NutriVax-Measles: Increasing measles vaccination coverage through supplementation with an SQ-LNS incentive in children aged 6-23 months

STATISTICAL ANALYSIS PLAN	Version	
	1.0	10/09/2024

PROJECT INFORMATION

Sponsor	ALIMA - The Alliance for International Medical Action
Funding Agency	The Eleanor Crook Foundation and GAVI
Protocol Number	NCT06387511 (clinicaltrials.gov)
Phase	Phase III pragmatic parallel cluster randomized trial

	WRITTEN BY	**REVIEWED BY**	**REVIEWED BY**	**VALIDATED BY**
Name	Delphine GABILLARD	Minh HUYEN	Cécile CAZES	Renaud BECQUET
Position	Biostatistician	Biostatistician	Project manager	Coordinating investigator
Date and electronic signature	GABILLARD Signature numérique de GABILLARD Date : 2024.09.13 10:14:49 +02'00'	Minh HUYEN Signature numérique de Minh HUYEN Date : 2024.09.16 14:49:16 +02'00'	Cazes Signature numérique de Cazes Date : 2024.09.13 15:53:19 +02'00'	Renaud Becquet Signature numérique de Renaud Becquet Date : 2024.09.18 09:30:51 +02'00'

EVOLUTION TRACKING		
Version	**Date**	**Modification**
1.0	10/09/2024	Add other definition of zero dose

This statistical analysis plan contains a technical description of the statistical analysis[1]. It does not include analysis details for study objectives concerning the acceptability, feasibility, cost-effectiveness, which will be addressed in a separate document

ABBREVIATIONS

CCS	Caregiver cost survey
CHW	Community Health Workers
CI	Confidence Intervals
DDF	Denominator degrees of freedom
GLMM	Generalized linear mixed model
HF	Health Facility
HHS	Household Survey
ICC	Intra-cluster Correlation Coefficient
LS	Longitudinal follow-up survey
LGA	Local Government Areas
PCT	Pragmatic cluster Randomized Trial
PHCC	Primary Health Care Center
PHC	Primary Health Clinics
PPS	Probability Proportional to Size Sampling
SQ-LNS	Small-quantity lipid-based nutrient supplements

Table des matières

1 SUMMARY OF TRIAL PROTOCOL

1.1 HYPOTHESIS

The NutriVax-Measles study will test the hypothesis that a SQ-LNS supplementation program coupled with a standard Expanded Program on Immunization (EPI) in health facilities for children aged 6-23 months (NutriVax program) could increase vaccination coverage, particularly measles, after twelve months of implementation. NutriVax-Measles will also assess the barriers, facilitators and cost of NutriVax plus EPI compared to EPI alone in children aged 6-23 months. Thanks to NutriVax-Measles study, we will be able to see if SQ-LNS as an incentive for immunizations leads to cost-efficient benefits compared to EPI alone. Thus, it will build evidence about whether it is possible to achieve a double impact on increased vaccination coverage and improved malnutrition indicators.

1.2 Study objectives and endpoints

1.2.1 Objectives

Primary objective:
To estimate the effectiveness of a SQ-LNS mass supplementation program added to routine immunization program compared to routine immunization program alone in terms of measles vaccine coverage, after 12 months of program implementation, in children aged 12-23 months in the endline cross-sectional household survey.

Main secondary objective:
To estimate the effectiveness of a SQ-LNS mass supplementation program added to routine immunization program compared to routine immunization program alone in terms of measles vaccine coverage, after 12 months of program implementation, in children aged 6-12 months at inclusion in the longitudinal follow-up survey.

Secondary objectives:

- To estimate the effectiveness of a SQ-LNS mass supplementation program added to routine immunization program compared to routine immunization program alone in children aged 6-23 months, after 12 months of program implementation, in terms of 1) all other infant vaccines uptake, 2) timeliness of age-eligible vaccinations; 3) anthropometric status, 4) uptake of pediatric curative and preventive health consultations and activities.
- To assess the barriers and facilitators to SQ-LNS mass supplementation implemented as part of a routine immunization program in health facilities, from the perspectives of parents/legal guardian of children aged 6-23 months, health care providers, community health workers and community representatives.
- To assess the cost-efficiency of a SQ-LNS mass supplementation program versus routine immunization program in terms of cost per child supplemented and vaccinated.

1.2.2 Endpoints

The primary endpoint is the measles vaccine coverage, defined as the proportion of children aged 12-23 months in the endline survey who have received at least one dose of measles vaccine, as reported on their vaccination card.

The main secondary endpoint is the measles vaccine coverage, defined as the proportion of children aged 6-12 months at inclusion in the longitudinal follow-up survey, who have received at least one additional dose of measles vaccine, as reported on their vaccination card, administered between inclusion and the end of follow-up.

Secondary endpoints:

- Proportion of children with at least one measles vaccines as reported on their vaccination card or by recall.
- Timeliness of measles 1 vaccination (i.e. within 30 days of turning 9 months), as reported on their vaccination card.
- Timeliness of measles 2 vaccination (i.e. within 30 days of turning 15 months), as reported on their vaccination card.
- Proportion of children with each of the following vaccine doses, as reported on their vaccination card or by recall: measles 1, measles 2, pentavalent 1 and 3, yellow fever and meningitis.
- Proportion of children with age-eligible vaccines such as measles 1, measles 2, yellow fever, meningitis, and catch-up vaccines like pentavalent 1 and 3, as reported on a vaccination card or by recall.
- Proportion of zero-dose children as reported on a vaccination card or by recall, two definitions : none dose, and no penta 1 dose.
- Proportion of fully immunized children, i.e. children who received all age-eligible vaccination.
- Anthropometric parameters mean change in children: MUAC, z scores for MUAC for age, weight for age, weight for height, and height for age.
- Proportion of vitamin A dose 1 given at 6 months and dose 2 given at 12 months
- Evolution of aggregate data collected routinely at health facilities, mean and proportion: paediatric curative and preventive consultations, vaccination activities, SQ-LNS activities, activities on vaccination and Infant and Young Child Feeding (IYCF) sensitization, vaccine and SQ-LNS consumption.
- Barriers and facilitators to access health care centres and receive SQ-LNS mass supplementation implemented as a routine preventive program nested in health facilities activities.
- Adherence to SQ-LNS supplementation.
- Barriers and facilitators to vaccination uptake at health facilities activities.
- Cost per child vaccinated and supplemented compared to cost per child vaccinated only, using a facility and caregiver perspective (see 5.1.4 Cost-efficiency survey section for precision) to identify all provider and caregiver costs associated with treatment.

Table 1: Primary and secondary endpoints (vaccination coverage and anthropometric parameter) in the NutriVax-Measles study

Endpoints	Measurement variable	Denominator and data collection mode	Method of aggregation	Timepoint
Primary				
Measles vaccine coverage	At least one dose of measles vaccine reported on vaccination card	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
Main secondary				
Measles vaccine coverage	At least one additional dose of measles vaccine reported on vaccination card administered between inclusion and the end of follow-up	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	Between inclusion and the end of longitudinal follow-up
Secondary				
Measles vaccine coverage	At least one measles vaccine as reported on a vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
	At least one additional dose of measles vaccine reported on vaccination card or by recall, administered between inclusion and the end of follow-up	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
	Measles 2 vaccine as reported on a vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
Timeliness of measles 1 vaccination.	Measles 1 vaccine received within 30 days of turning 9 months by card	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
	Measles 1 vaccine received within 30 days of turning 9 months by card or by recall	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
Timeliness of measles 2 vaccination.	Measles 2 vaccine received within 30 days of turning 15 months by card or by recall	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
	Measles 2 vaccine received within 30 days of turning 15 months by card or by recall	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
Other infant vaccines coverage	Pentavalent 1 and 3, yellow fever, and meningitis vaccine by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
		Children aged 6-12 months at inclusion in	Proportion	End of follow-up

		the longitudinal follow-up survey		
Additional infant vaccines coverage	Catch-up pentavalent 1 and 3, and age-eligible yellow fever, meningitis, by vaccination card or by recall	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
Zero-dose	No measles + pentavalent + yellow fever + meningitis vaccines reported by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
	No pentavalent 1 vaccine reported by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
Proportion of children fully immunized	Children who had received all childhood vaccinations recommended by the Nigeria MOH, by vaccination card or by recall.	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up
Anthropometric parameters	MUAC, z scores for MUAC for age, weight for age, weight for height, and height for age	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Median/mean	End of follow-up
Vitamin A supplementation coverage	Dose 1 given at 6 month and dose 2 given at 12 months by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	Proportion	After one year of the SQ-LNS program (measured in the endline HHS)
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	Proportion	End of follow-up

1.3 Study design

This NutriVax-Measles study will consist in conducting a pragmatic parallel cluster randomized trial (PCT) with baseline measure with different populations and data collection modes: 1) a baseline and an endline cross-sectional household surveys (HHS) of children aged 12-23 months, 2) a longitudinal 12 months follow-up household survey (LS) of children aged 6-12 months at inclusion, 3) two cross-sectional Qualitative Feasibility and Acceptability Survey (QFAS) of parents/legal guardian of children aged 6-23 months, health care providers, community health workers, 4) caregiver cost survey (CCS) of the caregivers of children from the longitudinal cohort (population 2), and 5) a health facility cost survey of a randomized sub-sample of health facilities.

Thanks to these different data collection modes, measles vaccination coverage and the quantitative secondary endpoints will be measured both at the community level using the endline HHS and at the individual level using the LS (Figure 1).

The NutriVax program will be assessed within a routine program implemented at scale. PCT pragmatic studies are designed to guide decision-making in the real-world clinical practice and policy settings while maintaining the strengths of individual randomized controlled trials if they are systematically designed and performed properly. The pragmatic attitude increases external validity to ensure that the results can be generalized in everyday practice in this context and other similar settings[2,3].

The unit of randomization (i.e. cluster) will be a ward and its catchment area. We will include exhaustively all wards of each LGA where the study will take place.

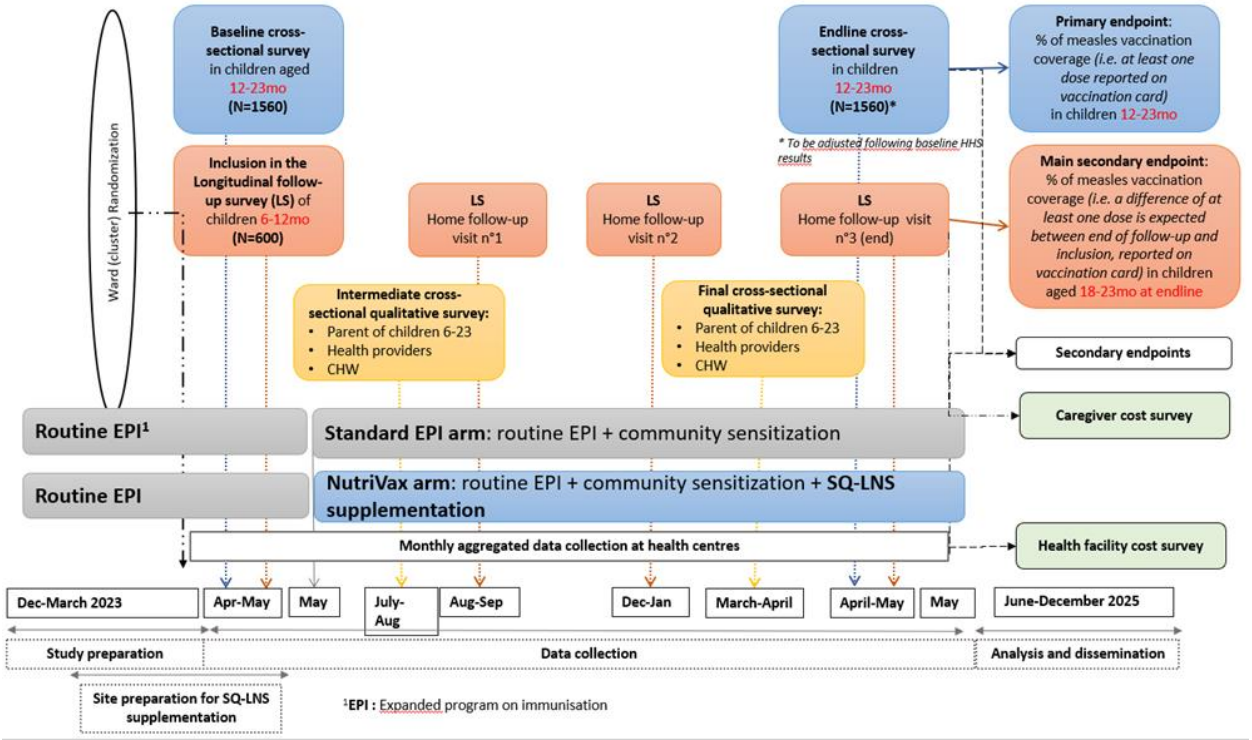

Figure 1. Study design chart.

1.4 Study population

1.4.1 Eligibility criteria

- Baseline and endline household surveys

Inclusion criteria

- Aged 12-23 months;

- With oral informed consent of parent or legal guardian;

- Residing in the catchment settlements of wards included in the study.

Exclusion criteria

- Already included in the longitudinal follow-up.

- Longitudinal follow-up survey

Inclusion criteria

- Aged 6-12 months;

- With written informed consent of parent or legal guardian;

- Residing in the catchment settlements of wards included in the study.

Exclusion criteria

- Acute malnutrition criteria as per the WHO definition (i.e. MUAC<125or WHZ < -2 or nutritional edema). Such children will be referred for treatment;

- Known medical complication that requires referral for hospitalization;

- Known allergies to SQ-LNS or SQ-LNS contraindication;

- Any other condition interfering with protocol adherence or the ability to give informed consent, in the judgment of the Field Investigator

For instance, medical, occupational or other conditions of the participant or parent/guardian may make routine home visits and evaluation difficult or make the child a poor candidate for retention. Any such potential exclusion must be validated by the Field Investigator.

1.4.2 Sampling methods

Both HHS and longitudinal follow-up surveys will be carried out at community level. Household and participants will be selected through a multi-stage sampling method. We will follow the WHO reference manual for vaccination coverage cluster surveys[4]. The sampling will be carried out in three stages in each randomized cluster (which will be a ward), namely: (1) random selection of 'settlement' in each cluster using probability proportional to size (PPS) method; (2) systematic random selection of households in each settlement with equal probability using population estimation used by the MoH for the planification of health activities; (3) selection of the youngest eligible child in the household, and random selection of one among the eligible child guardians (Figure 2).

We will use the following definition to identify a household: "A person or a group of persons, related or unrelated, who live together and share a common source of food and livelihood, and recognize one person as a head.", as per the usual definition used in population survey in Nigeria (26).

A specific and detailed standardized operation procedure for the sampling process will be developed prior to the baseline HHS and the LS inclusion.

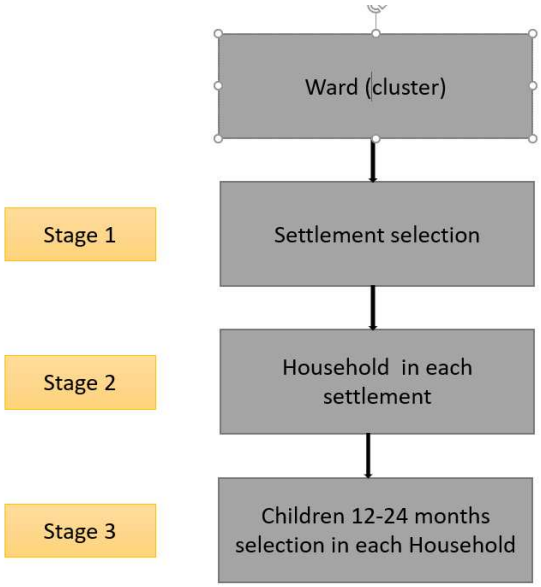

Figure 2 : Sampling survey using 3-stage random sampling method

The same sampling methods will be used for the inclusion of children in the HHS and for the inclusion of children in the LS.

1.5 Study interventions

The NutriVax-Measles study will compare two interventions randomly assigned to either:
1) The standard Expanded Program on Immunization (EPI) in children aged 6-23 months delivered according to the Ministry of Health's routine plans in the community and at health centres (i.e. PHCCs) and health posts, named the standard EPI arm,

2) The NutriVax program combining the EPI in children aged 6-23 months according to the Ministry of Health's routine plans in the community and at health centres (i.e. PHCCs) and health posts associated with SQ-LNS mass supplementation integrated into pre-existing services delivered at health centres (i.e. PHCCs) for children 6-23 months of age, named the NutriVax arm.

1.6 Study sites

Research and operational activities will take place at community level and in the 20 PHCCs of the 2 LGAs of Karasuwa and Nguru LGAs. Each LGA has 10 wards and there is one PHCC per ward.

1.7 Randomisation

The unit of randomization (cluster) will be the ward and its catchment area.
Randomization will be stratified by LGA in order to promote balance of the intervention among both LGAs and to give the same opportunity to receive the intervention in both LGAs. Also, the two LGAs may have different populations or contexts that could be a potential confounding factor.

Randomization will be done prior to the baseline survey, because of logistical constraints required for preparing SQ-LNS distribution at health centres (PHCCs), but kept concealed to the data collector until the completion of the baseline survey. After baseline survey completion, the study will be open, as it will not be possible to blind participants or study staff to SQ-LNS supplementation.

In each LGA, the 10 wards will be randomized the same day, by lottery from an envelope, in presence of PHCC department, local community representatives and held in the local language (Hausa). Clusters will be randomly allocated at a ratio of 1:1 either to the standard EPI arm or to the NutriVax arm.

1.8 SAMPLE SIZE

1.8.1 HHS – primary endpoint

The primary endpoint will be collected among children aged 12-23 months included in the endline HHS about 12 months after the baseline HHS. Based on a baseline measles vaccination (i.e. at least one dose) coverage reported on vaccination card of 30% [4], a power of 90%, and an intra-cluster correlation of 0.2, an alpha risk of 5%, it will be necessary to include 74 children per cluster. With a rate of unusable data of 5%, we will need 78 children per cluster or a total of 1,560 children to demonstrate a minimum of +10% points difference between the NutriVax and the standard EPI arms, in measles vaccination coverage, i.e. the primary outcome. This calculation was performed using the Shiny CRT calculator used specifically to estimate sample size for cluster randomised trials[5].

1.8.2 LS - Main secondary endpoint

The main secondary endpoint will be collected among the children aged 6-12 months at baseline included in the LS. Based on the baseline measles vaccination coverage reported on vaccination card of 30% [4], a power of 80%, and an intra-cluster correlation coefficient of 0.2, an individual auto-correlation of 50%, an alpha risk of 5% and a rate of unusable data of 5%, it will be necessary to include 30 children per cluster or 600 children in total to demonstrate a minimum of +10% points difference between arms in measles vaccination coverage. This calculation was performed using the Shiny CRT calculator used specifically to estimate sample size for cluster randomised trials[5].

1.8.3 Precision

These two estimates were validated by the senior statistician for this study but they may need to be adjusted thereafter according to the actual intra-cluster correlation coefficient and the baseline measles vaccination proportion results from the baseline survey.

2 STATISTICAL ASPECTS

2.1 STUDY SAMPLES FOR ANALYSIS

2.1.1 Intention-to-treat sample

Baseline HHS was already carried out in all randomised clusters by the time this SAP was written. All randomised clusters will be included in the planned analysis.

All individuals with available endpoint data will be included in the planned analyses.

2.1.2 Per-protocol sample

All individuals with available endpoint data and meeting criteria for study participants, as defined in section 1.4, will be included in the planned analyses.

2.2 STATISTICAL APPROACH

Variables will be described overall, by arm, by arm and by LGA, by arm and by cluster using appropriate summary statistics for the variable type. For quantitative variables, means, standard deviations, medians, interquartile ranges, and ranges values will be given. Data may be transformed or normalized as necessary based on review of the data distribution.

For qualitative variables, absolute numbers and percentages will be given, and 95% binomial proportion confidence intervals (CI) will be calculated; the exact method will be used when appropriate.

Categorical and discrete data may be aggregated into classes depending on frequency. Statistical testing between arms or subgroups will be carried out using the appropriate statistical test for the variable type (χ^2, corrected χ^2 or the Fisher's exact test for categorical variables, depending on the observed frequencies and Student's t test, Kruskal Wallis test or the Wilcoxon test, depending on the distribution for continuous variables).

2.3 CALCULATION CONVENTIONS FOR STATISTICAL ANALYSES

2.3.1 Missing data management

For the cross-sectional survey with endpoints using data reported on vaccination card, we have 1 type of missing data: children without vaccination card.

For the longitudinal with endpoints using data reported on vaccination card, we have 2 types of missing data: children without vaccination card and children loss-to-follow-up.

For the longitudinal with endpoints using data reported on vaccination card or recall we have 1 type of missing data : children loss-to-follow-up.

We will describe these missing data.

For all endpoints using data reported on vaccination card, if the children don't have a vaccination card then vaccination will be considered as failure.

For the longitudinal survey, at first, analyses will be performed on available data only, so without loss-to-follow-up. The endpoint will be measured at the end of follow-up, so a child may be loss-to-follow-up at M12, but at the end of follow-up may have received vaccine, and therefore be considered as in success for an endpoint.

A particular case is that of a child who does not have a vaccination card at inclusion and who has a vaccination card during follow-up, so if he is vaccinated during follow-up, he will be considered as a success.

In addition, we will perform sensitivity analyses to assess the potential impact of missing data on the study conclusions, using the most extreme scenarios (like described in chapter 3.4.2.2).

2.3.2 Type I error

We will use a two-sided type I error α of 5% to compare endpoints between arms without any correction for multiple comparisons.

2.3.3 Statistical significance

The level of statistical significance will be $p<0.05$, and estimates will be reported together with their 95% CI.

2.4 SOFTWARE

We will use either SAS Enterprise Guide version 8.3 or higher, or R software versions 4.2.2 or higher to prepare datasets, and conduct statistical analyses.

2.5 REPORTING

We will follow the Consort 2010 statement: extension to cluster randomised trials[6] for reporting the results of the following analyses, including reporting ICC of the primary endpoint.

3 ANALYSES PLAN

3.1 CLUSTER CHARACTERISTICS

3.1.1 Before baseline cross-sectional survey

The following characteristics of clusters collected before the baseline cross-sectional survey will be described between arms, between LGA, between LGA*arm*clusters.

- Number of villages
- Population
- Number of Households
- Number of days with no cold chain
- Data aggregated of OPD (Out-patient department): total consultation of children aged 12-59 months ; and OTP (Out-patient therapeutic program) register: total number of children 6-59 months admitted.

- Data aggregated of IYCF register: total number of mothers of child 6-23 months sensitized on IYCF practices.
- Data aggregated of Health facility nutrition/growth monitoring and promotion register: total number of children 6-59 months receiving Vitamin A; total number of children 12-59 months receiving deworming

3.1.2 Baseline measles vaccination coverage

The baseline coverage level will be calculated globally, by LGA, by cluster, by arm : $\frac{Number\ of\ participants\ with\ endpoint}{Number\ of\ participants\ responding\ to\ the\ HHS}$

When used for the analyses, it will be affected to all participant belonging to the same ward, under the likely hypothesis that all participant in the ward area do not attend another ward.

We will estimate the baseline ICC and its 95% CI from our data by performing various calculations using the R ICCbin library[7] or R ICCest library. First, we will use the Anova estimator (option R ICCbin ci.type=aov), that is originally proposed for continuous data but that is also used for binary data[8]. Then, we will use the Fleiss-Cuzick estimator that is a kappa-type estimator (option R ICCbin ci.type=fc) and the Pearson estimator that is based on direct calculation of the correlation between observations within each cluster (option R ICCbin ci.type=peq). If the results are different, we will keep the more conservative calculation, so the highest ICC estimator.

If necessary, we will refine thereafter the sample size of the endline cross-sectional survey according to the actual intra-cluster correlation coefficient and the baseline measles vaccine coverage proportion results from the baseline HHS survey.

3.1.3 After baseline cross-sectional survey

The following characteristics of clusters collected at the start of SQ-LNS supplementation will be described in NutriVax arm only, globally, between LGA, between LGA*clusters:

- Aggregated data from the SQ-LNS supplementation programme register (NutriVax arm only) : total number of new admissions to the SQ-LNS supplementation program, new ; Total number of new admissions to the SQ-LNS supplementation program, OTP cured ; Total new admissions to the SQ-LNS supplementation program (new and OTP cures), children 6-11 months ; Total new admissions to the SQ-LNS supplementation program (new and OTP cures), children 12-23 months ; Total number of children discharged from the SQ-LNS supplementation program ; Total number of sachets of SQ-LNS distributed ; Total Number of days out of stock of SQ-LNS.
- Aggregated data from the child immunization register (both arms): total doses of different vaccines, total number of days of stock of measles vaccines and others vaccines.

3.2 SURVEY PARTICIPANTS CHARACTERISTICS

The following participant characteristics will be described both for the longitudinal study, for the cross-sectional baseline survey and for the endline cross-sectional survey, by arms, by LGA, by LGA*arm*clusters.

No adjustment for clustering will be applied when summarising the baseline characteristics.

3.2.1 Respect of eligibility criteria

Analyses to be done in order to check compliance with the eligibility criteria are described below.

- Inclusion criteria for the baseline and for the endline surveys:
  - **Residence**: check that all the participants residing in the place where the survey is taking place
  - **Age**: check that all the participants are 12-23 months old
  - **Oral consent**: check that oral informed consent form is given by the child's a parent or legal guardian
- Non-inclusion criteria for the baseline and for the endline surveys
  - **Included in the cohort:** check that all participants are not included in the cohort of the longitudinal survey

3.2.2 Characteristics of children participating in the cross-sectional baseline survey

Variables	**Modalities**
Demographic characteristics	
Age	In months
Sex	Categories
Settlement	Categories
Sibling and number	Binary Yes/No; quantitative
Immunization	
Vaccination card or a family folder	Categories
Different vaccines and doses (BCG, HepB-0, OPV-0, OPV-1, PCV-1, Penta-1, Rota-1, IPV-1, OPV-2, PCV-2, Penta-2, Rota-2, OPV-3, PCV-3, Penta-3, Rota-3, IPV-2, Measles-1, Yellow fever, Meningitis, Measles-2, Vitamin A-1, Vitamin A-2, Other)	Binary Yes/No
Age at which vaccines were administered	In months
Child has received all the vaccines according to caregiver	Categories
Child has received vitamin A supplementation over the last 6 months	Binary Yes/No
Anthropometric data	
Middle upper arm circumference (MUAC)	Quantitative and/or relevant classes

Nutritional edema	Categories
Child breastfeeding	Binary Yes/No
Medical history	
Chronic disease	Binary Yes/No
Allergies	Binary Yes/No
Hospitalization event in the last 3 months	Binary Yes/No
Child has received deworming supplementation over the last 6 months	Categories
Child currently receiving treatment with RUTF	Binary Yes/No
Child currently receiving treatment with RUSF	Binary Yes/No
Actual medical problem	
Fever in the last 24 hours	Binary Yes/No
Current presence of different pathologies (non-bloody diarrhoea, suspected anaemia, suspected malaria, respiratory infection, vomiting, dehydration, parasitosis, dermatosis, upper respiratory infection, non-malarial fever, abscess, other)	Binary Yes/No
Number of days with different pathologies	Quantitative

3.2.3 Characteristics of families of children participating in the baseline cross-sectional survey

Variables	**Modalities**
Family demographic characteristics	
Number of people in the household	Quantitative
Demographic characteristics of fathers	
Alive	Binary Yes/No
Age	In years and/or relevant classes
Literacy	Binary Yes/No
Main activity	Categories
Native language	Categories
Demographic characteristics of mothers	
Number of births	Quantitative
Alive	Binary Yes/No
Age	In years and/or relevant classes
Literacy	Binary Yes/No
Main activity	Categories
Marital status	Categories
For married, number of domestic partners	Quantitative
Native language	Categories
Legal representative demographic characteristics	
Accompanist	Categories
Legal representative	Categories
Age	In years and/or relevant classes
Literacy	Binary Yes/No

Education level	Categories
Native language	Categories
Time living in settlement	In months and/or relevant classes
Medical	
Participated in training session for using a MUAC	Binary Yes/No

3.2.4 Characteristics of children participating in the longitudinal survey

The same variables as those used for children participating in the HHS cross-sectional surveys will be described, with additional variables:

Variables	**Modalities**
Anthropometric data	
Weight	Quantitative and/or relevant classes
Length/Height	Quantitative and/or relevant classes
Weight for Length/Height z-score	Quantitative and/or relevant classes
Weight for age z-score	Quantitative and/or relevant classes
Length/Height for age z-score	Quantitative and/or relevant classes
Household hunger score	
Household hunger score	Quantitative
Household hunger score conclusion	Categories
Nutrition data	
Minimum dietary diversity score	Quantitative and/or relevant classes
Breastfeeding	Binary Yes/No
SQ-LNS supplementation	Binary Yes/No
SQ-LNS consumption during the previous 7 days	Quantitative and/or relevant classes
SQ-LNS sharing if SQ-LNS supplementation is yes	Categories
Reasons for sharing SQ-LNS if SQ-LNS sharing is yes	Categories
Seasonal malaria prevention treatment received	Binary Yes/No
Months when Seasonal malaria prevention treatment was received	Categories
Cost-effectiveness	

3.3 BASELINE RESULTS DESCRIPTION

At the end of the HHS baseline cross-sectional survey, all primary and secondary endpoints will be described at cluster level.

Individual characteristics will be described between arms to assess for sample representativity and possibly identify imbalances. Since the allocation of clusters between arms is carried out randomly, it is known that any differences that do occur must have occurred by chance. This means that the null hypothesis is known to be true, so there is no logic in testing it. The point of displaying between-arm comparisons is not to carry out a significance test, but to describe in quantitative terms how large any differences were.[9(p200)]

We will use the findings of the baseline analysis to decide which variables need to be adjusted for in the final analysis.

3.4 STRATEGY OF ANALYSIS

All endpoints in NutriVax intervention will be compared to the standard arm, using appropriate model. No weighting will be applied for the PPS sampling.

3.4.1 Analysis of the primary endpoint:

The primary endpoint is the proportion of children with at least one dose of measles vaccine reported on vaccination card, measured during the endline cross-sectional household survey.

If the children don't have a vaccination card then this endpoint will be considered as failure.

3.4.1.1 Main analysis first intention:

In first intention, analysis will be carried out at the individual level, adjusting for clustering and for baseline assessment of outcome in that cluster.

Analysis using the endline cross-sectional household survey will be an individual-level analysis approach, using a logistic regression with random effects (a member of the family of generalised linear mixed model GLMM) with:

- a binomial distribution if the events/trials data syntax is used, or if necessary a binary distribution if an individual-level data syntax is used, depending on adjustments variables if the individual level is necessary or due to problematic of boundary effects with the binomial distribution.[9(p192),10]

- a logit link function;

- study arm as fixed effect;

- adjustment by cluster level baseline coverage estimate (all patients of a cluster will have the same proportion of vaccination coverage);

- ward (cluster) as random intercept term.

The intervention effect will be estimated as the cluster-specific odds ratio (OR) (with 95% CI) of vaccine between the intervention and the control arm.

The Kenward-Rodger correction[11] will be applied to estimate the number of degrees of freedom as it has been shown to improve estimation with a small number of clusters[12–14].

The ICC at endline will be reported. It will be calculated using the same methods as the ICC at baseline (see chapter 3.1.2). It will also be stratified by study arm.

Additional models will be considered for sensitivity analyses:

- Re-running the same model but without using the Kenward-Roger correction, using between-within method for DDF[12].
- Model with an additional nested random effect term (LGA : cluster within LGA), or adjusted on LGA as fixed effect.

Adjusted analyses

The logistic regression with random effects will be re-run after adjustment for the individual baseline covariates selected at baseline analysis. We will also adjust on cluster covariates:

- Number of days with no cold chain;
- Number of days of stock of measles vaccines in classes.

It will be performed to identify individual and contextual factors associated with measles vaccine coverage.

3.4.1.2 Main analysis second intention:

In second intention, in case the model does not converge due to low numbers of clusters, we will use the cluster-level analysis. Individual data will be aggregated by cluster as summary statistics for a cluster-level analysis approach with a Student or Wilcoxon test depending on the endpoint distribution to determine the intervention effect.[9(p197),15]

Assumptions will be verified and alternative approach will be considered in case assumptions are not met.

3.4.1.3 Sensitivity analysis:

We are planning to perform the following two sensitivity analyses:

1) Difference-in-difference (DiD) analysis

To provide a robustness check for the results, a difference-in-difference (DiD) analysis will be performed as an impact analysis.

This DiD analysis calculates the effect of the intervention on the outcome by comparing the average change over time in the outcome variable for the intervention arm to the average change over time for the control arm.

DiD focuses on estimating the average treatment effect by comparing changes over time.

The DiD estimate will be calculated using a logistic regression with the period, the arm and an interaction between period and arm as covariates. We will also adjust on selected covariates. Results of DiD in percentage points (pp) and in relative odds ratio (ROR) (with 95% CI) will be reported.

Should the main analysis and this DiD analysis yield to similar findings, that will increase confidence in the validity of our conclusions.

2) Sub-group analysis
Excluding children aged 21-23 months in the cross-sectional household surveys.

This sensibility analysis is planned because these children could have one dose of measles vaccine, but administered before the intervention period.

3.4.2 Analysis of the main secondary endpoint:

The main secondary endpoint is the measles vaccine coverage, defined as the proportion of children aged 6-12 months at inclusion in the longitudinal follow-up survey, who have received at least one additional dose of measles vaccine, as reported on their vaccination card, administered between inclusion and the end of follow-up.

3.4.2.1 Description

We will describe at each visit, globally and by age classes at inclusion ([6-9[months, [9-12] months):

- proportion of measles vaccine dose 1,
- proportion of measles vaccine dose 2,
- proportion of patients of at least one additional dose of measles vaccine.

For children included at [6-9[months old, measles vaccine dose 1 is supposed to be administered before the first follow-up visit (i.e V1) occurring 4 months after inclusion (i.e M4); measles vaccine dose 2 is supposed to be administered before V2 (M8) if included at [7-9[months old or before V3 (M12) if included at [6-7[months old.

For children included at [9-12] months old, measles vaccine dose 1 is supposed to be administered before inclusion, and measles vaccine dose 2 is supposed to be administered before V1 (M4) if included at [11-12] months old or before V2 (M8) if included at [9-11[months old.

3.4.2.2 Main analysis first intention:

In first intention, analysis will be conducted at the individual level, adjusting for clustering.

Analysis using an individual-level analysis approach, using a logistic regression with random effects (a member of the family of generalised linear mixed model GLMM) with:

- a binary distribution (we analyse the change in the endpoint at individual level at the end of the follow-up)

- a logit link function;

- study arm as fixed effect;

- ward (cluster) as random intercept term.

The intervention effect will be estimated as the cluster-specific odds ratio (OR) (with 95% CI) of vaccine between the intervention and the control arm.

We won't adjust for the baseline value because the number of doses expected depends on the inclusion age. For children aged 6-9 months at inclusion, it is normal to have 0 dose at inclusion.

The Kenward-Rodger correction11 will be applied to estimate the number of degrees of freedom as it has been shown to improve estimation with a small number of clusters12–14.

The ICC will be reported. It will be calculated using the same methods as the ICC at baseline of primary endpoint (see chapter 3.1.2). It will also be stratified by study arm.

Additional model will be considered for sensitivity analyses:

- Re-running the same model but without using the Kenward-Roger correction, using between-within method for DDF[12].

Adjusted analyses

The logistic regression model will be re-run after adjustment on a selection of baseline covariates, with additional variables selected by the results of the qualitative survey.

We will adjust on the following individual baseline covariates:

- Mother's age;
- Study level;
- Number of children in the family;
- Distance of home from health facility;
- Level of food insecurity.

And on the following cluster baseline covariates:

- Number of days with no cold chain
- Number of days of stock of measles vaccines

Handling of missing data

The primary analysis will use all available data with no imputation, i.e. without loss-to-follow-up.

In addition, we will perform sensitivity analyses to assess the potential impact of missing data (for children loss-to-follow-up before additional vaccination) on the study conclusions[16]. We will start by re-running the Model described above under the following two most extreme scenarios:

- Extreme scenario 1 ('worst' case for intervention): all patients with missing data assumed as failure (no additional dose of measles vaccine) in the Nutrivax arm and success (at least one additional dose of measles vaccine) in the standard arm.
- Extreme scenario 2 ('best' case for intervention): all patients with missing data assumed as success (at least one additional dose of measles vaccine) in the Nutrivax arm and failure (no additional dose of measles vaccine) in the standard arm.

We will check whether the study conclusion changes as indicated by the resulting p-values. In the case where both extremes result in the same statistical significance as the main analysis, no further tipping point analysis will be performed. In case the conclusions disagree, we will perform a tipping point search by identifying combinations where the level of significance switches from significant to non-significant (or vice versa) as described by Yan et al.[17]

3.4.2.3 Main analysis second intention:

In second intention, in case the model does not converge due to low numbers of clusters, we will use the cluster-level analysis. Individual data will be aggregated by cluster as summary statistics for a cluster-level analysis approach with a Student or Wilcoxon test depending on the endpoint distribution to determine the intervention effect.

Assumptions will be verified and alternative approach will be considered in case assumptions are not met.

3.4.3 Analysis of secondary endpoints:

3.4.3.1 Anthropometric endpoints:

We will analyse anthropometrics parameters mean change in children included in the longitudinal follow-up survey: MUAC, z scores for MUAC for age, weight/length for age, weight for weight/length, and height for age, as follow.

We will describe evolution at each visit of each parameter. And also, the evolution in absolute change at each visit of each parameter compared to the baseline measure.

In first intention, analysis will be conducted at the individual level, adjusting for clustering.

Analysis using an individual-level analysis approach, using a mixed effects linear regression with:

- a normal distribution,

- an identity link function,

- study arm as fixed effect,

- ward (cluster) as random intercept term,

- random intercept to account for within-individual correlation nested inside ward over time.

This model will be for the change in the endpoint of interest at different timing with baseline value.

The intervention effect will be estimated as the difference of means of delta (with 95% CI) between the intervention and the control arm.

The Kenward-Rodger correction[11] will be applied to estimate the number of degrees of freedom as it has been shown to improve estimation with a small number of clusters[12–14].

We will adjust by individual baseline parameter estimate and by cluster baseline mean parameter estimate.[14,18]

Assumptions will be verified and alternative approach will be considered in case assumptions are not met.

In case of non-normal distribution of the endpoint, categorization into classes will be considered. If the outcome is binary, we will use logistic regression with random effects; if the

outcome is a nominal with more than two classes, we will fit multinomial model with random effects[10,19], or we will keep continuous data using a transformation or a generalized additive mixed model GAMM [20,21].

3.4.3.2 Others secondary endpoints

Table 2 summarises each remaining secondary endpoint and the planned analyses.

All endpoints in the NutriVax arm will be compared to the control arm, using appropriate model.

Concerning the longitudinal follow-up survey, for secondary endpoints using data reported on a vaccination card or by recall, there is only 1 type of missing data: children loss-to-follow-up.

In these models, we won't adjust for the baseline value because the number of doses expected depends on the age at inclusion. For children aged [6-9[months at inclusion, it is normal to have 0 dose of measles at inclusion.

Table 2. Summary of secondary endpoints and planned analyses

Endpoints	Measurement variable	Denominator and data collection mode	Timepoint	Method
Secondary				
Measles vaccine coverage	At least one measles vaccine as reported on a vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
	At least one additional dose of measles vaccine reported on vaccination card or by recall, administered between inclusion and the end of follow-up	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
	Measles 2 vaccine as reported on a vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
Timeliness of measles 1 vaccination.	Measles 1 vaccine received within 30 days of turning 9 months by card	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
	Measles 1 vaccine received within 30 days of turning 9 months by card or by recall	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2

Timeliness of measles 2 vaccination.	Measles 2 vaccine received within 30 days of turning 15 months by card or by recall	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
	Measles 2 vaccine received within 30 days of turning 15 months by card or by recall	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
Other infant vaccines coverage	Pentavalent 1 and 3, yellow fever, and meningitis vaccine by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
Additional infant vaccines coverage	Catch-up pentavalent 1 and 3, and age-eligible yellow fever, meningitis, by vaccination card or by recall	Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
Zero-dose	No measles + pentavalent + yellow fever + meningitis vaccines reported by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
	No pentavalent 1 vaccine reported by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
Proportion of children fully immunized	Children who had received all childhood vaccinations recommended by the Nigeria MOH, by vaccination card or by recall.	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
		Children aged 6-12 months at inclusion in the longitudinal follow-up survey	End of follow-up	Like 3.4.2.2
Vitamin A supplementation coverage	Dose 1 given at 6 month and dose 2 given at 12 months by vaccination card or by recall	Children aged 12-23 months in the cross-sectional endline survey	After one year of the SQ-LNS program (measured in the endline HHS)	Like 3.4.1.1
		Children aged 6-12 months at inclusion in	End of follow-up	Like 3.4.2.2

		the longitudinal follow-up survey		

4 REFERENCES

1. 2006 - E 9 Statistical Principles for Clinical Trials.pdf.

2. MacPherson H. Pragmatic clinical trials. *Complement Ther Med*. 2004;12(2-3):136-140. doi:10.1016/j.ctim.2004.07.043

3. Treweek S, Zwarenstein M. Making trials matter: pragmatic and explanatory trials and the problem of applicability. *Trials*. 2009;10(1):37. doi:10.1186/1745-6215-10-37

4. World Health Organization Vaccination Coverage Cluster Surveys: Reference Manual. Accessed June 6, 2024. https://www.who.int/publications-detail-redirect/WHO-IVB-18.09

5. Hemming K, Kasza J, Hooper R, Forbes A, Taljaard M. A tutorial on sample size calculation for multiple-period cluster randomized parallel, cross-over and stepped-wedge trials using the Shiny CRT Calculator. *Int J Epidemiol*. 2020;49(3):979-995. doi:10.1093/ije/dyz237

6. Campbell MK, Elbourne DR, Altman DG. CONSORT statement: extension to cluster randomised trials. *BMJ*. 2004;328(7441):702-708. doi:10.1136/bmj.328.7441.702

7. Chakraborty H, Hossain A. R package to estimate intracluster correlation coefficient with confidence interval for binary data. *Comput Methods Programs Biomed*. 2018;155:85-92. doi:10.1016/j.cmpb.2017.10.023

8. Wu S, Crespi CM, Wong WK. Comparison of methods for estimating the intraclass correlation coefficient for binary responses in cancer prevention cluster randomized trials. *Contemp Clin Trials*. 2012;33(5):869-880. doi:10.1016/j.cct.2012.05.004

9. Hayes RJ, Moulton LH. *Cluster Randomised Trials*. CRC Press; 2017. https://books.google.fr/books?id=ikIrDwAAQBAJ

10. Kiernan K. Insights into Using the GLIMMIX Procedure to Model Categorical Outcomes with Random Effects.

11. Kenward MG, Roger JH. An improved approximation to the precision of fixed effects from restricted maximum likelihood. *Comput Stat Data Anal*. 2009;53(7):2583-2595. doi:10.1016/j.csda.2008.12.013

12. Leyrat C, Morgan KE, Leurent B, Kahan BC. Cluster randomized trials with a small number of clusters: which analyses should be used? *Int J Epidemiol*. 2018;47(1):321-331. doi:10.1093/ije/dyx169

13. McNeish D. Estimation Methods for Mixed Logistic Models with Few Clusters. *Multivar Behav Res*. 2016;51(6):790-804. doi:10.1080/00273171.2016.1236237

14. Hooper R, Forbes A, Hemming K, Takeda A, Beresford L. Analysis of cluster randomised trials with an assessment of outcome at baseline. *BMJ*. Published online March 20, 2018:k1121. doi:10.1136/bmj.k1121

15. Statistics in Medicine - 2000 - Omar - Analysis of a cluster randomized trial with binary outcome data using a multi-level.pdf.

16. Billot L, Copas A, Leyrat C, Forbes A, Turner EL. How should a cluster randomized trial be analyzed? *J Epidemiol Popul Health*. 2024;72(1):202196. doi:10.1016/j.jeph.2024.202196

17. Yan X, Lee S, Li N. Missing Data Handling Methods in Medical Device Clinical Trials. *J Biopharm Stat*. 2009;19(6):1085-1098. doi:10.1080/10543400903243009

18. Klar N, Darlington G. Methods for modelling change in cluster randomization trials. *Stat Med*. 2004;23(15):2341-2357. doi:10.1002/sim.1858

19. Schabenberger O. 196-30: Introducing the GLIMMIX Procedure for Generalized Linear Mixed Models.

20. Pedersen EJ, Miller DL, Simpson GL, Ross N. Hierarchical generalized additive models in ecology: an introduction with mgcv. *PeerJ*. 2019;7:e6876. doi:10.7717/peerj.6876

21. Atelier 8: Modèles additifs généralisés. Accessed May 13, 2022. https://r.qcbs.ca/workshop08/pres-fr/workshop08-pres-fr.html#105