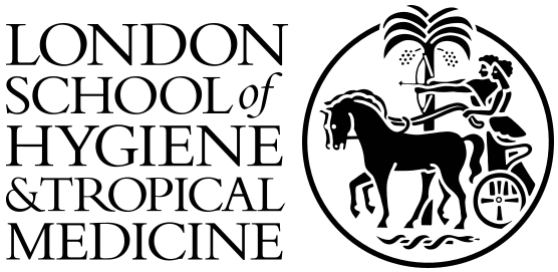

Comparison of an interactive 24-hour recall and weighed food record for measuring energy and nutrient intakes from complementary foods among 9-10 month-old Malawian infants consuming Lipid-Based Nutrient Supplements

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Comparison of an interactive 24-hour recall and weighed food record for measuring energy and nutrient intakes from complementary foods among 9-10-month-old Malawian infants consuming Lipid-Based Nutrient Supplements

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Abstract

Fortifying complementary foods with lipid-based nutrient supplements (LNS) may improve energy and nutrient intakes of infants at risk for undernutrition. We aimed to determine the relative validity of an interactive 24-hour dietary recall (i-24-HR) for assessing the impact of an LNS intervention on dietary intakes of energy and nutrients among rural Malawian 9-10-month-old infants (n=132) participating in the iLiNS dose trial. Dietary data were collected for the same day via i-24-HRs and weighed food records. Inter-method agreements were estimated overall and by intervention group, using Bland-Altman plots and paired t-tests; measurement error models (differential error); and percentage of food omissions and intrusions were estimated. Overall, inter-method differences in mean intakes of energy and most nutrients were not significant. When stratified by group, recalled energy intakes were under-estimated (-88kcal p=0.01) in the control but not in the intervention group (-10kcal; p=0.6). This differential reporting error was related to an over-estimation of recalled LNS (8.1g vs 4.5g; p<0.001) in the intervention group, compensating for an under-estimation of energy and nutrient intakes from complementary foods. Sources of measurement error in the i-24-HR were under-estimations of starchy staples, meat/fish/eggs and legumes/nuts/seeds (overall percent agreement between 38-89%; p<0.028); and over-estimations of added sugar, soups/broths and LNS (overall percent agreement between 138-149%; p<0.001). Common (>30% eating occasions) omissions were milk/fish/egg, starchy roots/vegetables, and sweetened snacks. Common intrusions were milk/yogurt. Starchy staples and LNS were recalled when consumed (>85%) (i.e. matched). These results emphasise the importance of considering differential error when interpreting dietary results in LNS trials.

Introduction

Undernutrition is common among young children living in low income countries (1). Both the short- and long-term adverse effects of under-nutrition impact health and future livelihoods. This underscores the need for comprehensive intervention packages, including effective dietary strategies. One such intervention is the use of lipid-based nutrient supplements (LNS) as home fortification of infant foods (2). Studies of the effectiveness of LNS for reducing undernutrition have shown mixed results (3-5). In cases where there was no association between LNS intake and growth outcomes (3), low adherence to the intervention (LNS consumption) and/or the displacement of other foods in the diet might partially account for the lack of a physiological effect. Thus, to correctly interpret LNS intervention trial results, accurate measurement of the LNS exposure and its influence on overall dietary intakes is fundamental.

The assessment of infant dietary intakes is complicated for several reasons: 1) infants eat very small quantities of food; 2) measuring intake includes measuring not only the amount served, but also amounts left over, spit-up, spilled or dropped; 3) infants are often cared for and fed by multiple people; and 4) infants are unable to report their own intakes (6). The weighed food record is considered the "gold standard" dietary assessment method for quantitative estimates of an individual's dietary intake, including for young children, because foods are weighed and recorded as they are consumed (7). However, for large surveys, the 24-hour recall is more practical because it is relatively rapid to conduct, has a low respondent burden and is less disruptive for low-literacy communities where, for the weighed food record, research assistants must weigh and record all foods consumed by participants. The disadvantages of 24-hour recalls are that they are prone to errors of memory, recall bias, errors in portion size reporting and potentially a social-desirability bias (8). The interactive multiple pass 24-hour recall (i-24-HR) was developed specifically for areas with low literacy rates, and includes a pictorial chart to prospectively record dietary intakes and reduce errors of memory (9).

Previous studies, in Malawi, Ghana, Sweden and the United States, have assessed the validity of the 24-hour dietary recall method relative to weighed food records (WFR) for estimating the energy and nutrient intakes of young children (10-13). They show recalled compared to weighed energy intakes are generally over-estimated (10, 12, 14), which for rural Malawian 15-m olds was by 13% (10). This pattern of over-estimation of energy intakes might be more pronounced for toddlers than infants, if accurate reporting becomes more difficult as the diet becomes more complex (12, 15). To our knowledge no study has validated the 24-hour recall for African infants under 12-months of age.

There is also evidence that certain foods are more accurately reported than others (16, 17). Such differences become important when assessing dietary exposures in a LNS intervention trial because LNS, which is an energy and nutrient dense food, is not present in the diet of the control group.

Systematic under- or over-estimation of LNS intakes would bias between-group comparisons by either exaggerating or attenuating the observed effect of LNS on infant dietary intakes, of energy and nutrients. An accurate assessment of dietary exposure is essential in dietary intervention trials to properly understand the association between dietary exposure and outcome (18-20). To our knowledge, the i-24-HR has not been validated for use among infants who are participating in an LNS intervention trial.

This study, therefore, aimed to assess the relative validity of the i-24-HR used in an LNS intervention trial, the iLiNS study (3). The iLiNS study aimed to evaluate the efficacy of three doses of LNS for the prevention of stunting among infants supplemented from 6 to 18 months of age. In this trial, inter-group differences in dietary intakes of energy and nutrients were assessed when the infants were 9-10 months of age (21). The specific objectives of the current study were to 1) assess the relative validity of the i-24-HR method for estimating dietary intakes of energy, protein, fat, iron, zinc, calcium and vitamin A from complementary foods using a 1-day WFR as the reference method; 2) assess whether there is a differential bias in i-24-HR measures of energy intake between the control group and intervention groups, and 3) describe potential sources of measurement error in the i-24-HR, including errors in the types or amounts of LNS and complementary foods reported.

Methods

Design and Study Population

A cross-sectional validation study was nested within a dietary assessment sub-study of infants participating in a 12-month LNS randomised control trial (iLiNS-DOSE trial) conducted in Mangochi district, Malawi from November 2009 and July 2012. Data collection for the dietary assessment sub-study took place between March 2010 and October 2011 when the infants were 9-10 m of age. Data collection for the dietary validation study took place between October 2010 and October 2011. The main trial was designed to assess the impact of three different doses of LNS (10g, 20g and 40g) on linear growth; which was delivered bi-weekly to households in the intervention groups. The objectives and methods of the iLiNS-DOSE trial (n=1980) and the dietary assessment sub-study (n=688) are described in more detail in Maleta, et.al. (3) and Hemsworth, et.al. (21), respectively. In the dietary assessment sub-study, two i-24-HRs were done exactly 7-days apart when the infants were between 9 and 10 months of age. One i-24-HR was done during the week LNS was delivered, and the other in the subsequent week. In the validation study the WFRs which were done one-day prior to a corresponding i-24-HR, were done just after the LNS delivery day to maximize capturing the presence of LNS in the

child's diet. The other i-24-HR was collected either 7-days before or 7-days after the i-24-HR that corresponded with the WFR day.

Sampling

A randomsample of 228 infant-mother dyads was obtained for the validation study (56 in each of the control, 10g, 20g, and 40g LNS groups). Thesample size for the validation study was calculated to allow detection of a difference of 55kcal (one 10g dose of LNS) between each of the four intervention groups with power of 80% and α=0.05, assuming a standard deviation of the difference between the methods (WFR minus i-24-HR) of 138 kcal (derived from a pilot study), and a 10% attrition rate (e.g. missed i-24-HR following the WFR).

The original inclusion criterion was participation in the dietary assessment sub-study of the iLiNS-DOSE trial. The validation study, however, began seven months after the trial began, which meant that one third of participants had already completed the dietary sub-study and were no longer eligible for the validation study. As a result, to meet our target sample size of 228 age-eligible infants, we selected additional infants (n=78) at random from the basic sub-study group (i.e., not randomised to any additional sub-study at baseline to minimise respondent burden) to reach the intended sample size. It introduced an imbalance in the number of infants from the control and 10g LNS groups versus the 20g and 40g LNS groups. As such, more infants were in the 20g LNS and 40g LNS groups than the other two groups in this validation study.

Ethical Approval

Ethical approval for this sub-study was granted by the London School of Hygiene and Tropical Medicine Research Ethics Board as well as by the College of Medicine Research Ethics Board in Malawi. Informed written consent was obtained from all participating caregivers in this study. The trial was registered at clinicaltrials.gov with the identifier: NCT00945698

Dietary Assessment

Interactive 24-hour Recall (i-24-HR)

Dietary data were collected using a 4-pass i-24-HR, developed for use in a rural African context (9). The method was modified specifically for a similar population and included pictorial charts (intended to reduce intrusions and omissions), bowls/cups/plates, and measured portion sizes using real food replicas and salted models. In the dietary assessment sub-study, caregivers were given the pictorial food chart and a plastic cup and bowl 2-days before the i-24-HR was done. On the day before the i-24-HR, caregivers were asked to prospectively record on the pictorial chart all foods, beverages, and LNS

(if appropriate) when given to the child to minimise memory errors; and to feed their child from the cup and bowl provided to minimise portion size estimation errors. In the first pass, during the i-24-HR interview, from memory, the caregiver was asked to serially recall all foods, supplements and beverages that their child had consumed in the previous 24 hours. In the second pass, information about the time, place, and description of the food or beverage was collected. In the third pass, portion sizes were estimated by the caregivers showing the amount served and the amount left-over using real food replicas (with or without excess salt to preserve them) and unit descriptions (e.g. package of biscuits). The amounts were weighed by the interviewers using digital kitchen scales (Home Elegance, accurate to ± 1g), and recorded. The amount consumed was calculated as the amount served minus the amount left-over. LNS portion sizes were measured using a pot of LNS, which was weighed before and after the caregiver had removed the amount of LNS used at each eating occasion. Left-overs were subtracted from the amount of LNS served. If LNS was mixed with other foods, the amount left over was calculated by multiplying the amount served by the proportion of the mixed dish that was consumed, assuming uniform mixing. The consumption of LNS was not specifically probed to prevent errors of intrusion (i.e. items listed but not actually consumed). To reduce potential differences in recording, interviewers were given extensive training and used standardised operating procedures, including a portion size estimation manual, detailing the specific methods for portion size estimations and probing. At the end of the third pass, interviewers asked for the pictorial chart. Any discrepancies between the pictorial chart and the food list of the i-24-HR were discussed. In the final pass, the data collector summarised and confirmed the food and drinks recorded in the i-24-HR.

Weighed Food Record (WFR)

All foods and beverages consumed by the child from 6 a.m. until the final meal of the day were weighed and recorded by a data collector, using digital kitchen scales (Home Elegance, accurate to ± 1g). Left-over foods were weighed either individually, if they could be separated on the plate, or as a mixture, assuming uniform mixing. Recipe data were collected by weighing all raw ingredients and the final cooked dish. The WFR data collector was not involved in the collection of the i-24-HR data.

Questionnaires

Socio-demographic background characteristics of the infants were collected within two weeks of baseline enrolment in the iLiNS study, when the infants were 6 months old, using an interviewer-administered questionnaire.

Data processing

Conversion factors were developed for the i-24-HR,and used to estimate the grams of food consumed. Average recipes were calculated for cooked dishes using the individual recipes collected from each household. These data were used to calculate intakes of ingredients from cooked dishes in the i-24-HRs. Intakes of energy and nutrients from the WFR and i-24-HRs were estimated, using a food composition table developed for this study (21).

The time each item was consumed was also recorded, and it was used to match the corresponding eating occasions for inter-method portion size comparisons. Meals and snacks consumed after 19:00 were removed from both the WFR and i-24-HR (i.e. a 12-hour WFR and recall were created) because there were occasions during the collection of the WFR when the final meal was consumed after the data collector had left the household.

Statistical Analysis

All data analyses were performed using Stata version 12 (StataCorp LLC, College Station, Texas). The three LNS intervention groups were collapsed to form one large group, for all analyses, because there were no significant inter-group differences in energy and nutrient intakes from complementary foods (including LNS), and the group sample sizes were small (21). In all analyses, except the analyses for an instrument effect (see below), data from only one of the two i-24-HR were used, which was the i-24-HR collected for the same day as the WFR. Energy and nutrient intake distributions from the WFR and i-24-HRs were mathematically transformed, when necessary, for the analyses.

Sociodemographic variables

A composite variable for socioeconomic status was calculated using principal component analysis (PCA), and the PCA scores were divided into quintiles using the first principal component. The following variables were used as part of the composite variable: maternal occupation, household crowding, source of electricity, source of water, sanitary facilities, material of roofing, and material of house walls.

Chi-squared tests, for categorical socio-demographic variables, and two-sample t-tests, for non-categorical socio-demographic variables, were used to check for variables associated with "missingness" of WFRs and for differences between intervention groups (control vs. LNS) in the validation study.

176 ***Assessment of agreement between dietary assessment methods***
177 Paired t-tests were used to compare mean intakes of energy and nutrients from the corresponding i-24-
178 HR and WFR. Absolute differences ("error") in amounts of energy and nutrients between the two
179 methods were calculated as follows: i-24-HR – WFR. A two-sample t-test with equal variances was
180 used to compare the absolute differences between the control and intervention groups. Bland-Altman
181 plots were used to estimate, for energy intakes, the level of agreement between the two methods and
182 the 95% limits of agreement.

183 ***Assessment of differential error***
184 Measurement error modelling was used to investigate whether error in the i-24-HR differed by
185 treatment group. We let S_1 denote the i-24-HR measurement (square-root transformed) made at the
186 same time as the WFR, and W_1 denote the WFR measurement itself (square-root transformed). The
187 second independent i-24-HR measurement (square-root transformed) was denoted S_2. The true, but
188 unobserved, intakes at time points 1 and 2 were denoted Y_1 and Y_2 respectively. At time point j ($j = 1,2$) the relationships between the observed measurements of dietary intake and the unobserved
189
190 underlying true intake were assumed to be of the following forms, where we allowed separate model
191 parameters for individuals in the control (C) and combined intervention (T) groups,
192 **Equation 1**
193

$$\text{Combined intervention group: } S_j = \gamma_{0T} + \gamma_{1T} Y_j + \epsilon_{Tj}$$
$$\text{Control group: } S_j = \gamma_{0C} + \gamma_{1C} Y_j + \epsilon_{Cj}$$

194

$$\text{Combined intervention group: } W_1 = Y_j + \delta_{Tj}$$
$$\text{Control group: } W_1 = Y_j + \delta_{Cj}$$

The ϵ and δ terms are random errors with mean zero and constant variance. The WFR is assumed to provide an unbiased estimate of true intake in both the control and intervention groups. The intercept parameters γ_{0T} and γ_{0C} , and slope parameters γ_{1T} and γ_{1C}, represent systematic error in the i-24-HR measurement. We assessed evidence for differential error based on estimates ofthe differences $\gamma_{1T} - \gamma_{1C}$ and $\gamma_{0T} - \gamma_{0C}$ and corresponding bootstrap confidence intervals. The parameters of the measurement error model in Equation 1 were estimated via a method of moments approach.

Sources of disagreement between the i-24-HR and WFR

To identify possible sources of disagreement between the two dietary assessment methods, we categorised each food and drink item (for composite dishes, we matched the individual ingredients) as an omission (present on WFR, absent on i-24-HR), an intrusion (absent on WFR, present on i-24-HR) or a match (present on both methods at matching meal/snack times). We calculated the frequency of each category across food groups (i.e., phala; nsima and rice; added sugar; sweetened snacks; savoury snacks; meat, fish and egg; legumes, nuts, and seeds; fruit; starchy roots and vegetables; milk and yogurt; non-dairy beverages; soup/broth from relish; and LNS), a method previously described by Smith, et.al. (22). We compared the median percentage agreement for each food group, (i.e. 100* reported amount (i-24-HR) / reference amount (WFR)), for the intervention and control groups, using Mann-Whitney rank sum test when the sample was at least five consumers. In the case where one food within a food group of these is an intrusion, this resulted in a reference amount of zero (at the individual food level only), and in the case where there is an omission, this resulted in a reported amount of zero. We also compared the overall inter-method differences, in the grams of food consumed in each food group, using the Wilcoxon signed-rank test.

Instrument Effect

We tested for an "instrument effect", because the presence of a data collector on the day of the WFR might have influenced the caregivers' ability to recall dietary intakes during its corresponding i-24-HR. This "instrument effect" was assessed using the Wilcoxon signed-rank test, by comparing the median intakes of energy and nutrients estimated using the i-24-HR corresponding to the WFR day and the i-24-HR collected on a day independent of the WFR (i.e., collected one week before or after the WFR). For this analysis, n=71 matched records were available.

Results

Participants

A total of 228 infants were selected to participate in the validation study. However, 78 were lost to follow-up and 18 did not have a matching WFR and i-24-HR. The final sample size analysed was 132 matching i-24HRs and WFRs (**Figure 1**). There were no significant differences in socio-demographic characteristics comparing those with missing data and those who completed the WFR (data not shown). Likewise, there were no differences in baseline characteristics between the intervention and control group (**Table 1)**.

Agreement between dietary assessment methods

The reported energy intakes were lower in the i-24-HR compared to the WFR, although the difference was not statistically significant (p=0.09) (**Table 2**). Reported protein intake was significantly underestimated and calcium intake was significantly over-estimated by the i-24-HR compared to the WFR (p<0.001). There were no significant between-method differences in intakes of fat, iron, zinc or vitamin A. The Bland-Altman plot showed a systematic bias for under-reporting recalled energy intakes compared to the WFR and poor agreement at the individual level, with 95% limits of agreement of -366 kcal to 316 kcal (Online supplement **Figure 1**).

When stratified by intervention group, however, there was a significant under-estimation of recalled energy intakes in the control group (p=0.010) but not in the intervention group (p=0.60) (**Table 2**). Recalled intakes of protein, fat, iron and zinc were also significantly underestimated in the control group. In the intervention group, recalled intakes of protein were significantly under-estimated, whereas recalled intakes of calcium and zinc were significantly overestimated (Table 2). Further, after comparing the absolute differences ("error") calculated between the WFR and i-24-HR in the control and intervention groups, we found significant differences ($p \leq 0.05$) for energy (kcal) and iron, and all other nutrients were considered non-significant (p>0.05). The Bland-Altman plot by intervention group (Online supplement **Figures 2a and 2b**) showed poor 95% limits of agreement (LOA) for energy at an individual level, for both the intervention (95% LOA -358, 337 kcal) and control (95% LOA -375 to 207 kcal) groups; and a mean systematic under-estimation of energy intakes in the control group only (-84 kcal)).

By fitting the measurement error models in equation 1, we found that $\hat{\gamma}_{1C} = -2.4$ (95% CI (-24.9, 29.7)) and $\hat{\gamma}_{1T} = 2.6$ (95% CI (-20.0, 20.2)), $\hat{\gamma}_{0C} = 63.2$ (95% CI (58.8, 67.3)) and $\hat{\gamma}_{0T} = -32.5$ (95% CI (-34.5,-30.6)). The confidence intervals were obtained from the 2.5 and 97.5 percentiles of 1000

bootstrap estimates, using bootstrap samples stratified by intervention group. The expected i-24-HR measure of energy intake (S) given the true intake (Y) is therefore $E(S|Y) = -32.5 + 2.6Y$ in the combined intervention group, and $E(S|Y) = 63.2 - 2.4Y$ in the control group. The estimates of the slope are in opposite directions in the intervention and control groups because the correlation between the independent i-24 and the WFR is positive in the intervention group, but negative in the control group; however the CIs are very wide and the 95% bootstrap CI for the difference $\gamma_{1T} - \gamma_{1C}$ was (-46.6, 56.5). However, there was strong evidence for a difference in the intercepts; the 95% bootstrap CI for the difference $\gamma_{0T} - \gamma_{0C}$ was (-100.1, -90.7) The model-based approach, therefore, suggests that the relationship between the i-24-HR measure of energy intake and the true intake may be different in the intervention and groups, i.e. potential differential error.

Sources of disagreement between thei-24-HR and WFR

LNS intakes

In the intervention group, there was a significant between-method difference in estimated LNS intakes. The median intake was significantly higher for the recalled (i-24-HR) than reference (WFR) amount (i.e., 8.1g (4.5, 11.8) vs 4.5g (2.0, 9.0); p<0.001) (**Online Supplement Table 1**). The median (IQR) percentage agreement (matched LNS portions) indicates recalled LNS consumption was over-estimated by over 50% compared to the WFR (**Table 3**). Close to 90% of the eating occasions matched on both the WFR and i-24-HR; and rates of intrusions and omissions were similar and low (**Table 4**).

Complementary food intakes

At the pooled group level, phala, legumes, nuts and seeds, and meat, fish and eggs were significantly under-estimated; whereas, soups/broths from relish and added sugar were significantly over-estimated in the i-24-HR compared to the WFR (**Online Supplement Table 1**). There were no significant differences between intervention- and control groups in reporting accuracy (i.e., percentage agreement for food groups), except for soups/broths from relish, where the control group showed a higher over-reporting rate than the intervention group. These comparisons, for four of the 12 food groups, were limited by the small sample size of the control group (Table 3).

In both the intervention and control groups, a comparison of food group matches, intrusions and omissions showed the highest reporting agreement for staples, where over 88% of the phala and nsima eating occasions matched between the two methods (Table 4). Episodically consumed foods such as meat, fish and eggs (which were frequently misreported as soup/broth from relish), starchy roots and vegetables, and sweetened snacks had poor reporting matching, with a higher tendency for respondents to omit (i.e. forget) as opposed to intrude (i.e. add in error).

The "instrument-effect"

There was no evidence of an "instrument effect". There were no significant differences in estimated intakes of energy or nutrients comparing the independent i-24-HR (performed either one week before or after the WFR) and the corresponding i-24-HR (i.e., for the same day as the WFR). The absolute differences ranged from zero RAE/d to 34 kcal/d (**Online supplement Table 2**).

Discussion

In the context of a LNS supplementation trial, we found there was no significant difference comparing energy intakes measured using the i-24-HR to the WFR when all groups were pooled. This comparison was not biased towards agreement by the weighing process, because the independent and corresponding i-24-HRs provided similar estimates of energy and nutrients intakes. However, this pooled comparison masked a difference between the intervention and control group. When stratified by intervention group, the i-24-HR systematically under-estimated dietary energy intakes compared with the WFR in the control group but not in the intervention group. The significant difference in the "error" or absolute difference between the methods in control and intervention groups suggest a differential for recalled energy intakes. This differential error, for estimating median energy intakes, primarily is the result of an over-estimation of the energy-dense supplement (LNS), which was only consumed by the intervention group. It compensated for the under-estimation of energy intakes from complementary foods because most caregivers were able to report whether their infant had consumed it. In contrast, when using dietary data collected via i-24-HRs to examine associations, the 95% LOA indicate poor agreement at the individual level, in both groups, which will attenuate associations. These results highlight, when aiming to estimate inter-group differences in median intakes of energy and nutrients in an intervention trial, the importance of examining whether systematic measurement error when quantifying intervention food consumption, contributes to a differential bias. In studies aiming to examine associations between dietary intakes and functional outcomes (e.g., growth), the i-24-HR is inferior to more accurate methods of dietary assessment. In our study considerable effort was made to accurately estimate LNS consumption. The caregivers were asked to spoon out the amount of LNS served to the infant and estimate the amount left-over, which were both weighed and recorded.

There were few differences, comparing the intervention and control group, for between-method agreement in the estimation of complementary foods intakes. In the pooled group analyses, the main sources of between-method disagreement were under-estimated recalled portion sizes of dietary staples (phala, rice and nsima by between 11 and 14%), meat, fish and eggs and legumes, nuts and seeds. Energy-dense foods, such as added sugar, were overestimated by over 40% compared with the WFR;

but it did not compensate for the under-estimation of energy from staples (phala, nsima and rice). This result is not surprising because dietary staples provide a high percentage of daily energy intakes for rural infants in Malawi.

Underestimation of certain food groups is not unique and has been reported among women in Malawi (9) as well as preschool aged children in Ghana (11). However, the underestimation in energy intakes relative to the WFR, in the control group of our study, is in contrast to results from a study of 10-13 month old Senegalese infants (n=45), which showed the 24-hour recall was a relatively good measure of intake compared to WFR (23, 24); and a study of 15-month old rural Malawian infants (n=169), which showed a systematic over-estimation in energy and nutrient intakes (10). The sources of measurement error, in the previous Malawian study, are unknown. These inter-study differences could be a function of inter-method or age group differences. In our study, we probed for left-overs and adjusted the portion sizes in the i-24-HR based on recalled left-overs. This adjustment was not reported in the other studies. It has been suggested that as a diet becomes more complex (as the infant ages), the reporting accuracy changes (12) and perhaps the direction of the error also changes.

The results of this validation study suggest that a differential error might be present when an i-24-HR is used to measure group mean dietary intakes, which is related to a systematic over-estimation of the exposure (LNS). Linear calibration techniques could be used to correct the systematic under-estimation of energy intakes from non-LNS foods. Previous studies have developed correction factors using the WFR as the reference standard to adjust i-24-HR energy intakes for a systematic overestimation of energy intakes compared to the WFR. This technique is not recommended for the current study because the reference method is subject to the same errors as the test method (19, 25), e.g. both the WFR and i-24-HR are subject to mis-estimation of items that were spilled or spit up. The linear calibration equations would only have been appropriate if we had used a biomarker, such as the stable isotope technique to measure total energy expenditure, which is an unbiased and independent measure of long-term energy intake (6, 20).

Study Limitations and Advantages

The main study limitations were the relatively low sample size and high rate of attrition. The study was underpowered to detect differential error in the i-24-HR between control vs. intervention groups. The high rate of attrition occurred because of the logistical demands of this validation study in a large catchment area (i.e. transportation, communication with households, etc.). No observed background characteristics were associated with missing the visit.

Another limitation was the reference method used. The WFR is the most common reference standard for comparison with a 24-hour dietary recall because it is less resource-intensive than collection of biomarkers, and it provides useful robust information about portion size estimation, intrusions and omissions. However, it does not meet the strict criteria for a valid reference method (26). To validate the i-24-HR (repeated to provide an estimate of usual intakes), for estimating energy intakes alone, the doubly labelled water method is the preferred reference method (25, 27). Further, the modelling approach we used to assess evidence for differential error (equation 1), relies on an assumption that the WFR provides an unbiased measure of intake, as well as additional assumptions about the form of the systematic errors.

This study also had many advantages. It was carried out severalmonths after the start of the intervention, which meant that the children were habituated to the intervention food. It was also conducted over a long period of time which allowed for seasonal variation in dietary patterns and episodically consumed foods to be captured. This study is also the first study that we are aware of that has assessed the relative validity of the i-24-HR for estimating the dietary intakes of rural African infants under 12 months of age who are participating in an LNS intervention trial. Such trials are important because the process of stunting predominantly occurs before 15 months of age in rural Africa (28). Detailed and accurate dietary intake information will contribute to an improved understanding of direct causes of stunting and undernutrition. The study results emphasise the importance of considering a potential differential bias to avoid the misinterpretation of intervention results.

Conclusions

At the pooled group level, the i-24-HR showed relatively good agreement to the WFR. However, there was an apparent differential bias whereby the mean intakes of energy and some nutrients were under-estimated compared with the WFR in the control group but not in the intervention group. Considering the cost and logistical implications of the WFR, the i-24-HR could be used in its place, for estimating mean intakes, but careful attention should be made during the design stage to the objectives of the study and whether only measures of absolute intakes or overall between-group differences are required. Absolute intakes might be under-estimated, if the i-24-HR is used to estimate dietary energy intakes of 9-10-month-old infants who are not consuming an energy dense supplement, such as LNS. Future interventions evaluating differential dietary exposures (such as LNS) should consider, when comparing groups, whether a systematic error in intervention food measurement introduced a differential bias. When designing the study, they should put effort into developing an accurate method of quantifying intervention food consumption; and where possible, evaluate it in a pilot study before commencing data

collection. For researchers aiming to examine associations between dietary intakes and functional outcomes, such as growth, if resources permit, they should include a dietary assessment validation study, with a biomarker reference method (or using a gold-standard reference method) to understand the dietary assessment method's measurement error structure to help avoid misinterpretation of dietary intakes in relation to final growth outcomes.

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Author contributions

J.H, C.K., K.M., U.A., M.A., & E.L.F designed the research and significantly contributed to the aim and structure of manuscript; J.H. & C.K. conducted the research; A.M.R. & R.K. provided statistical guidance and assistance with methods; J.H, R.K. & E.L.F analysed data and performed statistical analyses; J.H drafted the paper with inputs from R.K. & E.L.F; J.H., R.K. & E.L.F had primary responsibility for the final content. R.K. & E.L.F have equal contribution to senior authorship. All authors have read and approved the final manuscript.

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Table 1 Characteristics of participants at enrolment into the main study (at 6 months of age)

	Control	Intervention	p-value
Participants (n)	26	106	
Female n (%)	14 (54)	49 (47)	0.50[a]
Socio-demographic Background Characteristics (n)	**24**	**105**	
Maternal age; mean (SD) years	28.8 (7.3)	26.6 (5.9)	0.12[b]
Maternal Education; mean (SD) years	3.9 (3.4)	4.4 (3.6)	0.52[b]
Female-headed household n (%)	2 (8.3)	12 (11.9)	0.78[a]
More than one child under 5 years old in household n (%)	11 (45.8)	44 (41.9)	0.06[a]
Maternal occupation n (%)			0.64[a]
Farming/Fishing	17 (77.3)	66 (66.0)	
House wife	3 (16.6)	27 (27.0)	
Indoor / office work	1 (4.6)	3 (3.0)	
Other	1 (4.6)	3 (3.0)	
Unknown	0 (0)	1 (1)	
Information collected during time of visit (n)	**26**	**106**	
Season (rainy: October - March) n (%)	12 (46.1)	56 (52.8)	0.80[a]
Infant Breastfeeding n (%)	25 (100)[c]	104 (98.1)	0.49[a]

a Chi-square
b Two-sample t-test
c n=25 breastfed, n=1 missing value in this control group

Table 2: Estimated intakes of energy and selected nutrients (Mean and 95 % Confidence Interval)[a] using the i-24-HR compared to WFR between the hours of 06:00 and 18:00 by intervention group and pooled group

	Control Group (n=26)				Intervention Group- LNS (n=106)					Pooled Group (n=132)			
Nutrient	WFR	i-24-HR Recall	Abs. Diff[b]	p-value[c]	WFR	i-24-HR Recall	Abs Diff[b]	p-value[c]	p-value[d]	WFR	i-24-HR Recall	Abs Diff[b]	p-value[c]
Energy (kcal/d)	376 (317, 437)	293 (246, 345)	-88	0.010	388 (352, 424)	379 (346, 412)	-10	0.60	0.052	385 (355, 416)	361 (333, 390)	-25	0.09
Protein (g/d)	9.6 (7.7, 11.6)	7.1 (5.8, 8.4)	-2.9	0.009	9.4 (8.4, 10.5)	8.2 (7.3, 9.0)	-1.6	0.007	0.36	9.5 (8.5, 10.4)	8.0 (7.3, 8.6)	-1.8	<0.001
Fat (g/d)	7.3 (5.3, 9.8)	5.3 (4.0, 6.8)	-2.8	0.05	10.0 (8.7, 11.5)	10.4 (9.1, 11.7)	0.1	0.62	0.10	9.6 (8.3, 10.7)	9.2 (8.2, 10.4)	-0.4	0.65
Iron (mg/d)	2.6 (2.1, 3.2)	1.8 (1.4, 2.2)	-0.1	<0.001	3.7 (3.3, 4.2)	4.0 (3.4, 4.5)	0.3	0.25	0.020	3.5 (3.1, 3.9)	3.5 (3.0, 3.9)	0.03	0.68
Zinc (mg/d)	1.6 (1.2, 1.9)	1.1 (0.9, 1.4)	-0.5	<0.001	3.3 (2.8, 3.8)	3.8 (3.1, 4.4)	0.6	0.020	0.07	2.9 (2.5, 3.3)	3.1 (2.6, 3.7)	0.4	0.18
Calcium (mg/d)	38 (25, 54)	53 (33, 77)	21.6	0.20	94 (77, 113)	128 (107, 152)	38.3	<0.001	0.41	81 (68, 96)	111 (93, 130)	35.1	<0.001
Vitamin A (µg RAE/d)	39 (18, 67)	24 (9, 46)	-18.8	0.19	143 (113, 176)	164 (130, 202)	24.1	0.10	0.23	117 (93, 144)	125 (99, 156)	15.9	0.37

[a] Data back-transformed from square root transformation for presentation
[b] Absolute mean difference - i-24HR Recall – WFR
[c] Matched pairs T-test
[d] Two-group t-test with equal variances between intervention and control group absolute differences

i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, RAE: retinol activity equivalents, WFR: weighed food record

Table 3: Percentage agreement for matching foods (items appearing both on the i-24-HR and the WFR) between intervention groups

	Median (25th, 75th percentile)				
	Control Group (n=25)		Intervention Group (n=106)		
	n[a,e]	Percentage Agreement[b]	n	Percentage Agreement[b]	p-value[c]
Phala, all types (full volume)	25	100.0 (78.5, 122.4)	99	87.5 (68.1, 118.6)	0.457
Nsima, Rice (full volume)	25	78.4 (61.7, 100.0)	98	95.4 (59.5, 141.5)	0.248
Added Sugar	14	141.5 (103.7, 250.0)	69	167.7 (111.2, 295.0)	0.776
Sweetened Snacks	5	61.4 (50.7, 166.0)	45	112.7 (61.1, 195.0)	0.258
Savoury Snacks	8	105.9 (84.6, 137.5)	18	100.0 (56.7, 175.0)	0.683
Meat, Fish and Egg (solid)	7	82.7 (62.9, 294.9)	26	107.8 (62.7, 151.9)	0.735
Legumes, Nuts, Seeds	8	36.1 (26.4, 76.6)	26	76.2 (37.5, 105.3)	0.680
Fruit	4	160.0 (88.1, 231.7)	27	94.0 (66.2, 140.0)	--
Starchy Root and Vegetables	2	29.2 (22.1, 36.3)	20	80.8 (48.2, 145)	--
Milk and Yogurt	3	90.2 (90.0, 103.7)	8	111.0 (53.0, 228.6)	--
Non-dairy beverages	5	115.3 (85.6, 173.7)	15	100.0 (66.8, 142.2)	--
Soup/Broth from Relish	14	239.0 (195.3, 308.3)	54	134.0 (85.7, 240.0)	0.038
LNS	-		65	154.0 (98.8, 298.3)[d]	--

[a] Includes all portion sizes from items that match between the reported and reference values at the same time (i.e.: meal or snack time)

[b] Report percentage = (Reported amount / reference amount) x 100

Reference amount observed during the weighed food record; Reported amount taken from the 24-hour dietary recall.

[c] Mann-Whitney two-sample rank sum test by food group

[d] LNS only present in the diets of the intervention group, which is why there is no between-group comparison. This is descriptive only, looking at the percentage agreement of LNS in the intervention group.

[e] One participant missing in the control group for these analyses

i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, WFR: weighed food record

Table 4: Number of eating episodes and percentages of matching food groups (items appearing both in the i-24-HR and the WFR), intrusions and omissions by intervention groups

	Control Group (n=25[d])			Intervention Group (n=106)		
	n (%)			n (%)		
	matching [a]	intrusion [b]	omission [c]	matching [a]	intrusion [b]	omission [c]
Phala, all types (full volume)	49 (92.5)	0 (0)	4 (7.6)	166 (94.3)	2 (1.1)	8 (4.6)
Nsima, Rice (full volume)	30 (88.2)	3 (8.8)	1 (2.9)	150 (89.8)	9 (5.4)	8 (4.8)
Added Sugar	22 (73.3)	5 (16.7)	3 (6.7)	105 (68.6)	26 (17.0)	22 (14.4)
Sweetened Snacks	6 (50.0)	2 (16.7)	4 (33.3)	59 (68.6)	15 (17.4)	12 (14.0)
Savoury Snacks	10 (76.9)	2 (15.6)	1 (7.7)	23 (69.7)	5 (15.2)	5 (15.2)
Meat, Fish and Egg (solid)	8 (53.3)	0 (0)	7 (46.7)	34 (56.7)	7 (11.7)	20 (32.8)
Legumes, Nuts, Seeds	13 (76.5)	1 (5.9)	3 (17.6)	39 (68.4)	4 (7.0)	14 (24.6)
Fruit	4 (66.7)	1 (16.7)	1 (16.7)	34 (70.8)	8 (16.7)	6 (12.5)
Starchy Root and Vegetables	2 (40.0)	0 (0)	3 (60.0)	22 (71.0)	4 (12.9)	5 (16.1)
Milk and Yogurt	3 (100)	0(0)	0 (0)	8 (47.1)	6 (35.3)	3 (17.6)
Non-dairy beverages	6 (75.0)	2 (25.0)	0 (0)	20 (62.5)	7 (21.9)	5 (15.6)
Soup/Broth from Relish	18 (62.1)	8 (27.6)	3 (10.3)	68 (64.7)	30 (28.6)	7 (6.7)
LNS	-	--		101 (89.4)	7 (6.2)	5 (4.4)

[a] The total of portions that were matched between the reference (WFR) and reported (i-24-HR), as a percentage of all items in the same group

[b] The total of portions that were reported (i-24-HR) but not observed in the reference data (WFR)

[c] The total of portions that were observed in the reference data (WFR), but not reported (i-24-HR)

[d] One participant missing for these analyses

i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, WFR: weighed food record

Online supplement Table 1: Average reported (i-24-HR) and reference (WFR) portion sizes by food group

	Median (25th, 75th Percentiles)				
	n [a]	**Reported amount (g)[b]**	**Reference Amount (g)[c]**	**Percentage agreement[d]**	**P-value[e]**
Phala, all types (full volume)	125	78.9 (48.5, 112.0)	99.0 (64.7, 136.0)	86.4 (66.1, 114.1)	<0.001
Nsima, Rice (full volume)	124	52.5 (29.1, 80.0)	56.8 (33.5, 89.8)	89.1 (56.6, 135.0)	0.028
Added Sugar	94	5.1 (3.6, 7.9)	3.0 (1.9, 5.5)	143.3 (99.2, 238.9)	<0.001
Sweetened Snacks	64	7.9 (4.1, 15.8)	9.0 (4.0, 15.5)	91.7 (38.0, 158.0)	0.64
Savoury Snacks	34	7.7 (3.5, 11.0)	6.0 (3.0, 10.0)	86.1 (51.9, 157.1)	0.59
Meat, Fish and Egg (solid)	57	6.0 (0, 12.4)	9.2 (4.9, 18.2)	59.7 (0, 110.7)	0.015
Legumes, Nuts, Seeds	50	2.4 (0.4, 5.8)	7.8 (3.9, 16.0)	37.5 (2.4, 83.8)	<0.001
Fruit	38	22.5 (10.0, 35.0)	17.0 (6.0, 32.5)	94.0 (52.0, 136.4)	0.64
Starchy Root and Vegetables	30	18.0 (7.0, 24.0)	15.5 (6.0, 43.0)	50.0 (19.4, 120.0)	0.12
Milk and Yogurt	15	11.8 (5.2, 41.0)	8.0 (1.0, 29.0)	90.1 (36.8, 183.2)	0.82
Non-dairy beverages	33	47.3 (27.5, 76.1)	27.7 (9.0, 86.3)	98.1 (43.8, 123.5)	0.28
Soup/Broth from Relish	94	17.0 (11.7, 26.0)	7.4 (0, 16.9)	138.5 (80.0, 243.1)	<0.001
LNS	68	8.1 (4.5, 11.8)	4.5 (2.0, 9.0)	148.7 (95.0, 274.0)	<0.001

[a] Refers to the number of participants where this food group was present on the WFR, i-24-HR, or both. This includes the average portion size estimation per food group per participant. In the case where one was an intrusion, this resulted in a reference value of zero, and in the case where there is an omission, this resulted in a reported amount of zero. This is the participant average per food group.

[b] median daily average per participant of reported amount derived from i-24-HR

[c] median daily average per participant of reference amount derived from WFR

[d] Percentage agreement: (Reported amount / reference amount) x 100

[e] p-value derived from Wilcoxon signed-rank test for matched pairs

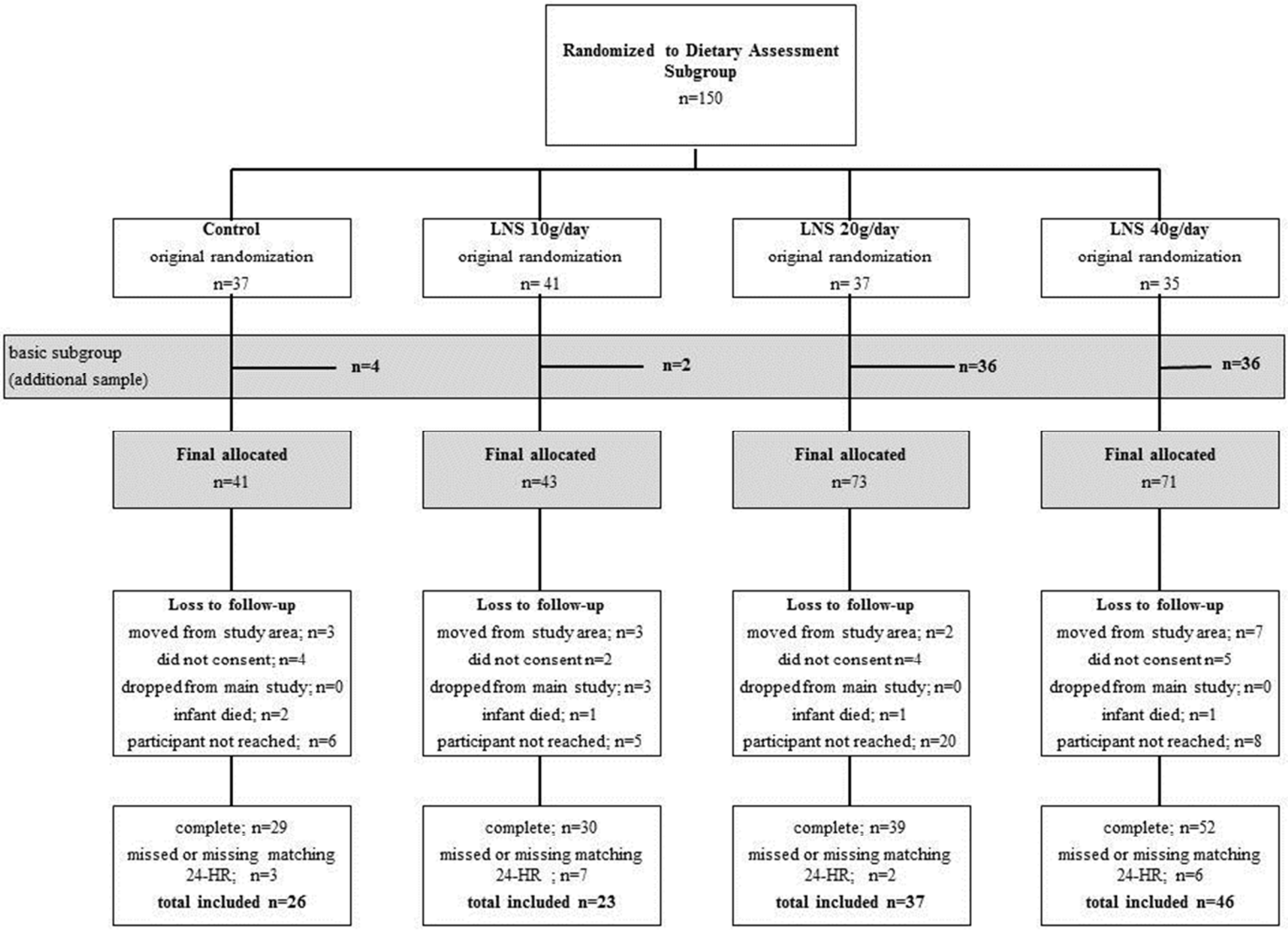
i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, WFR: weighed food record

Online supplement Table 2: Comparison of i-24-HRs that corresponded to and were independent of the WFR. An Assessment of bias in reporting related to the presence of the WFR: the "instrument effect".

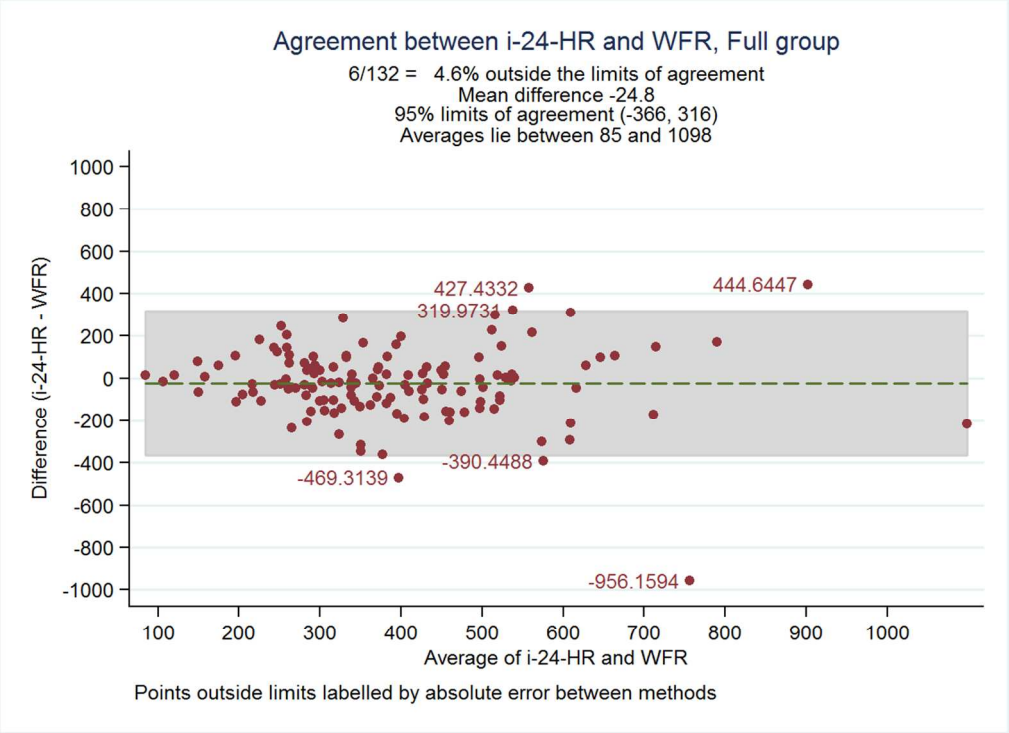
	N=71 Median Intake (25th,75th percentile)			
Nutrient	**Independent 24-HR Recall**	**i24-HR WFR**	**Absolute Difference[a]**	**p-value[b]**
Energy (kcal/d)	375 (273, 553)	327 (246, 463)	-34	0.10
Protein (g/d)	8.8 (5.8, 12.5)	7.6 (5.0, 10.3)	-0.78	0.06
Fat (g/d)	9.8 (5.0, 15.4)	8.1 (4.2, 11.8)	-1.9	0.06
Fe (mg/d)	3.2 (1.9, 5.8)	2.6 (1.7, 5.3)	-0.2	0.50
Zn (mg/d)	2.2 (1.2, 5.9)	2.0 (1.2, 6.1)	-0.1	0.97
Ca (mg/d)	115.9 (41.5, 204.3)	104.9 (34.7, 208.5)	-1.1	0.48
Vitamin A (µg RAE/d)	122.9 (30.3, 262.9)	107.9 (20.5, 292.9)	0	0.79

[a] i-24HR WFR – Independent 24-HR
[b] Wilcoxon signed rank matched-pairs test

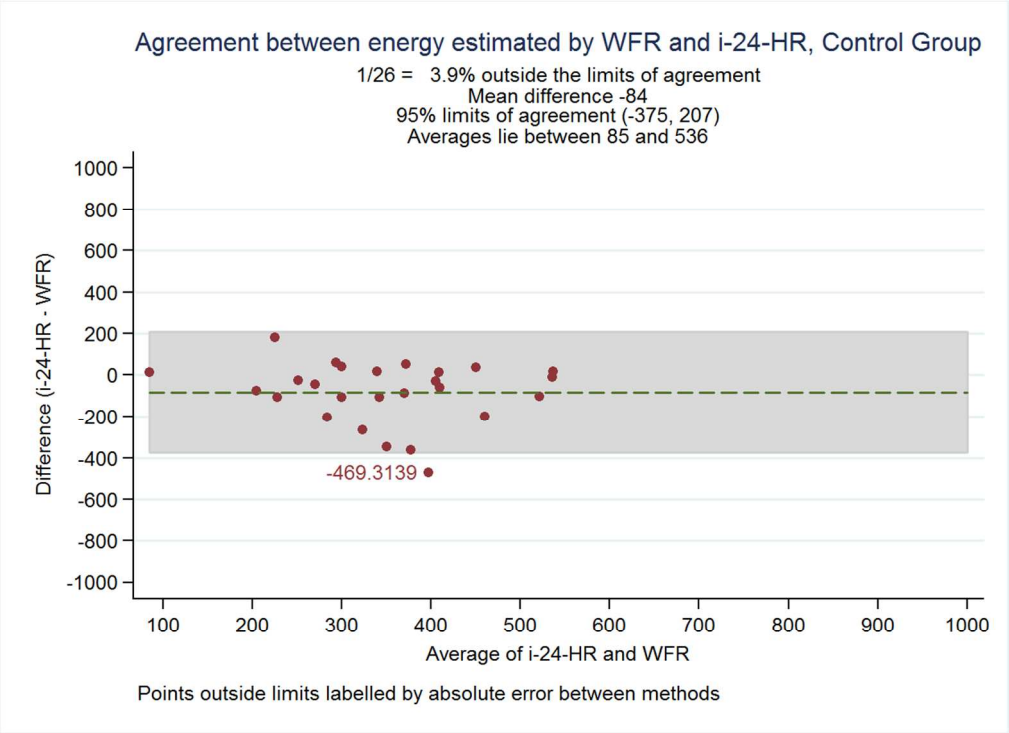
Figure 1: Consort Flow Diagram of Participant Enrolment and Inclusion in the Validation Sub-Study



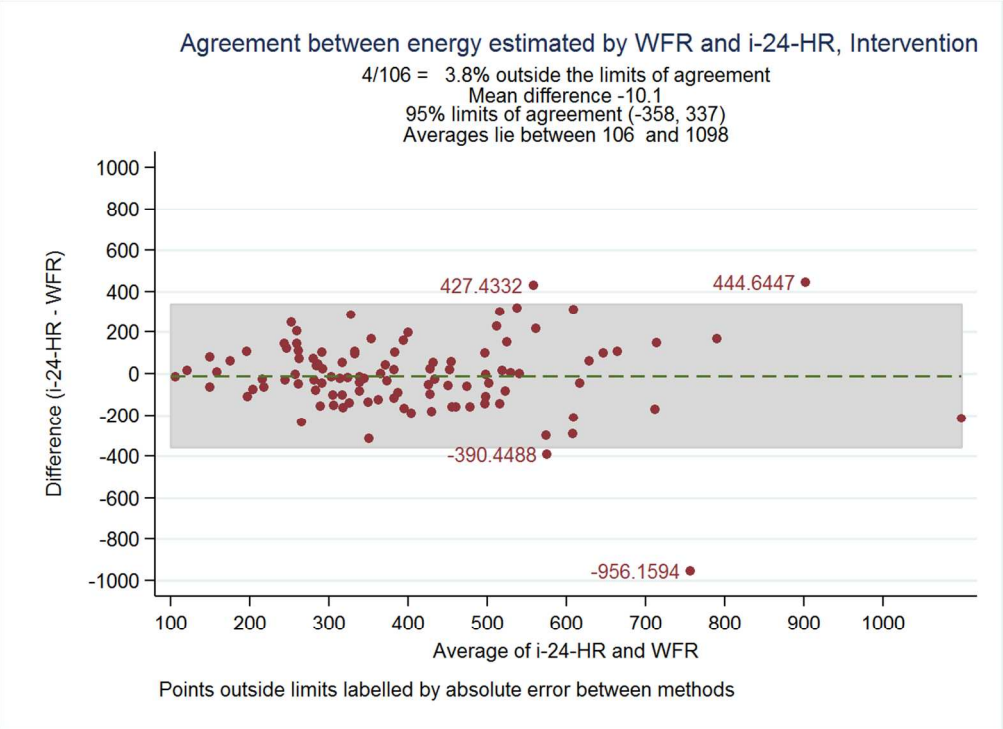
Online supplement Figure 1: Bland Altman Plot Showing Relative Agreement in energy (kcal/day) estimation between WFR and i-24-HR: Pooled Group



Online Figure 2a: Bland Altman Plot Showing Relative Agreement in Energy (kcal) estimation between WFR and i-24-HR: Control Group



Online supplement Figure 2b: Bland Altman Plot Showing Relative Agreement in Energy (kcal) estimation between WFR and i-24-HR: Intervention Group



Comparison of an interactive 24-hour recall and weighed food record for measuring energy and nutrient intakes from complementary foods among 9-10-month-old Malawian infants consuming Lipid-Based Nutrient Supplements

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Abstract

Fortifying complementary foods with lipid-based nutrient supplements (LNS) may improve energy and nutrient intakes of infants at risk for undernutrition. We aimed to determine the relative validity of an interactive 24-hour dietary recall (i-24-HR) for assessing the impact of an LNS intervention on dietary intakes of energy and nutrients among rural Malawian 9-10-month-old infants (n=132) participating in the iLiNS dose trial. Dietary data were collected for the same day via i-24-HRs and weighed food records. Inter-method agreements were estimated overall and by intervention group, using Bland-Altman plots and paired t-tests; measurement error models (differential error); and percentage of food omissions and intrusions were estimated. Overall, inter-method differences in mean intakes of energy and most nutrients were not significant. When stratified by group, recalled energy intakes were under-estimated (-88kcal p=0.01) in the control but not in the intervention group (-10kcal; p=0.6). This differential reporting error was related to an over-estimation of recalled LNS (8.1g vs 4.5g; p<0.001) in the intervention group, compensating for an under-estimation of energy and nutrients intakes from complementary foods. Sources of measurement error in the i-24-HR were under-estimations of starchy staples, meat/fish/eggs and legumes/nuts/seeds (overall percent agreement ~~overall report rates~~ between~~ranged from~~ 38-89%; p<0.028); and over-estimations of added sugar, soups/broths and LNS (overall percent agreement between~~overall report rates ranged from~~ 138-149%; p<0.001). Common (>30% ~~of~~ eating occasions) omissions were milk/fish/egg, starchy roots/vegetables, and sweetened snacks. Common intrusions were milk/yogurt. ~~Common (>20% eating occasions) omissions were meat/fish/eggs, legumes/nuts/seeds and starchy roots/vegetables, and intrusions were milk/ yogurt, beverages and soup/broths.~~ Starchy staples and LNS were recalled when consumed (>85%) (i.e. ~~well~~ matched). These results emphasise the importance of considering differential error when interpreting dietary results in LNS trials.

Introduction

Undernutrition is common among young children living in low income countries (1). Both the short- and long-term adverse effects of under-nutrition impact health and future livelihoods. This underscores the need for comprehensive intervention packages, including effective dietary strategies. One such intervention is the use of lipid-based nutrient supplements (LNS) as home fortification of infant foods (2). Studies of the effectiveness of LNS for reducing undernutrition have shown mixed results (3-5). In cases where there was no association between LNS intake and growth outcomes (3), low adherence to the intervention (LNS consumption) and/or the displacement of other foods in the diet might partially account for the lack of a physiological effect. Thus, to correctly interpret LNS intervention trial results, accurate measurement of the LNS exposure and its influence on overall dietary intakes is fundamental.

The assessment of infant dietary intakes is complicated for several reasons: 1) infants eat very small quantities of food; 2) measuring intake includes measuring not only the amount served, but also amounts left over, spit-up, spilled or dropped; 3) infants are often cared for and fed by multiple people; and 4) infants are unable to report their own intakes (6). The weighed food record is considered the "gold standard" dietary assessment method for quantitative estimates of an individual's dietary intake, including for young children, because foods are weighed and recorded as they are consumed (7). However, for large surveys, the 24-hour recall is more practical because it is relatively rapid to conduct, has a low respondent burden and is less disruptive for low-literacy communities where, for the weighed food record, research assistants must weigh and record all foods consumed by participants. The disadvantages of 24-hour recalls are that they are prone to errors of memory, recall bias, errors in portion size reporting and potentially a social-desirability bias (8). The interactive multiple pass 24-hour recall (i-24-HR) was developed specifically for areas with low literacy rates, and includes a pictorial chart to prospectively record dietary intakes and reduce errors of memory (9).

Previous studies, in Malawi, Ghana, Sweden and the United States, have assessed the validity of the 24-hour dietary recall method relative to weighed food records (WFR) for estimating the energy and nutrient intakes of young children (10-13). They show recalled compared to weighed energy intakes are generally over-estimated (10, 12, 14), which for rural Malawian 15-m olds was by 13% (10). This pattern of over-estimation of energy intakes might be more pronounced for toddlers than infants, if accurate reporting becomes more difficult as the diet becomes more complex (12, 15). To our knowledge no study has validated the 24-hour recall for African infants under 12-months of age.

There is also evidence that certain foods are more accurately reported than others (16, 17). Such differences become important when assessing dietary exposures in a LNS intervention trial because LNS, which is an energy and nutrient dense food, is not present in the diet of the control group.

Systematic under- or over-estimation of LNS intakes would bias between-group comparisons by either exaggerating or attenuating the observed effect of LNS on infant dietary intakes, of energy and nutrients. An accurate assessment of dietary exposure is essential in dietary intervention trials to properly understand the association between dietary exposure and outcome (18-20). To our knowledge, the i-24-HR has not been validated for use among infants who are participating in an LNS intervention trial.

This study, therefore, aimed to assess the relative validity of the i-24-HR used in an LNS intervention trial, the iLiNS study (3). The iLiNS study aimed to evaluate the efficacy of three doses of LNS for the prevention of stunting among infants supplemented from 6 to 18 months of age. In this trial, inter-group differences in dietary intakes of energy and nutrients were assessed when the infants were 9-10 months of age (21). The specific objectives of the current study were to 1) assess the relative validity of the i-24-HR method for estimating dietary intakes of energy, protein, fat, iron, zinc, calcium and vitamin A from complementary foods using a 1-day WFR as the reference method; 2) assess whether there is a differential bias in i-24-HR measures of energy intake between the control group and intervention groups, and 3) describe potential sources of measurement error in the i-24-HR, including errors in the types or amounts of LNS and complementary foods reported.

Methods

Design and Study Population

A cross-sectional validation study was nested within a dietary assessment sub-study of infants participating in a 12-month LNS randomised control trial (iLiNS-DOSE trial) conducted in Mangochi district, Malawi from November 20090~~10~~ and July 2012. Data collection for the dietary assessment sub-study took place between March 2010 and October 2011~~xxx~~ when the infants were 9-10 m of age. Data collection ~~Data collection~~ for the dietary validation study took place between October 2010 and October 2011. The main trial was designed to assess the impact of three different doses of LNS (10g, 20g and 40g) on linear growth; which was delivered bi-weekly to households in the intervention groups. The objectives and methods of the iLiNS-DOSE trial (n=1980) and the dietary assessment sub-study (n=688) are described in more detail in Maleta, et.al. (3) and Hemsworth, et.al. (21), respectively. In the dietary assessment sub-study, two i-24-HRs were done exactly 7-days apart when the infants were between 9 and 10 months of age. One i-24-HR was done during the week -LNS was delivered, and the other in the subsequent week. In the validation study the WFRs which were done one-day prior to a corresponding i-24-HR, were done just after the LNS delivery day to maximize

capturing the presence of LNS in the child's diet. The other i-24-HR was collected either 7-days before or 7-days after the i-24-HR that corresponded with the WFR day.

Sampling

A ~~stratified random~~ random ~~-~~sample of 228 infant-mother dyads was obtained ~~calculatedselected~~ for the validation study (~~i.e.,~~ 56 in each of the control, 10g, 20g, and 40g LNS groups). The~~is~~ sample size for the validation study was ~~chosen~~ calculated to allow detection of a difference of 55kcal (one 10g dose of LNS) between each of the four intervention groups with power of 80% and α=0.05, assuming a standard deviation of the difference between the methods (WFR minus i-24-HR) of 138 kcal (derived from a pilot study), and a 10% attrition rate (e.g. missed i-24-HR following the WFR).

The original inclusion criterion was participation in the dietary assessment sub-study of the iLiNS-DOSE trial. The validation study, however, began seven months after the trial began, which meant that one third of participants had already completed the dietary sub-study and were no longer eligible for the validation study. As a result, to meet our target sample size of 228 age-eligible infants, we selected additional infants (n=78) at random from the basic sub-study group (i.e., not randomised to any additional sub-study at baseline to minimise respondent burden) to reach the intended sample size. It introduced an imbalance in the number of infants from the control and 10g LNS groups versus the 20g and 40g LNS groups. As such, more infants were in the 20g LNS and 40g LNS groups than the other two groups in this validation study.

Ethical Approval

Ethical approval for this sub-study ~~study~~ was granted by the London School of Hygiene and Tropical Medicine Research Ethics Board as well as by the College of Medicine Research Ethics Board in Malawi. Informed written consent was obtained from all participating caregivers in this study. The trial was registered at clinicaltrials.gov with the identifier: NCT00945698

Dietary Assessment

Interactive 24-hour Recall (i-24-HR)

Dietary data were collected using a 4-pass i-24-HR, developed for use in a rural African context (9). The method was modified specifically for a similar population ~~to~~ and included pictorial charts (intended to reduce intrusions and omissions), bowls/cups/plates, and measured portion sizes using real food replicas and salted models. In the dietary assessment sub-study, caregivers were given the pictorial food chart and a plastic cup and bowl 2-days before the i-24-HR was done. On the day before the i-24-HR, caregivers~~they~~ were asked to prospectively record on the pictorial chart all foods,

beverages, and LNS (if appropriate) when given to the child to minimise memory errors; and to feed their child from the cup and bowl provided to minimise portion size estimation errors. In the first pass, during the i-24-HR interview, from memory, the caregiver was asked to serially recall all foods, supplements and beverages that their child had consumed in the previous 24 hours. In the second pass, information about the time, place, and description of the food or beverage was collected. In the third pass, portion sizes were estimated by the caregivers~~respondent~~s showing the amount served and the amount left-over using real food replicas (with or without excess salt to preserve them) and unit descriptions (e.g. package of biscuits). The amounts were weighed by the interviewers using digital kitchen scales (Home Elegance, accurate to ± 1g), and recorded. The amount consumed was calculated as the amount served minus the amount left-over. LNS portion sizes were measured using a pot of LNS, which was weighed before and after the caregiver had removed the amount of LNS used at each eating occasion. Left-overs were subtracted from the amount of LNS served. If LNS was mixed with other foods, the amount left over was calculated by multiplying the amount served by the proportion of the mixed dish that was consumed, assuming uniform mixing. The consumption of LNS was not specifically probed to prevent errors of intrusion (i.e. items listed but not actually consumed). To reduce potential differences in recording, interviewers were given extensive training and used standardised operating procedures, including a portion size estimation manual, detailing the specific methods for portion size estimations and probing. At the end of the third pass, interviewers ~~data collectors~~ asked for the pictorial chart. Any discrepancies between the pictorial chart and the food list of the i-24-HR were discussed. In the final pass, the data collector summarised and confirmed the food and drinks recorded in the i-24-HR.

Weighed Food Record (WFR)

All foods and beverages consumed by the child from 6 a.m. until the final meal of the day were weighed and recorded by a data collector, using digital kitchen scales (Home Elegance, accurate to ± 1g). Left-over foods were weighed either individually, if they could be separated on the plate, or as a mixture, assuming uniform mixing. Recipe data were collected by weighing all raw ingredients and the final cooked dish. The WFR data collector was not involved in the collection of the i-24-HR data.

Questionnaires

Socio-demographic background characteristics of the infants were collected within two weeks of baseline enrolment in the iLiNS study, when the infants were 6 months old, using an interviewer-administered questionnaire. ~~analysed(maternal occupation, maternal education level, household size, head of household, and presence of other child under 5 years in the household) of the infants were~~

~~collected using an interviewer administered questionnaire within two weeks of baseline enrolment (when infants were 6 months of age).~~

Data processing

Conversion factors were developed for the i-24-HR, and used to estimate the grams of food consumed. Average recipes were calculated for cooked dishes using the individual recipes collected from each household. These data were used to calculate intakes of ingredients from cooked dishes in the i-24-HRs. Intakes of energy and nutrients from the WFR and i-24-HRs were estimated, using a food composition table developed for this study (21).

The time each item was consumed was also recorded, and it was used to match the corresponding eating occasions for inter-method portion size comparisons. Meals and snacks consumed after 19:00 were removed from both the WFR and i-24-HR (i.e. a 12-hour WFR and recall were created) because there were occasions during the collection of the WFR when the final meal was consumed after the data collector had left the household.

Statistical Analysis

All data analyses were performed using Stata version 12 (StataCorp LLC, College Station, Texas). The three LNS intervention groups were collapsed to form one large group, for all analyses, because there were no significant inter-group differences in energy and nutrient intakes from complementary foods (including LNS), and the group sample sizes were small (21). In all analyses, except the analyses for an instrument effect (see below), data from only one of the two i-24-HR were used, which was the i-24-HR collected for the same day as the WFR. Energy and nutrient intake distributions from the WFR and i-24-HRs were mathematically transformed, when necessary, for the analyses.

Sociodemographic variables

A composite variable for socioeconomic status was calculated using principal component analysis (PCA), and the PCA scores were divided into quintiles using the first principal component. The following variables were used as part of the composite variable: maternal occupation, household crowding, source of electricity, source of water, sanitary facilities, material of roofing, and material of house walls.

Chi-squared tests, for categorical socio-demographic variables, and two-sample t-tests, for non-categorical socio-demographic variables, were used to check for variables associated with "missingness" of WFRs and for differences between intervention groups (control vs. LNS) in the validation study.

Assessment of agreement between dietary assessment methods

Paired t-tests were used to compare mean intakes of energy and nutrients from the corresponding i-24-HR and WFR. Absolute differences ("error") in amounts of energy and nutrients between the two methods were calculated as follows: i-24-HR – WFR. A two-sample t-test with equal variances was used to compare the absolute differences between the control and intervention groups. Bland-Altman plots were used to estimate, for energy intakes, the level of agreement between the two methods and the 95% limits of agreement.

Assessment of differential error

Measurement error modelling was used to investigate whether error in the i-24-HR differed by treatment group. We let S_1 denote the i-24-HR measurement (square-root transformed) made at the same time as the WFR, and W_1 denote the WFR measurement itself (square-root transformed). The second independent i-24-HR measurement (square-root transformed) was denoted ~~the square-root transformed measure~~ S_2. The true, but unobserved, intakes at time points 1 and 2 were denoted Y_1 and Y_2 respectively. At time point j ($j = 1,2$) the relationships between the observed measurements of dietary intake and the unobserved underlying true intake were assumed to be of the following forms, where we allowed separate model parameters for individuals in the control (C) and combined intervention (T) groups,

Equation 1

$$\text{Combined intervention group: } S_j = \gamma_{0T} + \gamma_{1T} Y_j + \epsilon_{Tj}$$

$$\text{Control group: } S_j = \gamma_{0C} + \gamma_{1C} Y_j + \epsilon_{Cj}$$

$$\text{Combined intervention group: } W_1 = Y_j + \delta_{Tj}$$

$$\text{Control group: } W_1 = Y_j + \delta_{Cj}$$

The ϵ and δ terms are random errors with mean zero and constant variance. The WFR is assumed to provide an unbiased estimate of true intake in both the control and intervention groups. The intercept parameters γ_{0T} and γ_{0C} , and slope parameters γ_{1T} and γ_{1C}, represent systematic error in the i-24-HR measurement. We assessed evidence for differential error based on estimates of~~bootstrap confidence intervals for~~ the differences $\gamma_{1T} - \gamma_{1C}$ and $\gamma_{0T} - \gamma_{0C}$ and corresponding bootstrap confidence intervals. The parameters of the measurement error model in Equation 1 were estimated via a method of moments approach.

Sources of disagreement between the i-24-HR and WFR

To identify possible sources of disagreement between the two dietary assessment methods, we categorised each food and drink item (for composite dishes, we matched the individual ingredients) as an omission (present on WFR, absent on i-24-HR), an intrusion (absent on WFR, present on i-24-HR) or a match (present on both methods at matching meal/snack times). We calculated the frequency of each category across food groups (i.e., phala; nsima and rice; added sugar; sweetened snacks; savoury snacks; meat, fish and egg; legumes, nuts, and seeds; fruit; starchy roots and vegetables; milk and yogurt; non-dairy beverages; soup/broth from relish; and LNS), a method previously described by Smith, et.al. (22). We compared the median percentage agreement for each food group, (i.e. 100* reported amount (i-24-HR) / reference amount (WFR)), for the intervention and control groups, using Mann-Whitney rank sum test when the sample was at least five consumers. In the case where one food within a food group of these is an intrusion, this resulted in a reference amount of zero (at the individual food level only), and in the case where there is an omission, this resulted in a reported amount of zero. We also compared the overall inter-method differences, in the grams of food consumed in each food group, using the Wilcoxon signed-rank test.

Instrument Effect

We tested for an "instrument effect", because the presence of a data collector on the day of the WFR might have influenced the caregivers~~respondent~~'s ability to recall dietary intakes during its corresponding i-24-HR. This "instrument effect" was assessed using the Wilcoxon signed-rank test, by comparing the median intakes of energy and nutrients estimated using the i-24-HR corresponding to the WFR day and the i-24-HR collected on a day independent of the WFR (i.e., collected one week before or after the WFR). For this analysis, n=71 matched records were available.

Results

Participants

A total of 228 infants were selected to participate in the validation study. However, 78 were lost to follow-up and 18 did not have a matching WFR and i-24-HR. The final sample size analysed was 132 matching i-24HRs and WFRs (**Figure 1**). There were no significant differences in socio-demographic characteristics comparing those with missing data and those who completed the WFR (data not shown). Likewise, there were no differences in baseline characteristics between the intervention and control group (**Table 1)**.

Agreement between dietary assessment methods

The reported energy intakes were lower in the i-24-HR compared to the WFR, although the difference was not statistically significant (p=0.09) (**Table 2**). Reported protein intake was significantly underestimated and calcium intake was significantly over-estimated by the i-24-HR compared to the WFR (p<0.001). There were no significant between-method differences in intakes of fat, iron, zinc or vitamin A. The Bland-Altman plot showed a systematic bias for under-reporting recalled energy intakes compared to the WFR and poor agreement at the individual level, with 95% limits of agreement of -3668 kcal to 3167 kcal (Online supplement **Figure 1**).

When stratified by intervention group, however, there was a significant under-estimation of recalled energy intakes in the control group (p=0.010) but not in the intervention group (p=0.60) (**Table 2**). Recalled intakes of protein, fat, iron and zinc were also significantly underestimated in the control group. In the intervention group, recalled intakes of protein were significantly under-estimated, whereas recalled intakes of calcium and zinc were significantly overestimated (Table 2). Further, after comparing the absolute differences ("error") calculated between the WFR and i-24-HR in the control and intervention groups, we found significant differences ($p \leq 0.05$) for energy (kcal) and iron, and all other nutrients were considered non-significant ($p > 0.05$). The Bland-Altman plot by intervention group (Online supplement **Figures 2a and 2b**) showed poor 95% limits of agreement (LOA) for energy at an individual level, for both the intervention (95% LOA -358, 337 kcal) and control (95% LOA -375 to 207 kcal) groups; and a mean systematic under-estimation of energy intakes in the control group only (-84 kcal~~, 95% LOA -375 to 207 kcal)~~~~84 kcal~~).

By fitting the measurement error models in equation 1, we found that $\hat{\gamma}_{1C} = -2.4$ (95% CI (-24.9, 29.7)) and $\hat{\gamma}_{1T} = 2.6$ (95% CI (-20.0, 20.2)), $\hat{\gamma}_{0C} = 63.2$ (95% CI (58.8, 67.3)) and $\hat{\gamma}_{0T} = -32.5$ (95% CI (-34.5,-30.6)). The confidence intervals were obtained from the 2.5 and 97.5 percentiles of 1000

bootstrap estimates, using bootstrap samples stratified by intervention group. The expected i-24-HR measure of energy intake (S) given the true intake (Y) is therefore $E(S|Y) = -32.5 + 2.6Y$ in the combined intervention group, and $E(S|Y) = 63.2 - 2.4Y$ in the control group. The estimates of the slope are in opposite directions in the intervention and control groups because the correlation between the independent i-24 and the WFR is positive~~,~~ in the intervention group, but negative in the control group; however the CIs are very wide and the 95% bootstrap CI for the difference $\gamma_{1T} - \gamma_{1C}$ was (-46.6, 56.5). However, there was strong evidence for a difference in the intercepts; the 95% bootstrap CI for the difference $\gamma_{0T} - \gamma_{0C}$ was (-100.1, -90.7) The model-based approach, therefore, ~~provides~~ suggests that the relationship between the i-24-HR measure of energy intake and the true intake may be different in the intervention and groups, i.e. ~~indication of a~~indicates a potential ~~evidence of~~ differential error.

Sources of disagreement between the~~measurement error in the~~ i-24-HR and WFR

LNS intakes

In the intervention group, there was a significant between-method difference in estimated LNS intakes. The median intake was significantly higher for the recalled (i-24-HR) than reference (WFR) amount (i.e., 8.1g (4.5, 11.8) vs 4.5g (2.0, 9.0); p<0.001) **(Online Supplement Table 1)**. The median (IQR) percentage agreement (matched LNS portions) indicates recalled LNS consumption was over-estimated by over 50% compared to the WFR (**Table 3**). Close to 90% of the eating occasions matched on both the WFR and i-24-HR; and rates of intrusions and omissions were similar and low (**Table 4**).

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Complementary food intakes

At the pooled group level, phala, legumes, nuts and seeds, and meat, fish and eggs were significantly under-estimated; whereas, soups/broths from relish and added sugar were significantly over-estimated in the i-24-HR compared to the WFR (**Online Supplement Table 1**). There were no significant differences between intervention- and control groups in reporting accuracy (i.e., percentage agreement for food groups), except for soups/broths from relish, where the control group showed a higher over-reporting rate than the intervention group. These comparisons, for four of the 12 food groups, were limited by the small sample size of the control group (Table 3).

In both the intervention and control groups, a comparison of food group matches, intrusions and omissions showed the highest reporting agreement for staples, where over 88% of the phala and nsima eating occasions matched between the two methods (Table 4). Episodically consumed foods such as meat, fish and eggs (which were frequently misreported as soup/broth from relish), starchy roots and

vegetables, and sweetened snacks had poor reporting matching, with a higher tendency for respondents to omit (i.e. forget) as opposed to intrude (i.e. add in error).

The "instrument-effect"

There was no evidence of an "instrument effect". There were no significant differences in estimated intakes of energy or nutrients comparing the independent i-24-HR (performed either one week before or after the WFR) and the corresponding i-24-HR (i.e., for the same day as the WFR). The absolute differences ranged from zero RAE/d to 34 kcal/d (**Online supplement Table 2**).

Discussion

In the context of a LNS supplementation trial, we found there was no significant difference comparing energy intakes measured using the i-24-HR to the WFR when all groups were pooled. This comparison was not biased towards agreement by the weighing process, because the independent and corresponding i-24-HRs provided similar estimates of energy and nutrients intakes. However, this pooled comparison masked a difference between the intervention and control group. When stratified by intervention group, the i-24-HR systematically under-estimated dietary energy intakes compared with the WFR in the control group but not in the intervention group. The significant difference in the "error" or absolute difference between the methods in control and intervention groups suggest a differential for recalled energy intakes. This differential error, for estimating median energy intakes, primarily is the result of an over-estimation of the energy-dense supplement (LNS), which was only consumed by the intervention group. It compensated for the under-estimation of energy intakes from complementary foods because most caregivers~~respondents~~ were able to report whether their infant had consumed it. In contrast, when using dietary data collected via i-24-HRs to examine associations, the 95% LOA indicate poor agreement at the individual level, in both groups, which will attenuate associations. These results highlight ~~the importance~~, when aiming to ~~of estimating differential measurement error to correctly interpret~~ estimate inter-group differences in ~~the impact of an energy- and nutrient-dense supplement on~~median intakes of energy and nutrients ~~dietary intakes (and growth outcomes)~~ in an intervention trial, the importance of examining whether systematic measurement error when quantifying intervention food consumption, contributes to a differential bias. In studies aiming to examine associations between dietary intakes and functional outcomes (e.g., growth), the i-24-HR is inferior to more accurate methods of dietary assessment. In our study considerable effort was made to accurately estimate LNS consumption. The ~~caregivers~~~~respondents~~caregivers were asked to spoon out the amount of LNS served to the infant and estimate the amount left-over, which were both weighed and recorded.

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There were few differences, comparing the intervention and control group, for between-method agreement in the estimation of complementary foods intakes. In the pooled group analyses, the main sources of between-method disagreement were under-estimated recalled portion sizes of dietary staples (phala, rice and nsima by between 11 and 14%), meat, fish and eggs and legumes, nuts and seeds. Energy-dense foods, such as added sugar, were overestimated by over 40% compared with the WFR; but it did not compensate for the under-estimation of energy from staples (phala, nsima and rice). This result is not surprising because dietary staples provide a high percentage of daily energy intakes for rural infants in Malawi.

Underestimation of certain food groups is not unique and has been reported among women in Malawi (9) as well as preschool aged children in Ghana (11). However, the underestimation in energy intakes relative to the WFR, in the control group of our study, is in contrast to results from a study of 10-13 month old Senegalese infants (n=45), which showed the 24-hour recall was a relatively good measure of intake compared to WFR (23, 24); and a study of 15-month old rural Malawian infants (n=169), which showed a systematic over-estimation in energy and nutrient intakes (10). The sources of measurement error, in the previous Malawian study, ~~is~~ are unknown. These inter-study differences could be a function of inter-method or age group differences. In our study, we probed for left-overs and adjusted the portion sizes in the i-24-HR based on recalled left-overs. This adjustment was not reported in the other studies. It has been suggested that as a diet becomes more complex (as the infant ages), the reporting accuracy changes (12) and perhaps the direction of the error also changes.

The results of this validation study suggest that a differential error might be present when an i-24-HR is used to measure group mean~~edian~~ dietary intakes, which is related to a systematic over-estimation of the exposure (LNS). Linear calibration techniques could be used to correct the systematic under-estimation of energy intakes from non-LNS foods. Previous studies have developed correction factors using the WFR as the reference standard to adjust i-24-HR energy intakes for a systematic overestimation of energy intakes compared to the WFR. This technique is not recommended for the current study because the reference method is subject to the same errors as the test method (19, 25), e.g. both the WFR and i-24-HR are subject to mis-estimation of items that were spilled or spit up. The linear calibration equations would only have been appropriate if we had used a biomarker, such as the stable isotope technique to measure total energy expenditure, which is an unbiased and independent measure of long-term energy intake (6, 20).

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Study Limitations and Advantages

The main study limitations were the relatively low sample size and high rate of attrition. The study was underpowered to detect differential error in the i-24-HR between control vs. intervention groups. The high rate of attrition occurred because of the logistical demands of this validation study in a large catchment area (i.e. transportation, communication with households, etc.). No observed background characteristics were associated with missing the visit.

Another limitation was the reference method used. The WFR is the most common reference standard for comparison with ~~the~~ a 24-hour dietary recall because it is less resource-intensive than collection of biomarkers, and it provides useful robust information about portion size estimation, intrusions and omissions. However, it does not meet the strict criteria for a valid reference method (26). To validate the i-24-HR (repeated to provide an estimate of usual intakes), for estimating energy intakes alone, the doubly labelled water method is the preferred reference method (25, 27). Further, the modelling approach we used to assess evidence for differential error (equation 1), relies on an assumption that the WFR provides an unbiased measure of intake, as well as additional assumptions about the form of the systematic errors.

This study also had many advantages. It was carried out several~~3~~ months after the start of the intervention, which meant that the children were habituated to the intervention food. It was also conducted over a long period of time which allowed for seasonal variation in dietary patterns and episodically consumed foods to be captured. This study is also the first study that we are aware of that has assessed the relative validity of the i-24-HR for estimating the dietary intakes of rural African infants under 12 months of age who are participating in an LNS intervention trial. Such trials are important because the process of stunting predominantly occurs before 15 months of age in rural Africa (28). Detailed and accurate dietary intake information will contribute to an improved understanding of direct causes of stunting and undernutrition. The study results emphasise the importance of considering a potential differential bias to avoid the misinterpretation of intervention results.

Conclusions

At the pooled group level, the i-24-HR showed relatively good agreement to the WFR. However, there was an apparent differential bias whereby the mean~~dian~~ intakes of energy and some nutrients were under-estimated compared with the WFR in the control group but not in the intervention group. Considering the cost and logistical implications of the WFR, the i-24-HR could be used in its place, for estimating mean~~dian~~ intakes, but careful attention should be made during the design stage to the objectives of the study and whether only measures of absolute intakes or overall between-group

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differences are required. Absolute intakes might be under-estimated, if the i-24-HR is used to estimate dietary energy intakes of 9-10-month-old infants who are not consuming an energy dense supplement, such as LNS. Future interventions evaluating differential dietary exposures (such as LNS) should consider, when comparing groups, whether a systematic error in intervention food measurement introduced a differential bias. When designing the study, they should put effort into developing an accurate method of quantifying intervention food consumption; and where possible, evaluate it in a pilot study before commencing data collection. For researchers aiming to examine associations between dietary intakes and functional outcomes, such as growth, if resources permit, they should include a dietary assessment validation study, ~~preferably~~ with a biomarker reference method (or using a gold-standard reference method) to understand the dietary assessment method's measurement error structure ~~and~~ to help avoid misinterpretation of dietary intakes in relation to final growth outcomes.

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Author contributions

J.H, C.K., K.M., U.A., M.A., & E.L.F designed the research and significantly contributed to the aim and structure of manuscript; J.H. & C.K. conducted the research; A.M.R. & R.K. provided statistical guidance and assistance with methods; J.H, R.K. & E.L.F analysed data and performed statistical analyses; J.H drafted the paper with inputs from R.K. & E.L.F; J.H., R.K. & E.L.F had primary responsibility for the final content. R.K. & E.L.F have equal contribution to senior authorship. All authors have read and approved the final manuscript.

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Table 1 Characteristics of participants at enrolment into the main study (at 6 months of age)

	Control	Intervention	p-value
Participants (n)	26	106	
Female n (%)	14 (54)	49 (47)	0.50[a]
Socio-demographic Background Characteristics (n)	**24**	**105**	
Maternal age; mean (SD) years	28.8 (7.3)	26.6 (5.9)	0.12[b]
Maternal Education; mean (SD) years	3.9 (3.4)	4.4 (3.6)	0.52[b]
Female-headed household n (%)	2 (8.3)	12 (11.9)	0.78[a]
More than one child under 5 years old in household n (%)	11 (45.8)	44 (41.9)	0.06[a]
Maternal occupation n (%)			0.64[a]
Farming/Fishing	17 (77.3)	66 (66.0)	
House wife	3 (16.6)	27 (27.0)	
Indoor / office work	1 (4.6)	3 (3.0)	
Other	1 (4.6)	3 (3.0)	
Unknown	0 (0)	1 (1)	
Information collected during time of visit (n)	**26**	**106**	
Season (rainy: October - March) n (%)	12 (46.1)	56 (52.8)	0.80[a]
Infant Breastfeeding n (%)	25 (100)[c]	104 (98.1)	0.49[a]

a Chi-square
b Two-sample t-test
c n=25 breastfed, n=1 missing value in this control group

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Table 2: Estimated intakes of energy and selected nutrients (Mean and 95 % Confidence Interval)[a] using the i-24-HR compared to WFR between the hours of 06:00 and 18:00 by intervention group and pooled group

	Control Group (n=26)				Intervention Group- LNS (n=106)					Pooled Group (n=132)			
Nutrient	WFR	i-24-HR Recall	Abs. Diff[b]	p-value[c]	WFR	i-24-HR Recall	Abs Diff[b]	p-value[c]	p-value[d]	WFR	i-24-HR Recall	Abs Diff[b]	p-value[c]
Energy (kcal/d)	376 (317, 437)	293 (246, 345)	-88	0.010	388 (352, 424)	379 (346, 412)	-10	0.60	0.052	385 (355, 416)	361 (333, 390)	-25	0.09
Protein (g/d)	9.6 (7.7, 11.6)	7.1 (5.8, 8.4)	-2.9	0.009	9.4 (8.4, 10.5)	8.2 (7.3, 9.0)	-1.6	0.007	0.36	9.5 (8.5, 10.4)	8.0 (7.3, 8.6)	-1.8	<0.001
Fat (g/d)	7.3 (5.3, 9.8)	5.3 (4.0, 6.8)	-2.8	0.05	10.0 (8.7, 11.5)	10.4 (9.1, 11.7)	0.1	0.62	0.10	9.6 (8.3, 10.7)	9.2 (8.2, 10.4)	-0.4	0.65
Iron (mg/d)	2.6 (2.1, 3.2)	1.8 (1.4, 2.2)	-0.1	<0.001	3.7 (3.3, 4.2)	4.0 (3.4, 4.5)	0.3	0.25	0.020	3.5 (3.1, 3.9)	3.5 (3.0, 3.9)	0.03	0.68
Zinc (mg/d)	1.6 (1.2, 1.9)	1.1 (0.9, 1.4)	-0.5	<0.001	3.3 (2.8, 3.8)	3.8 (3.1, 4.4)	0.6	0.020	0.07	2.9 (2.5, 3.3)	3.1 (2.6, 3.7)	0.4	0.18
Calcium (mg/d)	38 (25, 54)	53 (33, 77)	21.6	0.20	94 (77, 113)	128 (107, 152)	38.3	<0.001	0.41	81 (68, 96)	111 (93, 130)	35.1	<0.001
Vitamin A (µg RAE/d)	39 (18, 67)	24 (9, 46)	-18.8	0.19	143 (113, 176)	164 (130, 202)	24.1	0.10	0.23	117 (93, 144)	125 (99, 156)	15.9	0.37

[a] Data back-transformed from square root transformation for presentation
[b] Absolute mean difference - i-24HR Recall – WFR
[c] Matched pairs T-test
[d] Two-group t-test with equal variances between intervention and control group absolute differences

i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, RAE: retinol activity equivalents, WFR: weighed food record

Table 3: Percentage agreement for matching foods (items appearing both on the i-24-HR and the WFR) between intervention groups

	Median (25th, 75th percentile)				
	Control Group (n=25)		Intervention Group (n=106)		
	n[a,e]	Percentage Agreement[b]	n	Percentage Agreement[b]	p-value[c]
Phala, all types (full volume)	25	100.0 (78.5, 122.4)	99	87.5 (68.1, 118.6)	0.457
Nsima, Rice (full volume)	25	78.4 (61.7, 100.0)	98	95.4 (59.5, 141.5)	0.248
Added Sugar	14	141.5 (103.7, 250.0)	69	167.7 (111.2, 295.0)	0.776
Sweetened Snacks	5	61.4 (50.7, 166.0)	45	112.7 (61.1, 195.0)	0.258
Savoury Snacks	8	105.9 (84.6, 137.5)	18	100.0 (56.7, 175.0)	0.683
Meat, Fish and Egg (solid)	7	82.7 (62.9, 294.9)	26	107.8 (62.7, 151.9)	0.735
Legumes, Nuts, Seeds	8	36.1 (26.4, 76.6)	26	76.2 (37.5, 105.3)	0.680
Fruit	4	160.0 (88.1, 231.7)	27	94.0 (66.2, 140.0)	--
Starchy Root and Vegetables	2	29.2 (22.1, 36.3)	20	80.8 (48.2, 145)	--
Milk and Yogurt	3	90.2 (90.0, 103.7)	8	111.0 (53.0, 228.6)	--
Non-dairy beverages	5	115.3 (85.6, 173.7)	15	100.0 (66.8, 142.2)	--
Soup/Broth from Relish	14	239.0 (195.3, 308.3)	54	134.0 (85.7, 240.0)	0.038
LNS	-		65	154.0 (98.8, 298.3)[d]	--

[a] Includes all portion sizes from items that match between the reported and reference values at the same time (i.e.: meal or snack time)

[b] Report percentage = (Reported amount / reference amount) x 100

Reference amount observed during the weighed food record; Reported amount taken from the 24-hour dietary recall.

[c] Mann-Whitney two-sample rank sum test by food group

[d] LNS only present in the diets of the intervention group, which is why there is no between-group comparison. This is descriptive only, looking at the percentage agreement of LNS in the intervention group.

[e] One participant missing in the control group for these analyses

i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, WFR: weighed food record

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Table 4: Number of eating episodes and percentages of matching food groups (items appearing both in the i-24-HR and the WFR), intrusions and omissions -by intervention groups

	Control Group (n=25[d])			Intervention Group (n=106)		
	n (%)			n (%)		
	matching [a]	intrusion [b]	omission [c]	matching ~~[a] matching~~	intrusion ~~[b] intrusion~~	omission ~~[c] omission~~
Phala, all types (full volume)	49 (92.5)	0 (0)	4 (7.6)	166 (94.3)	2 (1.1)	8 (4.6)
Nsima, Rice (full volume)	30 (88.2)	3 (8.8)	1 (2.9)	150 (89.8)	9 (5.4)	8 (4.8)
Added Sugar	22 (73.3)	5 (16.7)	3 (6.7)	105 (68.6)	26 (17.0)	22 (14.4)
Sweetened Snacks	6 (50.0)	2 (16.7)	4 (33.3)	59 (68.6)	15 (17.4)	12 (14.0)
Savoury Snacks	10 (76.9)	2 (15.6)	1 (7.7)	23 (69.7)	5 (15.2)	5 (15.2)
Meat, Fish and Egg (solid)	8 (53.3)	0 (0)	7 (46.7)	34 (56.7)	7 (11.7)	20 (32.8)
Legumes, Nuts, Seeds	13 (76.5)	1 (5.9)	3 (17.6)	39 (68.4)	4 (7.0)	14 (24.6)
Fruit	4 (66.7)	1 (16.7)	1 (16.7)	34 (70.8)	8 (16.7)	6 (12.5)
Starchy Root and Vegetables	2 (40.0)	0 (0)	3 (60.0)	22 (71.0)	4 (12.9)	5 (16.1)
Milk and Yogurt	3 (100)	0(0)	0 (0)	8 (47.1)	6 (35.3)	3 (17.6)
Non-dairy beverages	6 (75.0)	2 (25.0)	0 (0)	20 (62.5)	7 (21.9)	5 (15.6)
Soup/Broth from Relish	18 (62.1)	8 (27.6)	3 (10.3)	68 (64.7)	30 (28.6)	7 (6.7)
LNS	-	--		101 (89.4)	7 (6.2)	5 (4.4)

[a] The total of portions that were matched between the reference (WFR) and reported (i-24-HR), as a percentage of all items in the same group

[b] The total of portions that were reported (i-24-HR) but not observed in the reference data (WFR)

[c] The total of portions that were observed in the reference data (WFR), but not reported (i-24-HR)

[d] One participant missing for these analyses

i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, WFR: weighed food record

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Online supplement Table 1: Average reported (i-24-HR) and reference (WFR) portion sizes by food group

	Median (25th, 75th Percentiles)				
	n [a]	Reported amount (g)[b]	Reference Amount (g)[c]	Percentage agreement[d]	P-value[e]
Phala, all types (full volume)	125	78.9 (48.5, 112.0)	99.0 (64.7, 136.0)	86.4 (66.1, 114.1)	<0.001
Nsima, Rice (full volume)	124	52.5 (29.1, 80.0)	56.8 (33.5, 89.8)	89.1 (56.6, 135.0)	0.028
Added Sugar	94	5.1 (3.6, 7.9)	3.0 (1.9, 5.5)	143.3 (99.2, 238.9)	<0.001
Sweetened Snacks	64	7.9 (4.1, 15.8)	9.0 (4.0, 15.5)	91.7 (38.0, 158.0)	0.64
Savoury Snacks	34	7.7 (3.5, 11.0)	6.0 (3.0, 10.0)	86.1 (51.9, 157.1)	0.59
Meat, Fish and Egg (solid)	57	6.0 (0, 12.4)	9.2 (4.9, 18.2)	59.7 (0, 110.7)	0.015
Legumes, Nuts, Seeds	50	2.4 (0.4, 5.8)	7.8 (3.9, 16.0)	37.5 (2.4, 83.8)	<0.001
Fruit	38	22.5 (10.0, 35.0)	17.0 (6.0, 32.5)	94.0 (52.0, 136.4)	0.64
Starchy Root and Vegetables	30	18.0 (7.0, 24.0)	15.5 (6.0, 43.0)	50.0 (19.4, 120.0)	0.12
Milk and Yogurt	15	11.8 (5.2, 41.0)	8.0 (1.0, 29.0)	90.1 (36.8, 183.2)	0.82
Non-dairy beverages	33	47.3 (27.5, 76.1)	27.7 (9.0, 86.3)	98.1 (43.8, 123.5)	0.28
Soup/Broth from Relish	94	17.0 (11.7, 26.0)	7.4 (0, 16.9)	138.5 (80.0, 243.1)	<0.001
LNS	68	8.1 (4.5, 11.8)	4.5 (2.0, 9.0)	148.7 (95.0, 274.0)	<0.001

[a] Refers to the number of participants~~respondents~~ where this food group was present on the WFR, i-24-HR, or both. This includes the average portion size estimation per food group per participant. In the case where one was an intrusion, this resulted in a reference value of zero, and in the case where there is an omission, this resulted in a reported amount of zero. This is the participant~~respondent~~ average per food group.

[b] median daily average per participant of reported amount derived from i-24-HR

[c] median daily average per participant of reference amount derived from WFR

[d] Percentage agreement: (Reported amount / reference amount) x 100

[e] p-value derived from Wilcoxon signed-rank test for matched pairs

i-24-HR: interactive 24-hour recall, LNS: Lipid-based nutrient supplement, WFR: weighed food record

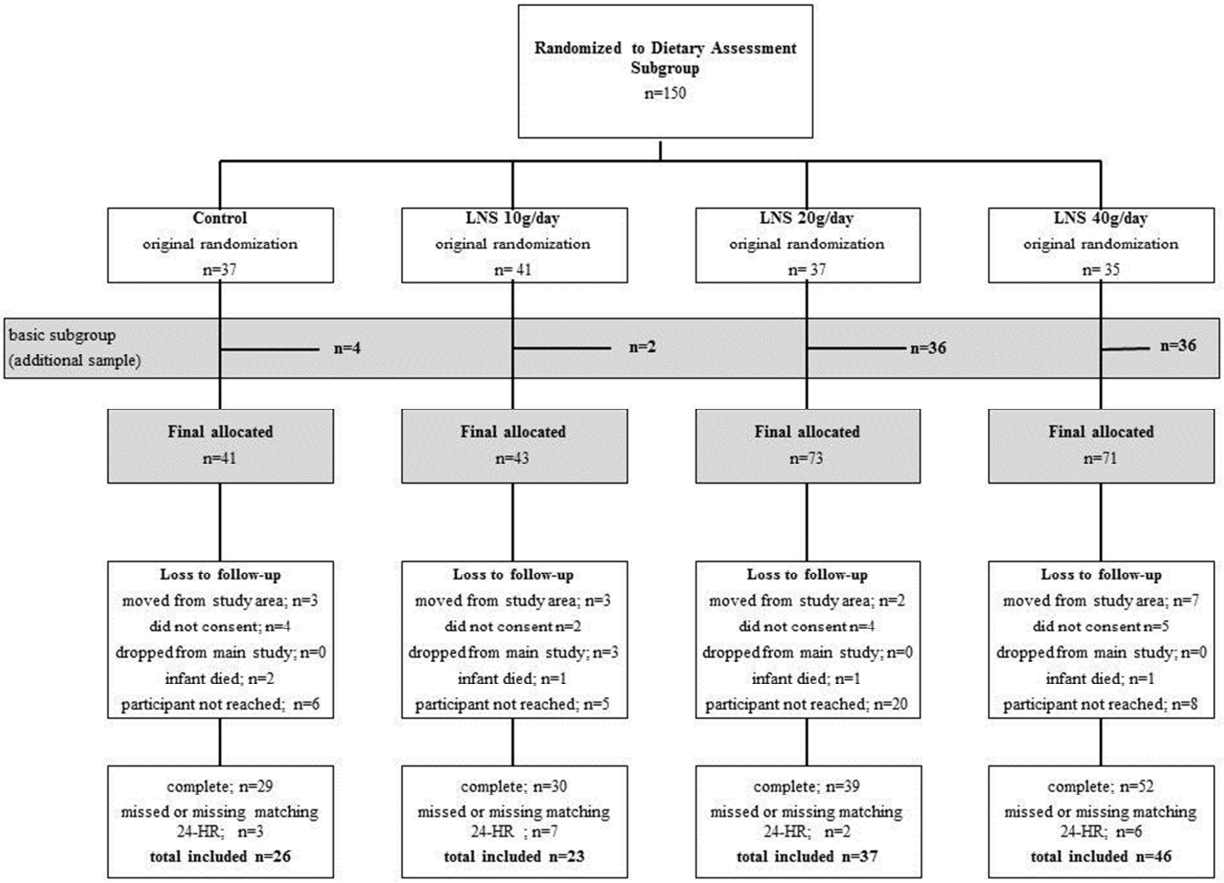
Online supplement Table 2: Comparison of i-24-HRs that corresponded to and were independent of the WFR. An Assessment of bias in reporting related to the presence of the WFR: the "instrument effect".

	N=71 Median Intake (25^{th},75^{th} percentile)			
Nutrient	**Independent 24-HR Recall**	**i24-HR WFR**	**Absolute Difference[a]**	**p-value[b]**
Energy (kcal/d)	375 (273, 553)	327 (246, 463)	-34	0.10
Protein (g/d)	8.8 (5.8, 12.5)	7.6 (5.0, 10.3)	-0.78	0.06
Fat (g/d)	9.8 (5.0, 15.4)	8.1 (4.2, 11.8)	-1.9	0.06
Fe (mg/d)	3.2 (1.9, 5.8)	2.6 (1.7, 5.3)	-0.2	0.50
Zn (mg/d)	2.2 (1.2, 5.9)	2.0 (1.2, 6.1)	-0.1	0.97
Ca (mg/d)	115.9 (41.5, 204.3)	104.9 (34.7, 208.5)	-1.1	0.48
Vitamin A (µg RAE/d)	122.9 (30.3, 262.9)	107.9 (20.5, 292.9)	0	0.79

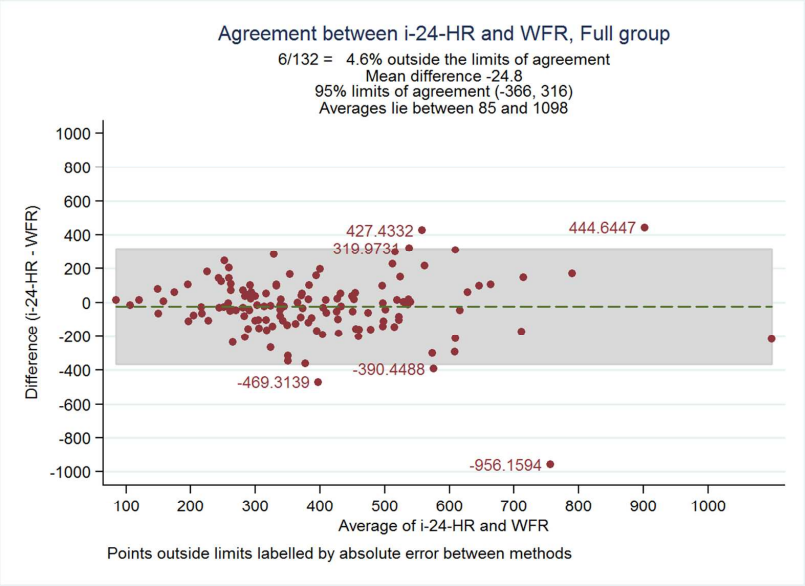
[a] i-24HR WFR – Independent 24-HR

[b] Wilcoxon signed rank matched-pairs test

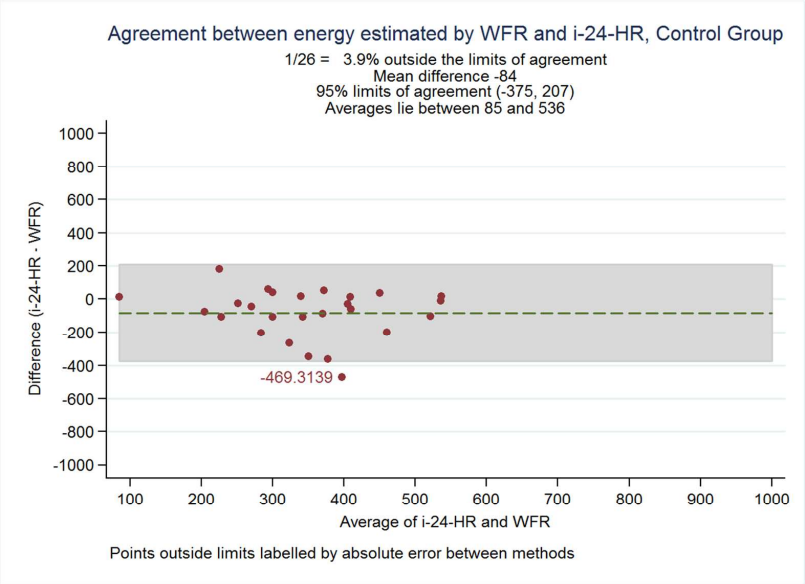
Figure 1: Consort Flow Diagram of Participant Enrolment and Inclusion in the Validation Sub-Study



Online supplement Figure 1: Bland Altman Plot Showing Relative Agreement in energy (kcal/day) estimation between WFR and i-24-HR: Pooled Group



Online Figure 2a: Bland Altman Plot Showing Relative Agreement in Energy (kcal) estimation between WFR and i-24-HR: Control Group



Online supplement Figure 2b: Bland Altman Plot Showing Relative Agreement in Energy (kcal) estimation between WFR and i-24-HR: Intervention Group