. college/tertiary ☐

10. Mother's occupation……………………………. *Mother's malaria status……………………………..

Marital status: a. married ☐ b. Single ☐ c. divorced/separated ☐

11. Child's age (months) ………… 12. Child's height (m) ………… 13. Child's weight (kg)…………

14. Child's Hb (g/l) ……………… sFe (ug/l)……… sTfR (nmol/l) ……

Number of siblings…………….. *Child's malaria status…………….

Household characteristics

15. Sex of household head ………… 16. Age of Household head ………… 17. No of adults in household ……

18. Number of Children <5yrs ………………… 19. Building type ………………… 20. Source of water ………………

21. Type of toilet……………………… 22 Lives with animals in same yard ……..…………………..

23. Does your household has any of the under listed items? (Please tick in check box when present)

a. Bicycle ☐ b. Motorbike ☐ c. Clock ☐ d. Radio ☐ e. Sewing Machine ☐ f. Bed ☐ g. Table ☐
h. Cabinet/Cupboard ☐ i. Mobile Phone ☐ j. Refrigerator ☐ k. Generator/Invertor ☐ l. Television ☐
m. Video Deck ☐ n. Dvd/Vcd ☐ o. Electricity ☐ p. Washing Machine ☐ q. Computer ☐
r. Digital Camera ☐ s. Non-Digital Camera ☐ t. car ☐ u. tractor ☐

18. Does this household own the under listed (If yes to any, input the corresponding numbers in the 1st column – N*)

	N*	1	2	3
a. Cattle				
b. Goat				
a. Sheep				
b. Pigs				
c. Rabits				
d. Grasscutter				
e. Chicken				
f. Guinea fowls				
g. other poultry				

N* = input number of animals, 1 = none, 2 = more than 95, 3 = don't know

Women empowerment

19. Who usually

	1	2	3
a. decides how the money you earn will be used?			
b. decides how your husband's/partner's earnings will be used?			
c. makes decisions about health care for yourself			
d. makes decisions about making major household purchases?			
e. makes decisions about making purchases for daily household needs?			
f. makes decisions about visits to your family or relatives?			
g. makes decisions about how many children to have?			

Key: 1 = you, 2 = partner, 3 = both

20. Who earns more money in your household? a. you ☐ b. partner ☐ c. about same ☐

24 HR Recall Questionnaire

Option A

I would like to ask you about liquids or foods that (NAME……...)/you may have had yesterday during the day or at night. I am interested in whether your child/you had the item even if it was combined with other foods.

Did (NAME ………...)/you drink (eat)	Child					Mother				
	Yes	No	DK	Freq	Qty (g)	Yes	No	DK	Freq	Qty (g)
1. Milk such as tinned, powdered, or fresh animal milk?										
2. Tea or coffee?										
3. Any other liquids (juice, cocoa)?										
4. Bread, rice, noodles, or other foods made from grains (kenkey, banku, koko,tuo zaafi, akple, weanimix)?										
5. Pumpkin, red or yellow yams, carrots, sweet potatoes that are yellow or orange inside?										
6. White potatoes, white yams, manioc, cassava, cocoyam, fufu or any other foods made from roots, tubers or plantain?										
7. Any dark green, leafy vegetables (kontomire, aleefu, ayoyo, kale, cassava leaves)?										
8. Ripe mangoes, pawpaw?										
9. Any other fruits or vegetables (e.g. bananas, avocados, tomatoes, oranges, apples)?										
10. Liver, kidney, heart or other organ meats?										
11. Any meat, such as beef, pork, lamb, goat, chicken, or duck?										
12. Eggs?										
13. Fresh or dried fish or shellfish (e.g. prawn, lobster)?										
14. Any foods made from beans, peas, lentils, or nuts?										
15. Cheese, yogurt or other milk products?										

16. Any oil, fats, or butter, or foods made with any of these?
17. Any sugary foods such as chocolates, sweets, candies, pastries, cakes, or biscuits?

Option A **procedure:** Ask for what has been consumed the previous day without mentioning food items to avoid prejudice. Tick as foods are mentioned, probe for frequency and usual quantities in the past 7 days. Probe for other foods consumed within the week, their frequencies and usual quantities consumed. Probe for alcohol consumption, smoking and substance abuse

Option B

24 hour recall **Day of the week: ……………………….**

Time / period of day	quantity of food eaten	Detailed food description	Source of food
1			
2			
3			
4			
5			
6			
7			

Probe for alcohol consumption, smoking and substance abuse

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