A randomized, double-blind, controlled trial in rural Burkina Faso to determine the optimal amount of zinc to include in small-quantity lipid-based nutrition supplements (iLiNS-Zinc)

Statistical analysis plan

Prepared for**:**
The International Lipid-based Nutrient Supplements-Zinc (iLiNS-Zinc) Project, Burkina Faso

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Version number	2
Version date	August 7th, 2014
Author	iLiNS-Zinc
Implementation date of current version	

Approved by	Name/Position	iLiNS site leaderships
	Signature	
	Date	
	Name/Position	
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	Name/Position	
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Version History Log

This table will detail the version history for this document. It will detail the key elements of the changes to the versions.

Version	Date implemented	Details of significant changes
1st	August 20th, 2013	
2nd		Addition of secondary outcomes. Minor corrections and updates to previously described analyses.

1. Introduction

The iLiNS-Zinc trial is a collaborative study designed and implemented by researchers of the Institute of Research in Health Sciences, Burkina Faso (Institut de Recherche en Sciences de la Santé – IRSS, BoboDioulasso) and the University of California, Davis, both of which are part of the International Lipid-based Nutrient Supplement (iLiNS) study group. The larger iLiNS project involves researchers from Burkina Faso, the United States, France, Malawi, Ghana and Finland whose goal is to expand the evidence base for using small-quantity lipid-based nutrient supplements (SQ-LNS) to prevent malnutrition in vulnerable populations. The study is investigator-initiated and funded by a grant from the Bill and Melinda Gates Foundation to the University of California. In Burkina Faso, the iLiNS-Zinc Project contributes to the options for community-based approaches to prevent malnutrition and stunting among young children in the intervention communities. Internationally, the results from iLiNS-Zinc study will be relevant to help develop the optimal amount of zinc to include in SQ-LNS formulations for young children. Primary data collection began in April 2010 and was finished in July 2012 and November 2012 for the nutrition and socio-economic data, respectively. The following sections describe the iLiNS-Zinc statistical analysis plan whilst data cleaning and analysis is still ongoing, before the identity of the study groups is unmasked.

2 Overview of study

2.1 The study

The iLiNS-Zinc study is a community-based, partially double-blind, placebo-controlled, randomized clinical trial which took place in the Dandé district in Burkina Faso. The District is home to more than 200,000 inhabitants; and has high prevalence of stunting and underweight of young children, poor food security, and endemic malaria transmission. Nine-month-old children were identified by periodic censuses in the study area. Communities selected after stratification by health clinic to ensure balance with respect to population, distance from a paved road, and distance from Bobo-Dioulasso in the target areas, were assigned to intervention or non-intervention cohorts. 2469 eligible children in the intervention cohort (IC) were randomly assigned to receive one of the following interventions from 9 to 18 months of age: 1) SQ-LNS without zinc and placebo tablet (LNS-Zn0), 2) SQ-LNS with 5 mg zinc and placebo tablet (LNS-Zn5), 3) SQ-LNS with 10 mg zinc and placebo tablet (LNS-Zn10), 4) SQ-LNS without zinc and 5 mg zinc tablet (LNS-ZnTab5). SQ-LNS and tablets were distributed weekly to provide 20 g of LNS and one tablet per day. Community-based treatment of malaria and diarrhea was provided free of charge during weekly household visits or when requested by the child caregiver, and confirmed by assessment of fever and/or malaria and reported diarrhea. Children in the non-intervention cohort (n=797) did not receive LNS, tablets or illness treatment during this period (NIC), but received LNS with 10 mg zinc, but no follow-up visits, from 18 to 27 months age.

2.2 Study population

During the initial census in the target communities of 32 villages, we screened (by means of a questionnaire) children who were less than 9 months and pregnant women in their last trimester. Children who were more than 10 months of age were excluded (not invited to the enrollment day). Additionally, to attain the target sample size, children in two more villages were invited to the enrollment day. All children who were 9 months of age were invited and enrolled in the study

if: 1) caregiver's written consent was provided, 2) all the inclusion criteria and none of the exclusion criteria were met (see 2.3 and 2.4); and 3) the child remained eligible after baseline medical, anthropometric and hematological tests were completed.
Enrolled children in the intervention cohort were randomly assigned to one of the four intervention groups.
Twins were both enrolled; however, only one of the twins will be randomly selected for inclusion in the data analysis.

2.3 Inclusion criteria

- Signed, informed consent from at least one caretaker
- Age 8.75 mo to 9.99 mo
- Permanent resident in the Dandé Health District, Burkina Faso
- Planned availability during the period of the study
- Residence within the health facility catchment areas
- Agreement to attend the study clinic for any febrile episode or other illness
- Acceptance of home visits

2.4 Exclusion criteria

- Hemoglobin <50 g/L
- Weight-for-height < 70% of the median NCHS reference.
- Presence of bipedal oedema
- Severe illness warranting hospital referral
- Congenital abnormalities potentially interfering with growth
- Chronic medical condition (e.g. malignancy) requiring frequent medical attention
- Known HIV-infected infant or mother
- History of allergy towards peanuts
- History of anaphylaxis or serious allergic reaction to any substance, requiring emergency medical care
- Concurrent participation in any other clinical trial

2.5 Objectives

The specific aim of the study is to assess zinc-related biochemical and functional responses among young Burkinabe children with a presumed high risk of zinc deficiency who received micronutrient products (lipid-based nutrient supplements (LNS) or zinc tablet) containing different amounts of zinc, provided with or between meals, and to compare the same outcomes among children who did or did not receive LNS, zinc tablet and selected health services.

The study had 3 **main objectives**:
a) To determine the optimal daily dose of zinc in SQ-LNS that promotes linear growth among 9- to 18-month-old infants in rural communities in Burkina Faso with high rates of stunting.
b) To determine the impact of different zinc doses in SQ-LNS on morbidity, micronutrient status, and neurobehavioral development.
c) To examine the extent to which household food insecurity and other individual, household, and village-level characteristics modify the effects of SQ-LNS on child outcomes.

The specific **hypotheses** for the trial were as follows:

a) Young Burkinabe children at risk of zinc deficiency who receive either 5 or 10 mg zinc per day in SQ-LNS or 5 mg zinc tablet per day provided between meals (and SQ-LNS to which no zinc has been added) have greater weight and length increments, decreased incidence of diarrhea and malaria, and a greater increment in plasma zinc concentration and fat-free mass compared with similar children who receive SQ-LNS that does not contain added zinc and a placebo tablet.
b) Children who receive SQ-LNS (with or without added zinc) or zinc tablets have greater physical growth (weight and/or length) compared with children in a non-intervention cohort.

2.6 Blinding

During data collection, all participants, field staff and investigators were blinded to the intervention group. The statistical analysis plan was written by the study investigators, all of whom are blinded to the group assignments, until after a consensus on the results and final conclusions of the primary outcomes were drawn by the study investigators.

2.7 Efficacy outcomes

2.7.1 Pre-primary outcomes

a) Presence of systemic inflammation as measured by acute phase response proteins (C-reactive protein (CRP) and alpha-1 glycoprotein (AGP))

2.7.2 Primary efficacy outcomes

a) Change in length and length-for-age Z-score (LAZ; based on WHO growth standards) between 9 and 18 mo of age
b) Change in weight and weight-for-age (WAZ), and weight-for-length (WLZ) Z-scores
c) Incidence and prevalence of diarrhea
d) Incidence of Rapid Diagnostic Test (RDT)
e) Change in plasma zinc concentration (PZC) adjusted for inflammation (measured in a subsample)
f) Change in body composition (fat mass and fat-free mass; measured in a subsample)

2.7.3 Secondary efficacy outcomes

Secondary outcomes include:

a) Adherence to the study supplements among children in the intervention cohort
b) Change in head circumference and mid-upper arm circumference
c) Prevalence of stunting, underweight, and wasting
d) Incidence of stunting, underweight, and wasting
e) Change in hemoglobin and micronutrient status at 18 months (iron status, measured by adjusted erythrocyte zinc protoporphyrin (ZPP), adjusted plasma ferritin (pF) and adjusted soluble transferrin receptor (sTfR); vitamin A status measured by adjusted retinol binding protein (RBP); and iodine status measured in a subsample of participants by urinary iodine (UI), dried blood spot thyroid stimulating hormone (TSH) and thyroxine (T_4), and plasma thyroglobulin (Tg) concentrations
f) Incidence of acute lower and upper respiratory tract infection (ALRI and AURI)
g) Neuro-behavioral development (acquisition of developmental milestones and more comprehensive analysis at the age of 18 mo)

2.8 Safety outcomes

Data were collected for adverse events during the intervention period from enrollment to 18 months of age in the intervention cohort. Adverse events were monitored by the field data collectors with a structured form (N07) at the weekly home visits (as reported by the caregiver). Additionally, severe adverse event (SAE) reports were filled in (N16a and N16b) for all hospitalisations or deaths of any of the participants in the intervention groups. Severe adverse events include all untoward medical occurrences that result in death, are life-threatening, require inpatient hospitalisation or prolongation of existing hospitalisation, result in persistent or significant disability, or are otherwise considered a serious medical condition by a study physician. Information on the causes of death were collected by a verbal autopsy method (N22).

2.8.1 Severe adverse events

Number of SAEs reported, i.e. if a subject has more than one occurrence of SAEs, all the SAEs will be counted and analyzed by group.

Conditions regarded as SAEs were:

a. Death of subject.
b. Hospitalization (at least 24-hour stay in the hospital because of illness).

2.8.2 Abnormally low anthropometric values

Abnormally low anthropometric outcomes were:

Percentage of children referred to a health center (Centre de Santé et de Promotion Sociale), i.e. with weight-for-length less than 70% of the median NCHS reference at 12, 15 (intervention cohort) or 18 months (all study children) of age.

2.8.3 Abnormally low hemoglobin values

Children with low hemoglobin values (<50 g/L) at 18 months who were referred to a health center (Centre de Santé et de Promotion Sociale).

2.9 Analysis principles

- The primary analysis will be by intention-to-treat. That is, results for all children enrolled (excluding one randomly selected twin per twin pair) will be analyzed according to the group to which they were assigned regardless of any protocol violations. Data on subjects, who were lost to follow-up because of death, travel from the study site, or refusal to continue with the study will be included in the analysis as available.
- For morbidity outcomes, data for children who were lost to follow up before 30 days will not be included in the analysis.
- All tests will be two-sided, at 5% level of significance.
- Where more than 10% of observations are missing for a dependent variable, we will report the number of observations used in the analysis.

3.0 Design

3.1 Sample size and power

The sample size estimates were based on the number of children needed in each group to detect (with a significance of P<0.05 and power >0.80) effect sizes that are consistent with the magnitude of effects observed in previous zinc supplementation trials. In particular, an effect size of >0.22 for diarrhea incidence, malaria incidence, and physical growth; and 0.6 for change in biochemical indicators of micronutrient status and change in fat-free body mass would be detectable. The estimated sample size for the non-intervention cohort was inflated for an assumed design effect of 1.5 due to the cluster sampling design and in all groups for an assumed attrition rate of 15%, except for the biochemical outcomes for which 20% attrition was assumed. A total of 3200 children (~267/month) were planned to be enrolled in the trial, of whom 468 (~38/mo) were to be selected for the biochemical studies and the body composition studies.

At the end of the study, a total of 3220 children were actually enrolled in the trial, of whom 701 were selected for the biochemical studies (see study 4.1. flowchart).

Iodine status was assessed in 3 out of 5 groups, i.e. LNS-Zn0, LNS-Zn10 and NIC. A total of 298 children in the three groups were randomized from the already randomly selected children in the biochemistry subgroup for the same effect size detection (Hess et al., 1999). Salt samples were collected in a sub-sample of the biochemical sub-group. The participants in the "salt sub-group" were randomly assigned by the statistician to have 20% and 50% from the participants in the IC and NIC, respectively (n=64 out of 320 in IC, and n=64 out of 128 in NIC). Assuming that approximately 15 – 20% of the mothers do not return with a salt sample, the sample size per type of intervention (IC vs. NIC) was approximately 50. The sample size estimates are based on the number of household salt samples needed in each type of intervention to detect (with a significance of p<0.05 and power >0.80) effect sizes that are consistent with the magnitude of differences observed in salt analyses of northern Côte d'Ivoire (Hess et al., 1999).

Table 1: Sample sizes for selected outcomes

Variable	Effect size	Unadjusted sample size*, treatment groups	Adjusted sample size*, treatment groups**	Total sample size, four treatment groups (intervention cohort)	Adjusted sample size, non-intervention cohort***	Total sample size, five groups
Weight, length Velocity	0.25 SD (0.17-0.35)[a]	383	451	1804	676	2480
Incidence of diarrhea	0.22 SD (~20% reduction)[a]	495	583	2332	--	--
Incidence of malaria	0.27 (~20% reduction)[b]	329	387	1548	--	--
Plasma zinc concentration	0.60 SD (0.6-0.8)[a]	68	85	340	128	468

Fat-free mass accrual [c]	0.60 [c]	68	85	340	128	468

*Sample size per group, five groups. **Adjusted for 15% or 20% attrition ***Adjusted for design effect (1.5) and 15% attrition.

[a] Data from two meta-analyses (Brown et al, 2002; Brown et al, 2009)

[b] Based on data from Bates et al, 1993 and Müller et al, 2001. Recent data from Burkina Faso showed 30% reduction in malaria incidence with both vitamin A and zinc (Zeba, 2008).

[c] Sample size based on discussion in Arsenault et al, 2008.

3.2 Informed consent issues

All potential participants received information about the study during their enrollment visit using a group discussion format. Those interested in participating were invited to a private discussion with study physician, during which the potential participants could ask additional questions about the study. One or both caregivers of every potentially eligible child wishing to participate signed or provided a thumbprint to indicate informed consent for the participation of their child prior to being enrolled. The consent procedure emphasized the voluntary nature of the study and the participants' right to discontinue follow-up at any point. When the caregiver was illiterate, an educated person from the same village read the consent form and witnessed the verbal explanation given by the educator. The witness observed the signature of the caregiver on the consent form and cosigned the form to confirm that the protocol was accurately explained.

Data on participants who refused to continue in the study will be included in the analysis on the intention-to-treat basis. However, the primary outcome data for such participants, if missing, will not be imputed.

3.3 Treatment cohorts

Target communities were stratified and assigned to intervention or non-intervention cohorts. A randomization list (ID from 1001 to 3200) was prepared by the study statistician for the intervention cohort. The list was randomized so that every concession had a 1/8 chance of receiving one of the eight color codes (two colors for each treatment group). Eligible children were assigned to the study group in the order of their enrollment. If a child belonged to an already participating concession (i.e. family compound), the child would be assigned the same study group (randomization at cluster level).

A second randomization which concerned the first child from every concession was done for the subgroups (biochemistry, 12-hour home observation, neuro-development (see Appendix B for neuro-development plan of analysis) and socio-economic status (SES)).

Combining the two colors that represent one group was done at the end of data collection, to facilitate the data cleaning.

3.4 Data collection and follow-up

The variables on which we collected data and the forms containing the variables are shown in Appendix A.

3.5 Interim data analysis

For most of the study outcome measures, interim analysis during the data collection period was performed. For anthropometric, hemoglobin, feeding practices, vaccination, breast feeding practices, behavioral development and laboratory testing for malaria outcomes, analysis was

done for 1) data cleaning purposes and 2) sharing data from the study area with the local health system. Indeed, a report detailing cross-sectional enrollment data (anemia prevalence, prevalence of positive rapid diagnostic tests for malaria, vaccination coverage, percent of children with moderate and severe malnutrition according to the Burkina National protocol [NCHS reference] and the WHO standards, and food frequency data) was shared every two months with the head of the Dandé Health District. Interim data analysis was not done by treatment group except for analyses of SAE (see 2.8.1).
Additionally, the principal investigators and the Institutional Review Board (IRB) at UC Davis-California and Centre Muraz-Burkina Faso were provided with a detailed report of the SAEs during the whole data collection period. In case of any evidence of a difference between treatment groups in hospitalization cases and/ or all-cause mortality, the PIs would have informed the Study Coordination Team and advised on the appropriate course of action.

3.6 Definition

3.6.1 Anthropometric z-scores

Anthropometric z-scores and the cut-offs will be defined according to the WHO 2006 Growth Standards (WHO Multicentre Growth Reference Study Group, 2006).

Table 2: Z-score' cut-offs for stunting, underweight wasting and other anthropometric outcomes

Term	Unit of measurement	Severe	Moderate	Moderate & severe
Stunting	Length-for-age z-score	< -3	-3≤ Z< -2	Z< -2
Underweight	Weight-for-age z-score	< -3	-3≤ Z< -2	Z< -2
Wasting	Weight-for-length	< -3	-3≤ Z< -2	Z< -2
Low MUAC for age	MUAC-for-age z-score	< -3	-3≤ Z< -2	Z< -2
Low head circumference for age	Head circumference-for-age z-score	< -3	-3≤ Z< -2	Z< -2

3.6.2 Micronutrient status

Most of the biochemical indicators measured in this study are acute phase proteins (APP), which increase or decrease during infection or inflammation. Measures of both CRP and AGP are needed for estimation of the effect of inflammation on PZC, ZPP, pF, sTfR and RBP concentrations and to correct their respective concentrations in the different groups (no elevated APPs [reference group], elevated CRP only, both AGP and CRP elevated and elevated AGP only) (Thurnham et al., 2010).

Table 3: Cut-offs of micronutrient deficiency

Term	Unit of measurement	Severe	Moderate	Mild	All grades combined
Anemia	Hemoglobin concentration (g/L)	< 70.0	>= 70.0 to < 90.0	>= 90.0 to < 110.0	< 110.0
Iron deficiency	Adjusted plasma ferritin (µg/L)				<12.0
Iron deficiency	Adjusted soluble plasma transferrin (mg/L)				>8.3
Iron deficiency	Adjusted zinc protoporphyrin in whole blood (µmol/mol heme)				>70.0
Vitamin A deficiency	Adjusted retinol binding protein (µmol/L)				<0.83[1]
Zinc deficiency	Adjusted plasma zinc concentration (µg/dL)				<65
Iodine deficiency	Urinary iodine (µg/L)				< 100.0
Subclinical hypothyroidism[2]	Thyroid simulating hormone (mU/L) and thyroxine (nU/L)				TSH >3.7 and normal T_4 (≥65)
Overt hypothyroidism[2]	Thyroid simulating hormone (mU/L) and thyroxine (nU/L)				TSH >3.7 and T_4 <65
Hypothyroxinemia[2]	Thyroxine (nU/L)				<65.0
Iodine status	Thyroglobulin (µg/mL)				n/a[3]

[1]Based on the cutoff corresponding to 0.70 µmol/L plasma retinol in 12-59 mo aged children in Cameroun (Engle-Stone et al., 2011). Analysis of plasma retinol in a subsample of 40 subjects at 9 and 18 mo from the current study will be carried out to derive and validate a specific cutoff of RBP for vitamin A deficiency in the study population.
[2]Bouhouch et al., 2014.
[3]No reference values for young children available. The results of the iLiNS-Zinc study participants will be compared with the normal reference values for school-age children (<4.0 and >40.0 µg/mL).

3.6.3 Body composition

Body composition will be defined using a two-compartment model including **fat mass** and **fat-free mass** (intracellular and extracellular water, minerals, glycogen and protein). Body composition will be expressed as a ratio of fat mass to body weight (**% fat mass**).

Total body water (TBW) will be calculated as:
[(^2H_2O dose [g])*1000)/ ^2H_2O enrichment)]/1.041] – [cumulative fluid intake (kg)]

Fat free mass (FFM) will be calculated as:
TBW*100/hydration coefficient
Hydration coefficient = 79.3 for male children; 79.0 for female children

Fat mass (FM) will be calculated as: Infant weight (kg) – FFM

% FM will be calculated as: (FM*100)/(Infant weight [kg])

3.6.4 Morbidity

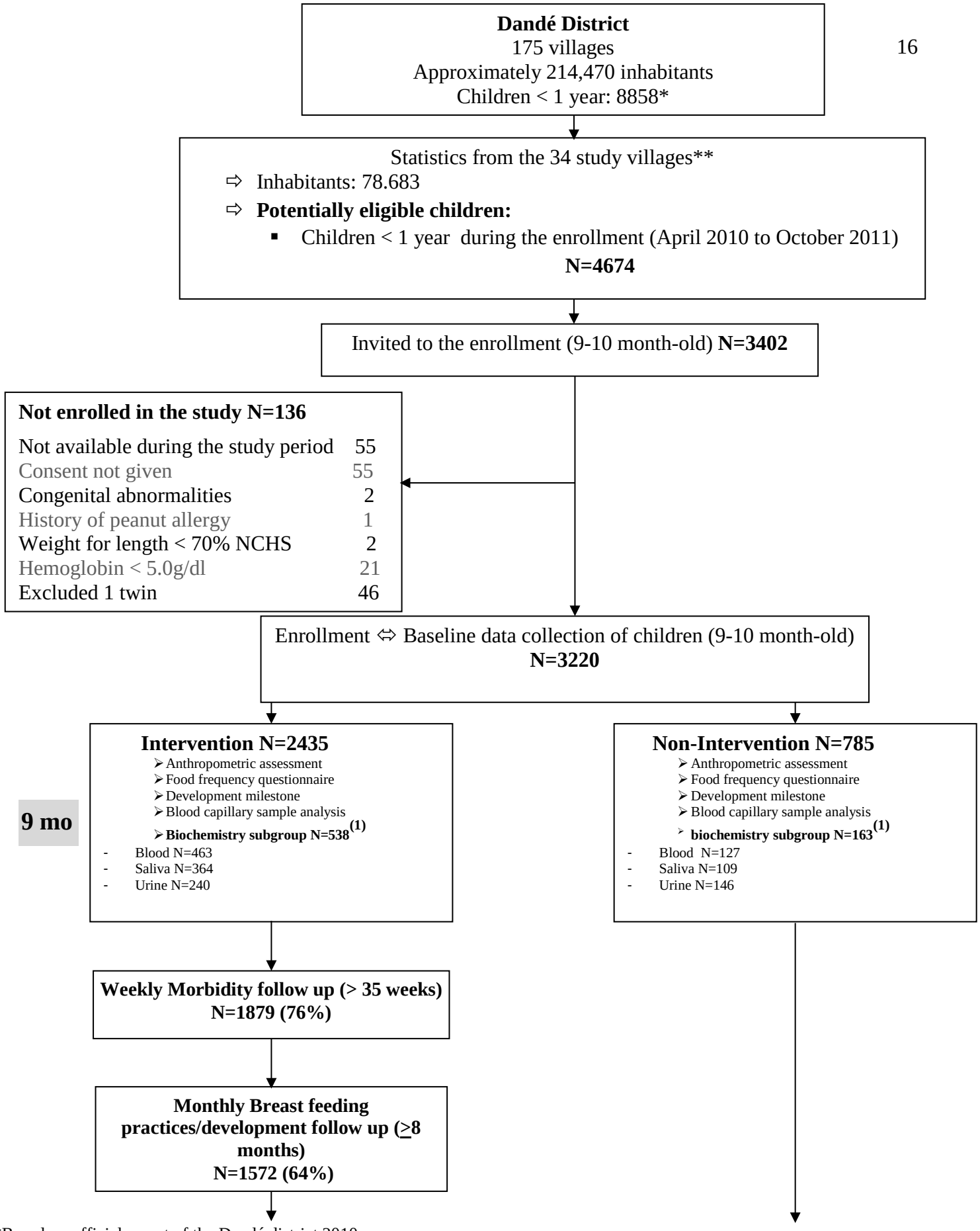
Morbidity terms will be defined as follows:

- **Diarrhea** will be defined in two ways: as the presence of three or more liquid or semi-liquid stools during 24-hour period, or as the presence of four or more liquid or semi-liquid stools during 24-hour period.
- An **episode of diarrhea** will be defined as the period starting the day the child has diarrhea but has not had diarrhea during the previous two days. The episode ends on the last day the child has diarrhea which is then followed by two or more diarrhea-free days.
- An episode of **severe diarrhea** will be defined as any episode of diarrhea that is **1)** associated with dehydration as assessed by the field worker; **2)** accompanied by reported presence of visible blood in at least one stool; **3)** characterized by the presence of six or more liquid or semi-liquid stools during 24-hour period; or **4)** lasts for $\geq$ 14 days.
- **Fever: 1) reported fever** was defined as any reported fever by the caregiver, whether or not confirmed by measured elevated temperature; **2) confirmed fever** was defined as measured temperature (T>37.5°C) by auricular thermometer.
- An **episode of fever** will be defined as the period starting the day when the child has fever but he has not had fever during the previous two days. The episode ends on the last day the child has fever which is then followed by two fever-free days.
- An **episode of malaria** will be defined as the presence of a new episode of reported or confirmed fever and a positive malaria rapid diagnostic test (RDT) obtained 21 days after the previous malaria episode
- An **episode of severe malaria** will be defined as any episode of malaria which is accompanied by any of the following danger signs at some point during the episode: seizure, unconsciousness, respiratory distress.
- **Acute lower respiratory illness** (ALRI) will be defined as any episode when the child exhibits cough or respiratory difficulties (wheezing/stridor or chest in-drawing, reported by caregiver). An episode ends on the last day the child has ALRI which is then followed by three respiratory distress-free days.
- **Acute upper respiratory illness** (AURI) will be defined as any episode when the child exhibits cough and a purulent nasal discharge (reported by caregiver). An episode ends the last the child has AURI which is then followed by seven purulent nasal discharge-free days.

4.0 Statistical analysis

4.1 Study flowchart

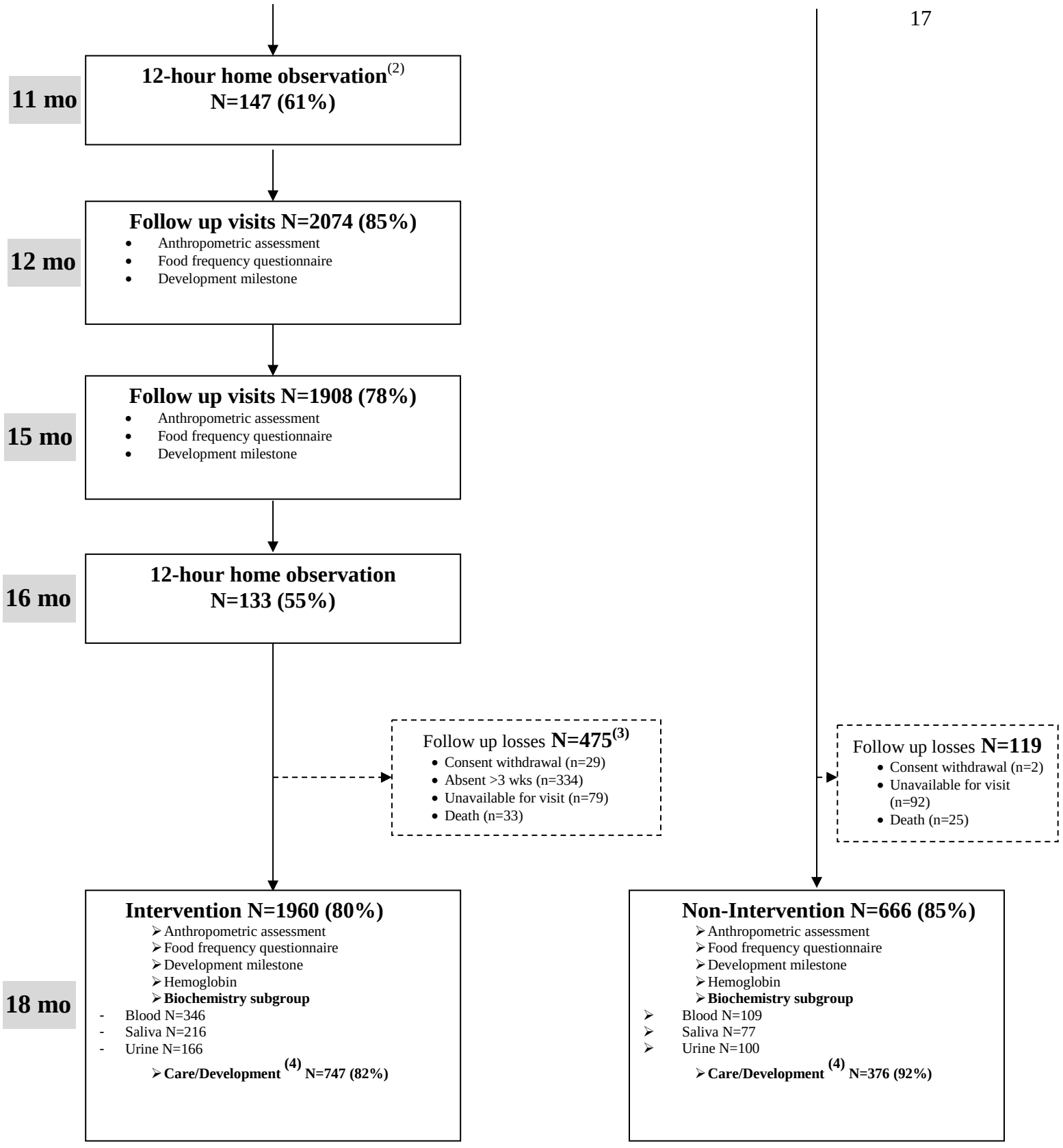
A participant flow diagram was prepared and will be shared with the study investigators. Percentages showed in the diagram represent percent of total enrolled children in each cohort that have completed data for this measure.



*Based on official report of the Dandé district 2010

** Badoville, Bakaribougou, Bambé, Colma, Dafinso, Dandé, Dawéra , Djirwal, Dorona, Faramana, Fo, Karna, Kogoma, Kokoroba, Koréba, Koundougou, Kuini, Lanfiera Coura, Loungo, Mangorotou, Moribougou, Mossibougou, Natéma, Padéma, Samandéni, Sangouléma, Sarfalaye, Séguéré, Siankoro, Soumorodougou, Soungalodaga, St jean, Tarama, Zangoma.

(1) Sample size: 468. Size of subgroup increased until the target sample size for plasma samples was achieved.



[2] Sample size: 240; [3] Number of children who dropped out between enrollment and 18 months visit; [4] Sample size 1322

4.2 Procedures for data cleaning

We performed data cleaning at various points during and after data collection:

a. At the field site, data collection supervisors checked all forms for completeness and consistency, and resolved queries before forms were sent to the study office.
b. At the study office, data quality agents/project coordinators checked some (anthropometric, development) or all the forms (biochemistry and morbidity) before sending them for data entry by two independent data entry clerks.
c. All data were double-entered. Data entry clerks validated the datasets and were allowed to resolve discrepancies themselves after checking the hard copy version. If it was not clear, they referred to the data entry supervisor or coordination team.
d. The data entry supervisor verified and/or compared the entries by the two entry clerks after validation. He generated a reconciled database sent to the data advisor to be merged with previous data bases. This data base is uploaded on the Smartsite.
e. Stata syntaxes were written by project coordinators to review all the datasets for errors in child ID, group assignment, inconsistent variables, biologically implausible values, and missing values. These were resolved by rechecking the original data form (including obtaining clarification from the field worker who completed the form when possible).

4.3 Outliers

- There were range checks on some variables (e.g. anthropometric variables); the same range checks were used during data entry and data cleaning.
- Outliers were further checked by visually inspecting Box plots and/or histograms of individual continuous variables, and scatterplots of related variables.
- Outliers which were clearly impossible or implausible values were corrected if possible, or recoded as missing if correction was not possible.
- Outliers which are plausible or possible will be kept. Variables with outliers will be transformed. If after appropriate transformation, outlying observations are still more than 3SD from the mean, a sensitivity analysis will be done to determine if such outliers have undue influence on the results.

4.4 Software

All analyses will be done using SAS version 9.3 (SAS Inst. Cary, NC, USA) or Stata version 10.1 (StataCorp, TX, USA). The WHO Child Growth Standards (WHO Multicentre Growth Reference Study Group, 2006) will be used for age-and-sex standardization of child weight, length, head circumference, arm circumference and weight-for-height using the SAS (Version 9.3, Cary, NC) macros for the WHO Child Growth Standards (2006).

4.5 Background characteristics of participants and baseline comparisons

For all variables measured, the available value at the time of enrollment, prior to the first intake of the study supplements will be considered as background or baseline value. The following background characteristics will be presented, by treatment group:

- Demographic and socio-economic characteristics (household) including (see separate statistical analysis plan for SES):
  - Age of caregiver

  - Maternal education at enrollment[1]
  - Maternal marital status/rank[2]
  - One or several proxy indices for socio-economic status
  - Number of children <5 years old who are still alive
  - Proxy for food insecurity (Household Food Insecurity Access Score, HFIAS, adjusted for season)[3]
- Baseline anthropometric characteristics (mothers):
  - Weight
  - Height
  - Body Mass Index (BMI)[4]
- Baseline anthropometric characteristics (children):
  - Weight
  - Length
  - Mid upper arm circumference (MUAC)
  - Head circumference (HC)
  - Length for age z-score (LAZ)
  - Weight for age z-score (WAZ)
  - Weight for length z-score (WLZ)
  - Mid upper arm circumference for age z-score (MUAC-AZ)
  - Head circumference for age z-score (HC-AZ)
- Hematologic and biochemical indices :
  - Hemoglobin (Hb)
  - Rapid diagnostic test for malaria (RDT)
  - Malaria microscopy
  - Body composition (fat and free fat body mass)
  - Acute phase response proteins (plasma C-reactive protein (CRP) and alpha-1 glycoprotein (AGP))
  - Iron status (zinc protoporphyrin (ZPP), plasma ferritin (pF), soluble transferrin receptor (sTfR))
  - Plasma zinc concentration (PZC)
  - Vitamin A status (retinol-binding protein (RBP))
  - Iodine status (urinary iodine (UI), and plasma thyroglobulin (Tg) concentrations) and thyroid function (dried blood spot thyroid stimulating hormone (TSH) and thyroxine (T_4))

[1] Three categories: 0- no education; 1- no formal education (Koranic school, alphabetization or professional) or formal education for less than one year or in a system without degree; 2- at least one year of formal education

[2] Four categories: 1- single, divorced, widow; 2- sole wife; 3- first wife in a polygamous household; 4- second wife or higher in a polygamous household.

[3] The Household Food Insecurity Access (HFIA) Score is a continuous measure of the degree of food insecurity in the household and is based on a set of questions that encompass three domains of food insecurity: (1) anxiety and uncertainty about the household food supply; (2) insufficient quality; and (3) insufficient food intake and its physical consequences (Coates et al. 2007). The higher the score, the higher the degree of household food insecurity experienced in the previous four weeks.

[4] Three categories: 1- lower than 18.5; 2- higher or equal to 18.5 and lower than 25; 3- higher or equal to 25

Analysis of baseline characteristics will be completed as follows:

- We will use frequencies and percentages to summarize categorical data. Percentages will be calculated based on the number of participants for whom data are available.
- Continuous variables will be summarized using either mean and SD for variables, or median and quartiles.
- Where data for any participant are missing, the number of participants included in the analysis will be indicated.

Baseline characteristics will be analyzed by cohort and by group. This will allow checking the success of the randomization, especially at the community level. Additionally, study completers and non-completers will be analyzed as well to determine their potential effect on the study outcomes and subject adherence. Non-completers were categorized into 1- Consent withdrawal; 2- Reported absence >3 wks from morbidity questionnaire, or time of end of follow up from the potential 18-mo visit >0.5 month, and number of weeks in the study <35 wks; 3- Absent the day of 18-mo visit, or time of end of follow up from the potential 18-mo visit <=0.5 month, or number of weeks in the study >=35 wks; or 4- Death.

4.6 Potential effect modifiers

The following variables will be considered as effect modifiers:

a. Baseline value for the outcome being analyzed (continuous variable, categorical considering the median, or at certain predefined baseline values using "at" option)
b. Baseline LAZ < -1.5 for no-anthropometric variables (categorical variable)
c. Median baseline LAZ of study population (categorical)
d. Sex of child (categorical)
e. Maternal height at enrollment (continuous variable, and categorical)
f. Maternal BMI at enrollment as categorical variable (three categories with predefined cutoffs) if test of correlation with the composite variable for food security shows a no-collinearity.
g. Proxy for food insecurity at enrollment (HFIAS, adjusted for season)
h. Proxy for hygiene/water quality at enrollment (morbidity outcomes only)[1]
i. Season during the study indicated by the season of the majority of months spent during the study (anthropometric and biochemical outcomes) or season at follow-up visit (infant and young child feeding practices)[2]
j. Month/year of enrollment (morbidity outcomes only)
k. Maternal education at enrollment
l. Maternal marital status/rank at enrollment
m. Iron supplementation (as provided by the study along with deworming at enrollment)[3]

1We will construct a restricted housing quality index for hygiene and water quality for each household based on the following baseline housing characteristics: drinking water supply, sanitation facilities, and flooring material. These housing quality characteristics are combined into an hygiene/water quality index using (with a mean of zero and standard deviation of one) principal components analysis (Vyas and Kumaranayake 2006).

[2] Season defined in two categories (rainy from May to September and dry from October to April, FAO) by year for all the outcomes except for dietary practices, where season is defined in three categories (rainy from May to September, dry/cool from October to January, and dry/hot from February to April)

[3] Three categories: 0- child did not receive any supplementation; 1- child Hb between 60 and 80 g/L; 2- child Hb between 50 and 60 g/L

n. Child PZC at baseline (biochemistry variables, adjusted for inflammation)

The specific effect modifiers that will be considered in each analysis have been indicated in **Section 4.10** below.

4.7 Covariate adjustment

For each outcome, we will adjust for the following variables when the correlation with outcome is significant ($p<0.1$):

1. Baseline child LAZ score
2. Baseline child WAZ score
3. Baseline child WLZ score
4. Baseline LAZ < -1.5
5. Baseline child Hb concentration
6. Baseline child capillary ZPP (adjusted for malaria)
7. Baseline PZC (biochemistry variables, adjusted for inflammation)
8. AGP and CRP (biochemistry variables)
9. Time of day of blood sampling (biochemistry variables)
10. Time since last breast-feeding (biochemistry variables)
11. Baseline percent fat mass (biochemistry variables)
12. Baseline body weight
13. Child sex
14. Child age (at enrollment and at the follow up visit)
15. Maternal height at enrollment
16. Maternal BMI at enrollment
17. Maternal education at enrollment
18. Maternal marital status/rank at enrollment
19. Number of children < 5 years old
20. Feeding practice variables at enrollment[1]
21. Proxy for food insecurity (HFIAS adjusted for season)
22. Proxy for hygiene/water quality (morbidity outcomes only)

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1 Four indicators adapted from "Indicators for assessing infant and young child feeding practices: conclusions of a consensus meeting held 6–8 November 2007 in Washington D.C., USA" : continued breastfeeding, minimum dietary diversity, minimum meal frequency and animal source foods.

5-1- Continued breastfeeding, three categories: 1) Child is not breastfed, 2) Child is breastfed for less than 6 times during the previous 24hours (or mother does not know how many times she breastfed the child), 3) Child is breastfed at least 6 times during the previous 24 hours.

5-2- Minimum dietary diversity, two categories: 1) Child received food from less than 4 food groups, 2) Child received food from 4 or more food groups.

5-3- Minimum meal frequency, two categories: 1) Breastfed child received less than 3 meals and snacks, or no-breastfed child received less than 4 meals and snacks during the previous 24 h; 2) Breastfed child received at least 3 meals and snacks during the previous 24 h, or no- breastfed child received at least 4 meals and snacks during the previous 24 h

5-4- Animal source food: Score between 0 and 3. ASF includes three food groups: flesh foods (meat, fish..), eggs and dairy products.

23. Proxy for livestock (small livestock and large livestock)[1]
24. Month/year of enrollment
25. Season during the study indicated by the season of the majority of months spent during the study (anthropometric and biochemical outcomes) or season at follow-up visit (young child feeding practices)
26. Month/year of enrollment (morbidity outcomes only)
27. Iron supplementation at enrollment
28. Vitamin A supplementation (frequency during the study period, or supplementation during the month preceding the last 18 mo visit, IC groups)
29. Mosquito net utilization (monthly frequency during the study period, IC groups)
30. Diarrhea prevalence (IC groups)[2]
31. Malaria incidence (IC groups)[3]
32. Fever prevalence (IC groups)[4]
33. Anthropometrist and assistant (anthropometric outcomes)

4.8 Timing of measurement of outcome variables

The outcome variables were measured after the eligible enrolled children had started taking the study supplements. The table below shows the timing of measurement of the outcome variables.

[1] We construct a composite measure of household livestock ownership by converting animals owned by a household to livestock units. Livestock units are a standardized animal unit calculated by generating a weighted sum of the number of animals owned, where the weights are determined by "feeding requirement" (FAO 2003). We used the following weights, identified by the FAO as relevant for Sub-Saharan Africa: cattle (0.5), sheep (0.1), goats (0.1), pigs (0.2), horses (0.8), chickens (0.01), ducks (0.03), turkeys (0.03), rabbits (0.02). Using the same concept, we constructed two separate indexes, index of household small and large animal ownership.

[2] Considered only in the analysis among IC groups at 12, 15 and 18 months

[3] Considered only in the analysis among IC groups at 12, 15 and 18 months

[4] Considered only in the analysis among IC groups at 12, 15 and 18 months

Table 4: Outcome variables form and timing for their collection

Variable	Measurement
Morbidity and supplement intake	Measured weekly during home visits from enrollment at 9 months (+1-3 days) to completed 18 months of age in the intervention cohort.
Breast feeding practices/development follow up	Monthly follow up from enrollment at 9 months to completed 18 months of age in the intervention cohort.
Food frequency	Data collected on weekly and 24 hour-recall at 9 and 18 months for the intervention and the non-intervention cohorts, and at 12 and 15 months for the intervention cohort
Anthropometric indices	Measured at 9 months and at 18 completed months of age for all the groups, and at 12 and 15 months for the intervention cohort.
Biochemical indices	Measured in a subgroup after enrollment (+1-3 days), and at 18 completed months of age before the final anthropometric visit.
12-hour home observation	In a 10%-subsample of the intervention cohort, where all supplement related behaviors, including who received the products and ways in which the LNS and tablet were given are reported at 11 and at 16 months (+ 1 to 2 months).
Family care indicators and neuro-development	In a subgroup at 18 months.

4.9 Hypothesis testing

The analysis will begin with testing the null hypothesis of no difference between the two study cohorts (all IC groups (LNS-Zn0; LNS-Zn5; LNS-Zn10; LNS-ZnTab5) *versus* the standard treatment cohort (NIC)) using Proc mixed, and controlling for pre-specified covariates including baseline values, household characteristics, season/year and community (as random effect as well). Only covariates significantly associated with an outcome at 10% level of significance in a bivariate analysis will be included in the final adjusted analysis. This means we may have different sets of covariates for each outcome.

The next test will be testing the null hypothesis of no difference among the five groups (NIC and IC groups) using Proc mixed, and controlling for the same covariates.

The last test will be testing the null hypothesis of no difference among the four IC groups using Proc mixed and controlling for the same covariates.

For the primary outcomes between 9, 12, 15 and 18 mo, the analysis will compare the difference (95% CI) between means in the intervention groups LNS-Zn5, LNS-Zn10, LNS-ZnTab5 *versus* LNS-Zn0.

For all analyses, if the global null hypothesis is rejected at 0.05 level, then we will perform post-hoc pairwise comparisons of the two cohorts, the entire five groups or the four IC groups using Tukey-Kramer adjustment in SAS.

The effects of potential effect modifiers will be assessed with an interaction term in the ANCOVA or logistic regression model. Significant interactions ($P < 0.10$) will be further examined with stratified analyses, estimation of separate regression lines, or estimation of adjusted means at key points of the covariate, in order to understand the nature of the effect modification.

Proc Glimmix will be used for all the categorical binary outcomes, using the above described approach as stated for Proc Mixed

4.10 Analysis of primary outcomes at a single time point

Below we list the primary outcomes to be analyzed (at 18 months for the comparison between the two study cohorts, and at 12, 15 and 18 months for the IC groups), and indicate the covariates and effect modifiers that will be used for each analysis. Each of the effect modifiers will be considered separately in the regression model to avoid collinearity.

Table 5: Analysis of primary outcome variables

Primary outcome variable	**Analysis**	**Covariates**	**Effect modifiers**
Child length	-Between 2 cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Procmixed) -Among IC groups at 12, 15 and 18 months: Repeated measurement (SAS Proc mixed)	- Child length at baseline - Baseline capillary ZPP adjusted for malaria - Child sex - Child age - Season during the study - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment - Anthropometrist and assistant - RDT at enrollment	- Child length at baseline - Child sex - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season at enrollment - Iron supplementation at enrollment
Length-for-age z-score at 18 months	-Between cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups	- Child LAZ at baseline - Baseline capillary ZPP adjusted for malaria - Child sex - Child age - Season during the study - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at	- Child LAZ at baseline - Child sex - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment

Primary outcome variable	**Analysis**	**Covariates**	**Effect modifiers**
	at 12, 15 and 18 months: Repeated measurement (SAS Proc mixed)	enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment - RDT at enrollment - Anthropometrist and assistant	
Child weight	-Between cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 12, 15 and 18 months: Repeated measurement (SAS Proc mixed)	- Child weight at baseline - Baseline capillary ZPP adjusted for malaria - Child sex - Child age - Season during the study - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment - RDT at enrollment - Anthropometrist and assistant	- Child weight at baseline - Child sex - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment
weight-for-age (WAZ)	-Between cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 12, 15 and 18 months: Repeated measurement (SAS Proc mixed)	- Child WAZ at baseline - Baseline capillary ZPP adjusted for malaria - Child sex - Child age - Season during the study - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment - RDT at enrollment - Anthropometrist and assistant	- Child WAZ at baseline - Child sex - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment
weight-for-length (WLZ) Z-scores	-Between cohorts at 18 months: ANCOVA	- Child WLZ at baseline - Baseline capillary ZPP adjusted for malaria	- Child WLZ at baseline - Child sex - Maternal height at enrollment

Primary outcome variable	**Analysis**	**Covariates**	**Effect modifiers**
	(SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 12, 15 and 18 months: Repeated measurement (SAS Proc mixed)	- Child sex - Child age - Season during the study - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment Maternal marital status/rank at enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment - RDT at enrollment - Anthropometrist and assistant	- Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment
Incidence of diarrhea	Among IC groups: - Logistic regression	- Child LAZ, WAZ and WLZ at baseline - Baseline LAZ < -1.5 - Child sex - Child age - Month/year of enrollment - Total number of days at risk of diarrhea - Maternal education at enrollment - Maternal marital status/rank at enrollment - Number of children < 5 years in the household - Proxy for hygiene/water quality - Feeding practice variables at enrollment - HFIAS adjusted for season - Small livestock number (given as Tropical Livestock Unit) - Iron supplementation at enrollment	- Child LAZ, WAZ and WLZ at baseline - Baseline LAZ < -1.5 - Child sex - Month/year of enrollment - Total number of days at risk of diarrhea - Maternal education at enrollment - Maternal marital status/rank at enrollment - Proxy for hygiene/water quality - HFIAS adjusted for season - Iron supplementation at enrollment
Incidence of positive RDT-confirmed malaria infection	Among IC groups: - Poisson regression	- Child LAZ, WAZ and WLZ at baseline - Baseline LAZ < -1.5 - Child sex - Child age - Month/year of enrollment - Total number of days at risk of malaria - Maternal education at enrollment - Maternal marital status/rank at enrollment	- Child LAZ, WAZ and WLZ at baseline - Baseline LAZ < -1.5 - Child sex - Month/year of enrollment - Total number of days at risk of malaria - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season

Primary outcome variable	**Analysis**	**Covariates**	**Effect modifiers**
		- HFIAS adjusted for season - Iron supplementation at enrollment	- Iron supplementation at enrollment
Final adjusted PZC	-Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 18 months: ANCOVA (SAS Proc mixed)	- Child length at baseline - Baseline LAZ < -1.5 - Child sex - Child age - Child zinc concentration at baseline - AGP and CRP - Time of day - Time since last breast-feeding - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season - Season during the study	- Child length at baseline - Baseline LAZ < -1.5 - Child sex - Child zinc concentration at baseline - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season - Season during the study
Prevalence of PZC<65 μg/dL	-Between cohorts at 18 months: logistic regression (SAS Proc Glimmix) -Among the five groups (NIC and IC) at 18 months: logistic regression (SAS Proc Glimmix)	- Child zinc concentration at baseline - AGP and CRP - Time of day - Time since last breast-feeding	- Child zinc concentration at baseline
Change in fat free mass	-Between cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 18 months: ANCOVA (SAS Proc mixed)	- Baseline fat free mass - Baseline body weight - Baseline capillary/venous ZPP - Baseline PZC - Child sex - Child age - Season during the study - Maternal education at enrollment - Maternal marital status/rank at enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Malaria incidence - Diarrhea prevalence - Fever prevalence	- Baseline LAZ < -1.5 - Median baseline LAZ of study population - Child sex - Season during the study - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season

Primary outcome variable	**Analysis**	**Covariates**	**Effect modifiers**
Change in percent fat mass	-Between cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 18 months: ANCOVA (SAS Proc mixed)	- Baseline percent fat mass - Baseline capillary/venous ZPP - Baseline body weight - Baseline PZC - Child sex - Child age - Season during the study - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season - Malaria incidence - Diarrhea prevalence - Fever prevalence	- Baseline LAZ < -1.5 - Median baseline LAZ of study population - Child sex - Season during the study - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season

4.11 Analysis of secondary outcomes at a single time point

Table 6 summarizes the secondary outcomes to be analyzed, and indicates the covariates and effect modifiers that will be used for each analysis.

Table 6: Analysis of secondary outcome variables

Secondary outcome variable	Analysis	Covariates	Effect modifiers
Head circumference, and HC for age Z-score	-Between cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 12, 15 and 18 months: Repeated measurement (SAS Proc mixed)	- Child HC at baseline - Baseline capillary ZPP adjusted for malaria - Child sex - Child age - Season during the study - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment - Anthropometrist and assistant - RDT at enrollment	- Child HC at baseline - Child sex - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season at enrollment - Iron supplementation at enrollment
Mid-upper arm circumference, and MUAC for age Z-score	-Between cohorts at 18 months: ANCOVA (SAS Proc mixed) -Among the five groups (NIC and IC) at 18 months: ANCOVA (SAS Proc mixed) -Among IC groups at 12, 15 and 18 months: Repeated measurement (SAS Proc mixed)	- Child MUAC at baseline - Baseline capillary ZPP adjusted for malaria - Child sex - Child age - Season during the study - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - Feeding practice variables at enrollment - HFIAS adjusted for season - Iron supplementation at enrollment - Anthropometrist and assistant - RDT at enrollment	- Child MUAC at baseline - Child sex - Maternal height at enrollment - Maternal BMI at enrollment - Maternal education at enrollment - Maternal marital status/rank at enrollment - HFIAS adjusted for season at enrollment - Iron supplementation at enrollment
Prevalence of total and severe stunting, underweight and	-Between cohorts at 18 months: logistic regression (SAS Proc Glimmix)	- Baseline LAZ, WAZ or WLZ - Child sex	

Secondary outcome variable	**Analysis**	**Covariates**	**Effect modifiers**
wasting at 18 mo	-Among the five groups (NIC and IC) at 18 months: logistic regression (SAS Proc Glimmix)		
Hemoglobin status at 18 mo	Continuous variable analyzed using Proc mixed, prevalence of anemia was analyzed using Proc Glimmix	- Baseline Hb concentration - Child sex - AGP and CRP - Season during the study	- AGP and CRP Season during the study
Acid glycoprotein and c-reactive protein at 18 mo	Continuous variables analyzed using Proc mixed, prevalence of high AGP, high CRP and high AGP and CRP were analyzed using Proc Glimmix	- Baseline value	
Adjusted plasma ferritin at 18 mo	Continuous variable analyzed using Proc mixed, prevalence of low plasma ferritin was analyzed using Proc Glimmix	- Baseline value - Child sex - AGP and CRP - Maternal BMI at enrollment - Indicators of feeding practices at enrollment - Iron supplementation at enrollment - RDT at enrollment - Recent iron supplementation during the previous month (IC groups only) - Recent vitamin A supplementation during the previous month (IC groups only) - Malaria incidence during the previous month (IC groups only)	-Baseline value - AGP and CRP - Recent iron supplementation during the previous month (IC groups only) - Recent vitamin A supplementation during the previous month (IC groups only) Malaria incidence during the previous month (IC groups only)
Adjusted plasma soluble transferrin receptors at 18 mo	Continuous variable analyzed using Proc mixed, prevalence of high plasma soluble transferrin receptors was analyzed using Proc Glimmix	- Baseline value - Child sex - Child age - AGP and CRP - Indicators of feeding practices at enrollment - Iron supplementation at enrollment - Season during the study	- Baseline value - Season during the study
Adjusted retinol binding protein at	Continuous variable analyzed using Proc	- Baseline value - Child sex	- Baseline value - Recent vitamin A

Secondary outcome variable	Analysis	Covariates	Effect modifiers
18 mo	mixed, prevalence of low RBP was analyzed using Proc Glimmix	- Child age - AGP and CRP - Indicators of feeding practices at enrollment - Season during the study - Maternal education - Recent vitamin A supplementation during the previous month (IC groups only) - Vitamin A rich food at 18 mo	supplementation during the previous month (IC groups only) - Vitamin A rich food at 18 mo
Adjusted venous zinc protoporphyrin at 18 mo	Continuous variable analyzed using Proc mixed, prevalence of high venous ZPP was analyzed using Proc Glimmix	- Baseline value - Child sex - AGP and CRP - Indicators of feeding practices at enrollment - Iron supplementation at enrollment - Season during the study - Maternal BMI at enrollment	- Baseline value - Indicators of feeding practices at enrollment
Iodine status (urinary iodine) at 18 mo	Continuous variable analyzed using Proc mixed, prevalence of low urinary iodine was analyzed using Proc Glimmix. Also Final urinary iodine concentrations were adjusted for baseline values using regression model (GLM). The median, the quartiles, and the highest 75^{th} percentile of the final adjusted urinary iodine concentration by cohort and by group was compared using Proc surveyfreq	- Baseline value - Child age - Child sex	- Baseline value - Child sex
Thyroid stimulating hormone (dried blood spot TSH concentration) at 18 mo	Continuous variable analyzed using Proc mixed, prevalence of elevated thyroid-stimulating hormone concentration was analyzed using Proc Glimmix	- Baseline value - Child age - Child sex	- Baseline value - Child sex
Thyroxin (dried	Continuous variable	- Baseline value	- Baseline value

Secondary outcome variable	Analysis	Covariates	Effect modifiers
blood spot T4 concentration) at 18 mo	analyzed using Proc mixed, prevalence of low thyroxin concentration was analyzed using Proc Glimmix	- Child age - Child sex	- Child sex
Plasma thyroglobulin concentrations	Continuous variable analyzed using Proc mixed, prevalence of high or low plasma thyroglobulin concentration[1] was analyzed using Proc Glimmix. Final Tg concentrations were adjusted for baseline values using regression model (GLM). The quartiles of the final adjusted Tg concentration by cohort and by group were compared using Proc surveyfreq	- Baseline value - Child age - Child sex	- Baseline value - Child sex

[1]Because no reference values exist for young children, the prevalence of low and high Tg concentrations were assessed by using the reference values for school-age children, by investigating -/+2 and -/+1 standard deviation (z Z-scores were calculated based on the distribution of the whole sample, by standardizing the distribution to a mean of 0 and standard deviation of 1), and by comparing the quartiles of the normally-transformed and adjusted values for baseline Tg concentrations.

4.12 Analysis of repeated measurements

For continuous variables, a linear mixed model (PROC MIXED) will be used to compare patterns in outcome variables across time among the IC groups. For categorical variables, a variant (mixed model logistic regression (SAS PROC GLIMMIX)) will be used. In these analyses, the covariates and effect modifiers indicated above will be considered.

4.13 Data transformation

Continuous outcomes will be assessed for conformance to the normal distribution and will be transformed appropriately (e.g. adjusted plasma ferritin, adjusted soluble transferrin receptor). If no suitable transformation can be found to optimize normality and homogeneity of variances, analysis will be done on ranked data. Also see section 4.3 (Outliers).

4.14 Data presentation

Continuous normally distributed variables will be presented as means +/- SD as obtained from proc means. Not normally distributed variables will be transformed as described above, and presented as geometric mean (95% confidence interval) or estimated median (95% confidence interval). Categorical variables will be presented as number (%) from proc surveyfreq which accounts for clustering and the random effect of village.

4.15 Confidence intervals

Estimates of treatment effects will be accompanied by a 95 % confidence interval, wherever possible.

5- References

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6 Appendix A: Data collection

What	When	Where	Who	Questionnaire/ Form
Screening	Children 9-9.9 months of age	At participant homes	Agent de terrain, field supervisors	• Form N01 Screening
Enrollment	Children 9-9.9 months of age	Central location in village (health clinic, school, etc)	Enrollment team (based in Bama)	• Form N02 Enrollment/Eligibility • Form N03 Baseline Anthro • Form N04 Hb RDT ZPP Vaccine • Form N05 FFQ • Form N06 Obs Dev Milestones • Form N19 Biochem subgroup
Biochemistry visits (subgroup)	1 day after enrollment 9 months after enrollment (final visit)	Health clinic	Biochem team Lab team	• Form N08 Blood collection • Form N09 Body composition • Form 17a/b Malaria microscopy • Form 18a Capillary ZPP • Form 18b Venous ZPP
SES visits	Within ~1 week of enrollment 6 months after enrollment 12 months after enrollment	At participant homes in village	SES team	• Form S01 Baseline SES • Form S02 Food security • Form S03 Risk game • Form S04 Household expenditures (subgroup) • Form S05 Household income (subgroup) • Form S07 Patience game (subgroup) • KAP (15 months only)
Morbidity visits	Weekly after enrollment (intervention villages)	At participant homes in village	Agent de terrain Physicians	• Form N07 Morbidity • Form N10 Home morbidity chart • Form N11 Maternal devlop milestones, breastfeeding (every 4 weeks) • Form N16a Serious Adverse Events AT • Form N16b Serious Adverse Event Assessment • Form N22 Verbal Autopsy
12-hr in-home observations (subgroup)	2 and 7 months after enrollment	At participant homes in village	12-hour in-home observation team	• Form N14 (on PDA)
Follow-up anthropometry visits	3 and 6 months after enrollment (intervention villages)	Central location in village (health clinic, school, etc)	Anthropometry team	• Form N12 Anthro/vaccines • Form N13 Obs dev milestones/FFQ
Final visit	9 months after enrollment	Central location in village (health clinic, school, etc)	Final visit team	• Form N21 Anthro • Form N15 Hb • Form N05 FFQ • Form N06 Obs Dev Milestones • Form N23 FCI • Form N24 Comprehensive Development Interview

1) **Form N02: Enrollment and eligibility at 9 months**

- Confirmation of parental consent
- Child birthdate
- Child sex
- Assessment of inclusion and exclusion criteria:
    - Permanent resident of Dandé Health District
    - Available during the study period
    - The child's family accept home visitors
    - Absence of bipedal edema
    - No severe illness warranting hospital referral
    - Absence of congenital abnormalities or other diseases that could interfere with growth
    - Absence of chronic medical condition requiring frequent medical attention
    - Not exposed to and/or infected with HIV
    - No history of allergy towards peanuts
    - Does the child have a history of anaphylaxis or serious allergic reaction to any substance, requiring emergency medical care?
    - Is the child currently participating in another clinical trial?
    - Child hemoglobin (g/dl) **≥5.0**
    - Child weight-for-length percentage **≥ 70%**
- In case of eligibility, assign study ID number
- Assign study group color

2) **Form N03: Anthropometry of child and mother at enrollment day**

- Weight of child (±0.01kg) in duplicate. Repeated if disagreement > 0.1 kg.
- Length of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- Mid upper arm circumference of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- Head circumference of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- Maternal clothing when weighed (heavy, light clothing and sweater or light)
- Whether mother is pregnant
- Maternal weight (±0.01kg) in duplicate. Repeated if disagreement > 0.1 kg.
- Maternal height (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.

3) **Form N04: Capillary blood collection at enrollment day**

- Hemoglobin
- Rapid Diagnostic Test for malaria.
- Confection of malaria blood smear slide.
- Collection of capillary blood sample for ZPP.
- Vaccination record (BCG, Polio 1, 2 and 3, DTCoq 1, 2 and 3, Anti-measles and Antiamaril).

4) **Form N05: Food frequency questionnaire and 9 and 18 months**

- Confirmation of respondent's knowledge of infant's consumption during previous 24 hours

- Number of breastfeeding episodes on previous 24 hours
- Age of child at weaning
- Reasons for weaning
- Consumption of liquids on previous day (milk, fresh or tinned; powdered milk; infant formula)
- Food groups eaten **on previous 24 hours**, including:
  - Cereals
  - Vitamin A-rich vegetables
  - Roots and tubers
  - Green leafy vegetables
  - Vitamin A-rich fruits
  - Other type of fruits
  - Other type of vegetables
  - Nutrient-rich organ meat
  - Other type of meat
  - Eggs
  - Fish (fresh, grilled, fried, etc).
  - Legumes, pulses and nuts.
  - Yoghurt and other milk derivatives
  - Oil and fats or foods made with these.
  - Sugary foods such as chocolates, sweets.
  - Condiments for flavor, such as hot pepper, garlic, dried fish, soumbala etc.
  - Grubs, snails or insects.
- Number of meals and snacks during previous 24 hours
- Food groups eaten **during previous seven days (by day)**, including:
  - Cereals
  - Vitamin A-rich vegetables
  - Roots and tubers
  - Green leafy vegetables
  - Vitamin A-rich fruits
  - Other type of fruits
  - Other type of vegetables
  - Nutrient-rich organ meat
  - Other type of meat
  - Eggs
  - Fish (fresh, grilled, fried, etc).
  - Legumes, pulses and nuts.
  - Milk, including fresh, powdered or tinned milk, yoghurt
  - Infant formula (Nursie or France Lait)
  - Infant cereal (Cerelac)
  - Oil and fats or foods made with these.
  - Sugary foods such as chocolates, sweets.
  - Condiments for flavor, such as hot pepper, garlic etc.
  - Grubs, snails or insects.
- Consumption and type of vitamins or mineral supplements during previous 7 days.

5) Form N06: Developmental outcomes at 9 and 18 months:

- State of alertness of child at the time of developmental assessment.
- Child's emotional state.
- Whether child is ill.
- Observation of child running (if yes, observation ended).
- Observation of child walking alone (if yes, observation ended).
- Observation of child standing alone (if yes, observation ended).
- Observation of child walking with assistance.
- Observation of child crawling.
- Observation of child standing with assistance.
- Observation of child sitting without support.

6) Form N07: Weekly home visit to collect information on morbidity and supplementation during previous seven days (from 9 to 18 months of age):

- Number of remaining tablets
- Number of remaining LNS sachets/estimated LNS leftover in pot
- Reported consumption of tablet per day
- Reported consumption of LNS (morning and afternoon)
- Reported morbidity symptoms on each day during previous seven days since last visit, including:
  - Rashes (none, some, severe or don't know)
  - Reduced activity (none, some, severe or don't know)
  - Poor appetite (none, some, severe or don't know)
  - Stool consistency (normal, slightly liquid, very liquid, don't know)
  - Stool pathology (none, blood, mucus, blood and mucus, or don't know)
  - Vomiting (none, some, a lot, or don't know)
  - Fever (none, some, high or don't know)
  - Nasal discharge (none, mild, thick, yellow or greenish or don't know)
  - Cough (none, some, severe or don't know)
  - Difficult breathing (none, nasal obstruction, wheeze, severe difficulty, or don't know)

- Assessment on day of visit, including:
  - Observed health status
  - Presence of dangerous signs
  - Measured temperature
  - RDT results
  - Confection of malaria slide
  - Respiratory distress
  - Respiratory rate

- Medication provided by field worker
- Referral to health center
- History of visits to any health centers and reasons for any such visit

7) **Form N08: Venous blood sample collection at 9 and 18 months (biochemistry subgroup)**
   - Confirmation of absence of fever and diarrhea during previous 48 hours
   - Time of last breastfeeding episode
   - Time of last meal/snack
   - Time of blood draw
   - Estimated volume of blood sample obtained
   - Confection of malaria slide
   - Time of plasma separation
   - Hemolysis of blood sample
   - Plasma sample collected for zinc (2x ≥ 500 μL)
   - Plasma sample collected for APP (2x 25-150 μL)
   - Plasma sample collected for retinol (2x ≥ 250 μL)
   - Plasma sample collected for reserve (≥ 500 μL)

8) **Form N09: Body composition at 9 and 18 months-Biochemistry subgroup**
   - Saliva A collection time (>2.5 mL) and volume (continuation only after successful collection of saliva A)
   - Deuterium oxide dose completely consumed
   - Child's weight (±0.001kg) in duplicate. Repeated if disagreement > 0.1 kg.
   - Time and volume of saliva B collection (at least 2hr 30min after deuterium administration)
   - Time and volume of saliva C collection (at least 30min after saliva B collection)
   - Collection of spot urine samples
   - Breastfeeding, and consumption of other foods/liquids

9) **Form N10: Home chart for caregiver's recording of morbidity symptoms, and LNS and tablets' consumption (intervention groups)**
   - Used as an aid to the memory during weekly morbidity home visit

10) **Form N11: Monthly collection of achievement of developmental milestones and breastfeeding practices during home visit (from 9 to 18 months of age in intervention groups):**

- Achievement of developmental outcomes:
  - Mother's report whether child is able to pronounce single words
  - Mother's report whether child is able to wave bye-bye
  - Mother's report whether child is able to drink from a cup by himself/herself.
  - Mother's report whether child is able to eat by himself/herself.
  - Mother's report whether child is able to run (if yes, interview stopped)
  - Mother's report whether child is able to walk alone (if yes, interview stopped)
  - Mother's report whether child is able to stand alone (if yes, interview stopped)
  - Mother's report whether child is able to walk with assistance
  - Mother's report whether child is able to crawl on hand and knees
  - Mother's report whether child is able to stand with assistance
  - Mother's report whether child is able to sit without support
- Number of breastfeeding episodes on previous 24 hours

- Breastfeeding at least once during previous 4 weeks
- Intention to continue breastfeeding during next 4 weeks
- Reasons for weaning
- Reported consumption of high-dose vitamin A capsule during previous month
- Location of receipt of vitamin A capsule
- Sleeping under mosquito net during previous night

11) Form N12: Anthropometric follow-up visit in the intervention group, at 12 and 15 months:

- Weight of child (±0.01kg) in duplicate. Repeated if disagreement > 0.1 kg.
- Length of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- Mid upper arm circumference of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- Head circumference of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- At 12 months only, type of vaccines received.

12) Form N13: Observed developmental outcomes and shortened food frequency questionnaire at 12 and 15 months (IC groups):

- State of alertness of child at the time of developmental assessment.
- Child's emotional state.
- Whether child is ill.
- Observation of child running (if yes, observation ended).
- Observation of child walking alone (if yes, observation ended).
- Observation of child standing alone (if yes, observation ended).
- Observation of child walking with assistance.
- Observation of child crawling.
- Observation of child standing with assistance.
- Observation of child sitting without support.
- Food groups consumed **yesterday and during last seven days (number of days)**, including:
    - Nutrient-rich organ meat
    - Other types of meat
    - Cereals porridge
    - Eggs
    - Fish (fresh, grilled, fried, etc)
    - Milk, including fresh, powdered or tinned milk, yoghurt
    - Legumes and pulses.
    - Grubs, snails or insects
    - Hot sauces, chili pepper
    - Nuts and oil rich food (peanut paste…)
    - Infant formula
    - Infant cereal (Cerelac)

13) Form N14: 12hour in home observation at 11 and 16 months (subgroup of the IC groups)

- Observation of child and environment at exactly 5 minute-interval, including:
  - Motor activity
  - Social activity
  - Emotional state
  - Caregiver
- Breastfeeding
- Administration of tablet
- Feeding of LNS
- Feeding

14) Form N15: Hemoglobin at 18 months

- Hemoglobin. Repeated if Hb < 5.0 g/dL
- Reported medical visit to health center (CSPS, Hospital) for illness during previous 9 mo.
- Reported hospitalization (CSPS, Hospital) during previous 9 mo.

15) Form N15b: Retrospective adverse event collection after 18 months (among children who did not attend 18 month follow up visit)

- Birthdate
- Reason of not attending 18 months follow up visit (if consent withdrawn, stop interview)
- Reported hospitalization (CSPS, Hospital) during previous 9 mo
- Number of visits to health center since enrollment
- Reported medical visit to health center (age, location, reason for visit)
- Death (date, location and reported reason)

16) **Form N16a: Serious adverse event reported by field agent (morbidity follow-up)**

- Reported hospitalization (location and date)
- Date of discharge from the hospital
- Death (date and location)

17) **Form N16b: Serious adverse event reported by study physician (morbidity follow-up)**

- Clinical course of the serious adverse event including:
  - Starting date of symptoms
  - Description of symptoms
  - Received treatment prior to hospitalization
  - Development of symptoms during hospitalization
  - Diagnosis and received treatment during hospitalization
- Evaluation of serious adverse event by study physician including
  - Duration of symptoms
  - Did the adverse event stop after stopping the study supplements?
  - Did the adverse event reappear after study supplements were reintroduced?
  - Outcome of the adverse event to date:
  - If fatal, date of death
- Conclusion of study physician
  - Classification of event as severe adverse event

- Categorization of serious adverse event
- Causality to the study
- Withdrawal of participant
- Participant continuing with study supplement

18) **Form N17a and N17b: Malaria slide microscopic reading**

- Slide date and study contact type (enrollment, biochemistry or morbidity)
- Amount of leukocytes
- Density of malaria parasites
- Type of *Plasmodium*

19) **Form N18a: Lab results of capillary ZPP**

20) **Form N18b: Lab results of washed venous ZPP**

21) Form N19: invitation for sample collection at 9 and 18 months (biochemistry subgroup)

- Confirmation of absence of fever and diarrhea during previous 48 hours
- Confirmation of no reference of child for treatment, except for anemia or moderate malnutrition
- Confirmation of child invitation for biological sample collection
- Household salt collection and distribution of dispositive for collection.

22) Form N21: Anthropometric follow-up visit at 18 months:

- Weight of child (±0.01kg) in duplicate. Repeated if disagreement > 0.1 kg.
- Length of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- Mid upper arm circumference of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.
- Head circumference of child (±0.1 cm) in duplicate. Repeated if disagreement > 0.5 cm.

23) **Form N22: Verbal autopsy the study physician**

- Date and place of death
- Child's age at time of death
- Hospital admission (location and date)
- History of disease leading to death
  - Description by caregiver of illness leading to death
  - Reported symptoms

7 Appendix B: Developmental outcomes at age 18 months

7-1 Contents

7-2 Study objectives

The main aim of the iLiNS-ZINC study was to assess zinc-related biochemical and functional responses among young Burkinabe children with a presumed high risk of zinc deficiency who receive micronutrient products (lipid-based nutrient supplements (LNS) or zinc tablet) containing different amounts of zinc, provided with or between meals from age 9 to 18 months, and to compare the same outcomes among children who do or do not receive LNS, a zinc tablet or selected health services from age 9 to 18 months.
The aim of the analyses described in this appendix is to compare infants in 4 different intervention groups: dietary supplementation with LNS containing 10 mg zinc per day with a placebo tablet (LNS-Zn10), LNS containing 0 mg zinc per day with a placebo tablet (LNS-Zn0), LNS containing 0 mg zinc per day with a tablet containing 5 mg zinc per day (LNS-ZnTab5), or nothing (non-intervention) on18-month motor development, language development, personal-social development, and interaction with caregivers

7-3 Hypotheses to be tested

1. 18-month scores in motor development, language development, personal-social development, and interaction with caregivers of infants provided with either 5 or 10 mg zinc per day in LNS or 5 mg zinc tablet per day provided between meals (and LNS to which no zinc has been added) will be greater than that of infants who receive LNS that does not contain zinc and a placebo tablet.

2. 18-month scores in motor development, language development, personal-social development, and interaction with caregivers of infants receiving LNS (with or without added zinc) from age 9 to 18 months will be greater than that of infants in the non-intervention cohort.

3. Hypotheses 1 and 2 will also be examined with regard to the prevalence of children in the bottom decile and bottom quartile of scores in motor development, language development, and personal-social development.

7-4 Definition of the 18-month developmental outcomes

The motor score is calculated as the sum of 32 Developmental Milestones Checklist II (DMC-II) motor items (*Form N24 Q 5.1 through 6.11, excluding Q 6.10*), each scored 0 (skill not yet acquired), 1 (emerging ability), or 2 (established ability). The bottom decile is the bottom 10% of our sample. The bottom quartile is the bottom 25% of our sample.
Language development is quantified as

a. The language score, calculated as the sum of the 16 DMC-II language items (*Form N24 Q 7.1 through 7.16*), each scored 0, 1, or 2. The bottom decile is the bottom 10% of our sample. The bottom quartile is the bottom 25% of our sample.

b. Expressive vocabulary > 10 words vs. <= 10 words, derived from *Form N24 Q 7.12*.
c. Word combining (Has the child started combining two words together into sentences? 0 = not yet, 1 = emerging ability, 2 = established ability) *Form N24 Q 7.14*

Personal-social development is calculated as the sum of the 28 DMC-II personal-social items (*Form N24 Q 8.1 through 8.30, excluding Q 8.6-8.7)*. The bottom decile is the bottom 10% of our sample. The bottom quartile is the bottom 25% of our sample.
Interaction with caregivers is calculated as the sum of the activities with adults in the past three days (*Form N23 Q 7.1, 8.1, 9.1, 10.1, 11.1, 12.1*).

7-5 Basis for the analysis: Intention to treat

The basis for the analysis and analysis principles will be the same as that for the primary outcomes, described in section 2.9 of the Statistical Analysis Plan.

7-6 Sample Size

We randomly selected a sub-sample from each of the four targeted intervention groups (LNS-Zn0; LNS-Zn10; LNS-ZnTab5; non-intervention) for comprehensive 18-month developmental assessment. We powered the study to detect an effect size of 0.3 *SD*. For the intervention groups the basic sample size was 244 per group, plus 20% attrition, we targeted 305 per group for comprehensive 18-month developmental assessment. For the delayed intervention group, the basic sample size was increased by one third, which was 325 plus 20% attrition, we targeted 407 children.

7-7 Hypothesis testing

7-7-1 Comparison of the developmental scores at 18 months of age between intervention groups versus the control group (non-intervention)

We will use mixed effects models (SAS PROC MIXED) with a random effect of village (to account for cluster randomization) and fixed effects of intervention group (with two levels: IC versus NIC) and any covariates, as specified in section 7.6, to test for differences between the two intervention groups. The null-hypothesis of the intervention having no impact on development will be rejected if the effect of intervention group yields a p-value < 0.05.

7-7-2 Comparison of the LNS-Zn0, LNS-Zn10, and LNS-ZnTab5groups

We will use mixed effects models (SAS PROC MIXED) with a random effect of village (to account for cluster randomization) and fixed effects of intervention group (with 4 levels: 3

intervention groups and 1 non-intervention group) and any covariates, as specified in section 7.6, to test for differences between the four intervention groups.
If the group effect is significant at the level of $p < .07$, we will use Tukey-Kramer's test for post-hoc pairwise comparisons between each intervention group and the control group (non-intervention). We will also use Tukey-Kramer's test for post-hoc pairwise comparisons between each group that received zinc (LNS-Zn10 and LNS-ZnTab5) and the LNS-Zn0 group. The null-hypothesis of zinc having no impact on development will be rejected for each comparison that yields a Tukey-Kramer's adjusted p-value < 0.05.

7-7-3 Prevalence of children in the bottom decile and bottom quartile

We will use logistic regression models (SAS PROC GLIMMIX), following the same approach outlined in sections 6.1 and 6.2.

7-8 General notes on statistical methods

7-8-1 Software

SAS for Windows Release 9.3 (Cary, NC) will be used for all analyses.

7-8-2 Calculating scores and z-scores

If a large percentage of data is missing for any item, we will exclude that item from the total score. For all other missing item scores, we will impute the scores based on the other items in the same subscale. We will use the imputation method described in Raghunathan et al. (2001).
Z-scores of developmental variables will be calculated based on the distribution of the iLiNS-ZINC sample, by standardizing the distribution to a mean of 0 and standard deviation of 1.

7-8-3 Multiple comparisons

The Tukey-Kramer adjustment method is used.

7-8-4 Outliers

Outliers will be handled in the same as for the primary outcomes, described in section 4.3 of the Statistical Analysis Plan.

7-8-5 Interaction and effect modification

We will follow the same procedure as specified for the primary outcomes to examine effect modifiers, as described in section 4.9 of the Statistical Analysis Plan.
We will examine the following effect modifiers, as described in section 4.6 of the Statistical Analysis Plan:

1. Baseline child LAZ < -1.5
2. Baseline child LAZ below median
3. Child sex
4. Maternal height at enrollment (continuous and categorical)

5. Maternal BMI at enrollment (quartiles or tertiles) if test of correlation with the composite variable for food security shows a non-colinearity
6. Proxy for food insecurity at enrollment (HFIAS, adjusted for season)
7. Proxy for hygiene/water quality at enrollment
8. Maternal education at enrollment
9. Maternal marital status/rank among wives at enrollment
10. Iron supplementation (as provided by the study along with deworming at enrollment)

In addition, we will examine the following effect modifiers

11. Baseline child WHZ score
12. Baseline child Hb concentration Family care indicators score
14. Adherence to supplementation

7-8-6 Covariate adjustment

For each outcome, we will examine the following three models:

1. No covariate adjustment
2. Adjustment for child age at developmental assessment
3. Adjustment for child age at developmental assessment, and for any of the variables listed below showing statistically significant association (at $p<0.1$ level) with the developmental score

We will consider the following variables for inclusion, as described in section 4.7 of the Statistical Analysis Plan:

34. Baseline child LAZ score
35. Baseline child WAZ score
36. Baseline child WHZ score
37. Baseline LAZ < -1.5
38. Baseline child Hb concentration
39. Baseline child capillary ZPP (adjusted for malaria)
40. Child sex
41. Maternal height at enrollment
42. Maternal BMI at enrollment
43. Maternal education at enrollment
44. Maternal marital status/rank among wives
45. Number of children < 5 years old
46. Feeding practice variables at enrollment
47. Proxy for food insecurity (HFIAS adjusted for season)
48. Proxy for hygiene/water quality
49. Proxy for livestock
50. Month/year of enrollment
51. Iron supplementation/deworming at enrollment
52. Adherence to supplementation

53. Diarrhea prevalence (IC groups)
54. Malaria incidence (IC groups)
55. Fever prevalence (IC groups)
56. Data collector

In addition, we will consider the following variables:

57. Family care indicators score, if this score is not different between supplement groups.
58. Baseline maternal age

7-9 References

Raghunathan, T. E., Lepkowski, J. M., Van Hoewyk, J., & Solenberger, P. (2001). A multivariate technique for multiply imputing missing values using a sequence of regression models. Survey Methodology, 27(1), 85-95.