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Reducing Child Undernutrition through Dietary Diversification, Reduced Aflatoxin Exposure, and Improved Hygiene Practices: The Immediate Impacts in Central Tanzania

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ABSTRACT

The study aimed to quantify the immediate effects of dietary diversification, food safety, and hygiene interventions on child undernutrition in four rural villages in Kongwa district of central Tanzania. One hundred mothers with their children of less than 24 months old were recruited for this study. The difference-in-difference (DID) method was used to assess the effects of intensive intervention through a learning-by-doing process on the topic of aflatoxin free diversified food utilization and improved hygiene practices. Periodic anthropometric measurements were conducted on the 0th, 7th, 14th, and 21st days, and DID estimator showed the significant and positive average marginal effects of the intervention on Z-Scores being 0.459, 0.252, and 0.493 for wasting, stunting, and underweight, respectively. Notably, at the end of the study, the mean aflatoxin M₁ level in urine samples decreased by 64% in the intervention group, while it decreased by 11% in the control group. The study provides quantitative evidence on intensive 21-day training for mothers incorporating integrated technologies yielded positive impacts on their children's nutritional outcomes.

KEYWORDS

Undernutrition;
complementary food;
aflatoxin exposure;
difference in difference

Introduction

The United Nations' Sustainable Development Goal 2 aims to end all forms of malnutrition by 2030, including a significant reduction in stunting and wasting in children under 5 years of age by 2025. Globally, undernutrition is

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a leading cause of one-third of deaths among children (Black et al. 2008). Undernutrition severely affects early child growth, cognitive development, social development, and ultimately, economic growth (Connell and Smith 2016; Jukes et al. 2002). Undernourished children are prone to frequent infections that can be severe, long lasting and may lead to a spiral of ever-worsening nutritional status than for well-nourished children (Neumann and Harrison 1994).

Wasting, stunting, and underweight are the three popular indicators for the assessment of undernutrition (Seetha et al. 2018b). Wasting can be caused by an extremely low energy intake, for example, due to hunger induced by poverty as well as famine due to crop failure and natural disasters, nutrient losses due to diseases such as diarrhea, or a combination of both. Stunting is a multifactorial impairment in linear growth as a result of undernutrition, recurrent infections from water-borne diseases, substandard health care, environmental enteropathy (EE) due to improper sanitation, and exposure to aflatoxin (Nandy and Miranda 2008; TFNC 2014). Underweight among children is a composite measure of wasting and stunting and is regarded as an overall measure of undernutrition (WHO 1997). According to the World Health Organization (WHO) classification, stunting rates above 40% are considered to be in the “very high” severity range and alarming to the economy (Onis and Blössner 2003; WHO 1997). In Tanzania, the rate of stunting or chronic undernutrition among children of ages between 0 and 59 months was estimated at 34% in 2015–2016, having decreased from 42% in 2010 (TDHS-MIS 2015–2016).

The most direct cause for child undernutrition is the deficiency in macro- and micro-nutrients. The CGIAR, through its Systems Level Outcome 2, aims to improve food and nutrition security by, among other things, improving diets for the poor and vulnerable, and food security and health for humans and animals (CGIAR 2016-2030), this study being one of those efforts. In Tanzania, more than 90% of the rural population depends on agriculture and predominantly cereal-based staple food consumption. In central Tanzania, maize is the major ingredient of porridge that is fed to children, with little or no vegetables or proteins rich products included in the meals (Muhimbula and Issa-Zacharia 2010). A previous study conducted in rural Tanzania reported that infants’ per-capita consumption of maize was relatively high at 43 g/day, as a single staple with limited diversification (Kimanya et al. 2009). Rice is the second most cultivated cereal crop in central Tanzania but is largely sold for income (Nakano et al. 2018). Notwithstanding the production and availability of diverse nutrient-dense crops such as groundnut (*Arachis hypogea*), pigeon pea (*Cajanus cajan*), pearl millet (*Pennisetum glaucum*), and sorghum (*Sorghum bicolor*), their inclusion in complementary foods remains low on average as maize forms major complementary food (Kimanya et al. 2009). The limited inclusion of

such nutritionally beneficial crops in diets is underpinned by social and economic drivers such as culture, drudgery associated with processing and cooking, and limited awareness of the health benefits, as the society's food systems have morphed over time to more maize-based diets. In general, rural households produce the nutrient-dense crops for sale benefiting affluent urban consumers and distant markets rather than local-undernourished communities.

Another important factor linked to growth impairment especially among children is the exposure to mycotoxins, in particular, aflatoxin B₁ (AFB₁) (Turner 2013; Wild and Gong 2010), a harmful mycotoxin which contaminates a variety of staple food crops (Wild and Gong 2010). Commonly grown crops in Kongwa district of central Tanzania such as maize and groundnut have AFB₁ contamination in freshly harvested produce, which is at even higher levels in stored produce due to improper post-harvest handling methods used by farm households (Turner 2013). Chronic exposure to AFB₁ impairs child growth (Gong et al. 2002) possibly by restricting dietary nutrient uptake. The presence of AFB₁ in maize samples from different agro-ecologies of Tanzania including Iringa, Tabora, and Kilimanjaro has been reported (Kimanya et al. 2008). This suggests that households from such agro-ecologies that consume contaminated cereals may be exposed to aflatoxins. Indeed, a longitudinal investigation on the exposure to aflatoxins by infants and young children in Tanzania suggested that it could be a contributory factor to early childhood growth impairment (Shirima et al. 2015). To date, only one study in Tanzania has found statistical evidence of the relationship between the presence of AFB₁ or fumonisin in diets, children's exposure thereto, and the presence or absence of growth impairment (Chen et al. 2018a).

Another crucial factor associated with undernutrition is poor hygiene practices. Enteric infections are common in the first year of child life, especially in low-income communities, and known to contribute to growth impairment (Arnold et al. 2013). Unclean and/or contaminated drinking water, especially with fecal and other solid and soluble matter is a leading cause of morbidity and mortality among children under 5 years in developing economies (Arnold et al. 2013; Hadi, Dibley, and West 2008). In all populations, frequent diarrhea and intestinal worm infestation, coupled with undernutrition in the first 5 years of life, lead to negative outcomes on health, cognitive development, and human capital (Adair et al. 2013; Crimmins and Finch 2006; Hadi, Dibley, and West 2008; Victora et al. 2008). WHO estimates that half of the undernutrition cases especially in developing economies are associated with repeated diarrhea or intestinal worm infestation originating from drinking unsafely contaminated water. However, cluster-randomized controlled trials conducted in Bangladesh and Kenya show that there was no benefit of integrating nutrition with water, sanitation and

handwashing on child linear growth and cognitive development (Luby et al. 2018; Stewart et al. 2018). It is noteworthy that in both sites of the intervention, most participants had access to basic latrines and had an improved drinking water source at baseline (Arnold et al. 2018). Moreover, nutrition interventions are not always one size fits all and have to be customized to specific cases. Nonetheless, it remains important to secure microbiologically safe water for drinking purposes in order to prevent diarrhea incidences and other health hazards.

There are no reports that examine the impacts of interventions that combine nutrition, food safety, and WASH on the growth of infants and young children. While these three issues closely interact with each other especially in rural households of developing economies, most of the past interventions did not incorporate all three aspects. In particular, the food safety aspect tends to be unintegrated. Aflatoxin contamination mitigation in food and food products is usually handled at all stages of food value chains, i.e., during crop production, post-harvest handling, storage, and marketing. This study is the second after Seetha et al. (2018b) that integrates the three aspects in order to inform the design of mitigation programs for a common but complex driver of nutrition-related poor health among infants and children. Accordingly, the aim of this study is to (i) quantify the short-term effects of intervention integrating dietary diversification, food safety, and hygiene on child growth, (ii) assess dietary AFB₁ levels in maize samples (food source), and 24-h exposure to dietary AFB₁ as measured in the form of aflatoxin M₁ (AFM₁) in urine, and (iii) determine the correlation between AFM₁ and child growth.

Methods

The intervention was continuously administered for 21 days in October 2015 in five villages of Kongwa and Kiteto districts in the Dodoma Region of central Tanzania. The difference-in-difference (DID) method (Seetha et al. 2018b; Ashenfelter and Card 1985) was adopted for which baseline, mid-line, and end-line measurements were performed with both the intervention group and control group. The different aspects of our methodology are described in more detail in the following subsections.¹

The difference-in-difference difference-in-difference method

The difference-in-difference (DID) method estimates intervention effects by systematically comparing pre- and post-intervention differences in the outcome between the intervention group and the nonintervention group (i.e., control group) (Donald and Lang 2007; Lechner 2011). Simply considering the change in the status of the beneficiaries before and after the intervention

fails to provide a reliable estimate of the intervention effects since the change would have occurred to non-beneficiaries anyway due to other factors than the intervention. Likewise, simply considering the difference in status between the intervention group and control group after the intervention fails to provide a reliable estimate of the intervention effects since the original status may have been different even before the intervention. The DID method controls for these biases and is regarded as best suited for controlled interventions with multiple-period measurements including the baseline (Tsusaka et al. 2016). This study has four periods, enabling monitoring of the over-time progress in intervention effects on anthropometric outcomes of the studied children. The most important assumption under the DID model is the parallel trends assumption. If this assumption holds true, then the estimation biases are minimized and thus credible inference on the intervention effects is upheld (Nakano et al. 2015). This assumption holds better when measurement intervals are short as was the case with this study.

Study participants

Mothers with infants aged between 6 months and 23 months old, who had started consuming complementary food, had no congenital disorder and were capable of swallowing complementary food were purposively selected from rural communities in Mlali, Moleti, Laikala, and Chitego villages of Kongwa district and Njoro village of Kiteto district which were chosen as the project sites due to the similarity in cropping systems across these areas. For the baseline, 100 mother-child pairs were selected based on a random sampling of 20 pairs from each of the five villages. The total sample size of 100 was sufficient to represent the targeted population according to Yamane Formula at a 10% margin of error. To avoid the knowledge spillover from the treatment group to the control group, the two groups were not mixed in the same village. Thus, a village-level assignment was used for providing the intervention. One village (Chitego village) was randomly selected as a control village from which all the 20 baseline pairs participated, whilst 68 out of the remaining 88 pairs agreed to participate from the four other villages as the intervention group, rendering the total sample size 88. The unequal sizes of the two groups were due to the project monitoring and evaluation requirement to reach the target number of beneficiaries, coupled with the limited budget to increase the control group size. The disadvantage of unequal sizes is a weak statistical power of estimation of the effects of intervention, resulting in underestimation of the effects. In other words, as long as significant effects are detected with unequal sample sizes, they will likely be detected with equal sample sizes as well (Dumville, Hahn, and Miles 2006; Gail et al. 1976; Pocock 1995). Ethical approval was obtained from the Ministry of Health, Community Development, Gender, Elderly and

Children², and personal written consent was obtained from mothers who participated in the study.

Data collection

On the day before the start of the trial (i.e., Day 0), baseline anthropometric measurements of children were recorded, and dietary assessment was conducted through collecting and recording the last 24-h dietary intake details using a survey questionnaire. The dietary data collected were used to estimate the nutrient content of the complementary food that was fed to children in the baseline condition using the Tanzania food composition table (Lukmajni et al. 2008).

A semi-structured questionnaire was administered to capture primary data on demographic and socio-economic characteristics of households, knowledge on nutrition, hygiene, and cleanliness, aflatoxin awareness, ingredients used for complementary foods, agricultural practices, and infant and young child feeding (IYCF) practices. During the 21-day trial, information on disease incidence and food acceptability was continuously collected daily, while mid-line and end-line anthropometric measurements were recorded on Day 7, Day 14, and Day 21. Weight was measured using calibrated Salter scales (Salter Brecknell, 235-6s), while height was measured by taking recumbent length using a horizontal height-measuring board.

Sample collection

Samples of the grains used for complementary food preparation and children's urine were collected for assaying AFB₁ and AFM₁ levels, respectively. Among the 68 participants from the intervention group, crop samples from 57 participants were collected of which 52 mothers also provided urine samples. The reduction in number was due to a combination of leakage in bags during transport and failure of mothers to collect urine samples. From the control group, samples of complementary food and urine were collected from all the 20 households. These samples were then subjected to laboratory analysis as elaborated in the subsequent subsections.

Urine samples were collected from children during the baseline and end line. They were advised to collect an early morning urine sample of their children, which is generally more concentrated and tends to contain higher levels of metabolite for analysis. To ensure that the urine was collected from the intended child, each mother or child caretaker was trained not to mistake the sample from unintended children and was provided with a pediatric urine collection bag with a label. After the collection, the pediatric urine collection bags were immediately transported in dry ice boxes to the laboratory for AFM₁ quantification. The urine samples collected from the children were given numbers before assay.

Quantification of AFB₁ contamination of food sources (maize and groundnut)

To assess the extent of contamination of food sources, which is the avenue for exposure of infants and children to aflatoxins, 100 g each of shelled maize and groundnut samples was collected from the participating households for AFB₁ assay. The representative sample was obtained by mixing 10 g each of 10 samples collected from multiple parts of the bag to constitute 100 g of sample. The samples were air-dried and immediately processed as mentioned earlier (Monyo et al. 2012; Seetha et al. 2018a). In brief, 100 g samples were powdered finely and two replicates of the samples of about 20 g each was mixed with 100 ml of 70% methanol (v/v), with 0.5% potassium chloride (KCl) and blended further. The mixture was shaken at 300 rpm for 30 min and filtered through Whatman No. 41 filter paper, and filtrate was subjected to enzyme-linked immunosorbent assay (ELISA), where the colorimetric reaction of enzyme-substrate was measured in an ELISA plate reader (Multiscan reader, Thermo Fisher scientific, China) at 405 nm to quantify the aflatoxin content (Monyo et al. 2012; Reddy et al. 2001). ELISA used for detecting AFB₁ has a detection limit of 1 µg/kg and quality control of the method was monitored using naturally contaminated known corn reference materials (4.2 and 23.0 µg/kg of AFB₁, product no. TR-A 100, batch number A-C-268 and A-C 271; R- Biopharm AG, Darmstadt, Germany).

Assay for dietary exposure to AFM₁

To assess the children's exposure to dietary aflatoxins, we assayed for AFM₁, which is regarded as a 24-h metabolic product of aflatoxin in urine (Ali et al. 2015; Chen et al. 2018b) Analysis was performed using a commercial ELISA kit (Sigma-Aldrich- Germany) according to the manufacturer's instruction. The urine samples collected from children were filtered and the clear solution was taken for further analysis as elaborated in Seetha et al. (2018a).

Integrated intervention on dietary diversity, food safety, and hygiene

The Positive Deviance (PD)/Hearth model (Kinfu 2013) was used for the delivery of knowledge to mothers on diversified food consumption, AFB₁ mitigation, and hygiene practices. That is, the practices of those mothers having well-nourished children were taken into account in designing the intervention package in order to ensure that the package was acceptable by the local culture, and such mothers were assigned leadership roles during the training process in order to help impart the improved practices recommended by the project. The key steps for this approach were adopted from the PD/Hearth program manual (CORE 2002; Pascale, Sternin, and Sternin

2010), and customized to the social and economic contexts of the locality. This includes the utilization of locally available and affordable crop produce, the use of culturally appropriate and acceptable recipes (which allow them to relate to what they know) according to the taste of the community, and the involvement of lead women farmers and lead health workers in the training. During the 21-day training period, all mothers and their children in the intervention group gathered in a commonplace and were trained by community nutrition extension staff, health staff, and project scientists. The training program underscored the importance of nutrition content in complementary food, the importance of following good hygienic practices, and how to choose quality grains for complementary food preparation to minimize exposure to AFB₁. The training was hands-on, with complementary food preparation for each day left to one or two mothers under the supervision of the study team.

More specifically, during the 21-day training period, the following activities were undertaken:

- (1) A nutritious complementary food package comprising locally available crops, namely, pigeon pea (for protein), finger millet (*Eleusine coracana*) (mainly for calcium and other micronutrients), soybean (*Glycine max*) (for protein), maize (for carbohydrate; the traditionally accepted ingredient), carrot, sweet potato, pumpkins or papaya (vitamin A rich vegetable and fruits), and leafy amaranth or locally available leafy vegetables (*Amaranthus blitum*) (for minerals) was developed by incorporating good practices identified within the community. The ingredient proportions were calculated to provide children with appropriate amounts of protein, fat, carbohydrates, and essential micronutrients including zinc, iron, and calcium as indicated in the first column of Table 1. Information on the importance of nutrition was provided through hands-on training on cooking the most accepted recipe that was formulated for their children. Mothers were also trained to choose ingredients using the food group approach to

Table 1. Comparison of nutrient values: recommended recipe vs. usual recipe at baseline in central Tanzania, 2015.

Nutrient (unit)	Nutrient value per 100 g of meal; Mean (SD)	
	Newly introduced complementary porridge	Usual complementary porridge at baseline
Energy (Kcal)	478 (154)	215 (104)
Protein (g)	19.4 (15.6)	7.9 (2.1)
Calcium (mg)	330 (88)	48 (18)
Iron (mg)	6.8 (1.8)	2.3 (1.4)
Zinc (mg)	3.0 (0.6)	2.9 (1.8)

NB: The values are based on the documented information on consumption.

include diverse nutrients in complementary food preparation. After the initial showcase, the mothers began preparing the meals using their own farm produce. Home vegetable gardening is one of the methods trained as part of the intervention to ensure sustainable production for household consumption and income.

- (2) The mothers (who were also farmers) were educated on the consequences of exposure to aflatoxins and how best to minimize contamination. Specifically, they were taught proper pre- and post-harvest crop handling methods such as mulching, harvesting without damaging pods or cobs, drying on a sheet rather than on soil, drying adequately to reduce moisture, sorting of damaged and rotten grain from the lot to be ground into flour, and using proper storage to ensure AFB₁ free grains with which to prepare complementary food, by incorporating the pre- and post-harvest practices in the same population as recommended by Seetha et al. (2017) and Munthali et al. (2016). Furthermore, crop samples were collected and tested in the laboratory for AFB₁ content, for which the result was communicated back to the participants.
- (3) Recommended hygiene practices were listed and explained to the mothers, which included boiling water for cooking and drinking; washing vessels before cooking; washing hands after using the toilet and before cooking and feeding; and maintaining cleanliness of the surroundings. These practices were implemented by the mothers every day during the 21-day intervention period.

Statistical analysis

Descriptive statistics were used to present the nutrient values, dietary habits, aflatoxin contamination in crop and urine samples, and the extent of undernutrition, whilst inferential statistics such as Spearman's correlation coefficient (Gauthier 2001) and regression analysis were employed to statistically examine the relation between key variables and the effects of the intervention on undernutrition. As indicators of child undernutrition, the Z-Scores for wasting, stunting, and underweight (Seetharaman, Chacko, and Shankar 2007) were calculated using WHO Anthro version 3.2.2. Use of Z-Scores in growth studies is essential and common since growth in Z-Scores indicates improvement in undernutrition compared to improvement in the reference population in general. Otherwise, the result would not be conducive to implying their improvement in growth, because rapid growth occurs intrinsically among such young children. These undernutrition indices were subjected to the analysis of the intervention effects using the DID method. Drawing on the availability of household-level panel data, the fixed effect and random effect regression models

were adopted (Tsusaka and Otsuka 2013a, 2013b) as well as the ordinary least squares (OLS) model for comparison. The panel regressions are capable of controlling for unobservable time-invariant individual heterogeneity among the sample children. One important note is that with multiple time points in data, the standard errors of the estimated coefficients need to be adjusted for autocorrelation (Bertrand, Duflo, and Mullainathan 2004). The easiest remedy, which was adopted in this paper, is clustering on the household identifier to allow for arbitrary correlation of the regression residuals within household-specific time series. In all the three regression models, the coefficients of the DID variables would capture the effects of the intervention. All the quantitative analyses, both descriptive and inferential statistics, were handled with STATA version 14 (StataCorp 2015).

Results

Current practices

According to the baseline survey, communities in the study district (Kongwa) were largely agrarian, where households mainly produced maize, sorghum, groundnut, pigeon pea, finger millet, sunflower (*Helianthus annuus*), and bambara nut (*Vigna subterranean*). These crops were produced as intercrops or mixed crops, primarily for income, with some consumed by families and some retained for seed. Crop management practices in different processes such as harvesting, drying, grading, sorting, and storage were found to be inadequate. For example, only 34% of the farmers adequately dried to less than 10% moisture content level in their produce before storage. In Laikala and Chitego, farmers dried their crop produce on bare ground in their homesteads, especially when the harvest was in large quantities. Only 14% of the farmers graded their grain by sorting out shriveled and rotten grain from healthy ones before storage.

The baseline 24-h dietary history shows that all the children were fed with watery maize porridge and/or breast milk. Although maize, finger millet, and groundnut were the most common ingredients used in complementary porridge, the consistency of porridge was generally light, leading to increased moisture content at the expense of nutrient content in the porridge. The mean intake of essential macro- and micro-nutrients namely protein, calcium, and iron was 7.9 g, 48.0 g, and 2.3 g, respectively, as shown in the second column of Table 1. Table 2 shows the indicators for dietary diversity and IYCF practices. In both the intervention and control groups, seven out of every 10 children were exclusively breastfed until the age of 6 months. The Mean Dietary Diversity (MDD) score for children was generally low. The percentage of children meeting the minimum dietary diversity was 39% in both groups, whilst that for minimum meal frequency and minimum

Table 2. Baseline dietary diversity and Infant and Young Child Feeding (IYCF) practices in central Tanzania, 2015.

Indicator	Intervention group (<i>n</i> = 67)	Control group (<i>n</i> = 20)
Exclusively breast fed (EBF) till 6 months; %	69.1	69.0
Dietary diversity score; Mean (SD)	2.0 (1.1)	3.2 (1.1)
Met minimum dietary score; %	39.0	38.8
Meal frequency; Mean (SD)	2.3 (0.9)	2.3 (0.9)
Met minimum meal frequency (MMF); %	43.7	50.0
Met minimum acceptable diet; %	18.4	22.2

NB: 6–23 months old.

acceptable diet was 44% and 18% respectively in the intervention group, and 50% and 22% respectively in the control group.

The main source of water for most (>90%) of the households was boreholes. The majority (87%) of the participants never treated their drinking water. One quarter of the households did not have access to protected pit latrines (i.e., with a slab). The use of unprotected pit latrines could provide media for transmitting microorganisms that cause pathogenic diarrhea and other symptoms. Nearly 80% of the children had experienced diarrhea prior to the intervention due to inadequate personal hygiene practices.

The grain samples intended for porridge preparation were contaminated with aflatoxins, with the mean and maximum AFB₁ levels of 38.3 µg/kg and 271 µg/kg, respectively, in the intervention group, and 34.0 µg/kg and 245 µg/kg, respectively, in the control group (Table 3). The urine samples collected from the children showed the mean and maximum AFM₁ levels of 57.1 pg/ml and 614 pg/ml, respectively, in the intervention group, and 60.3 pg/ml and 431 pg/ml, respectively, in the control group. Spearman's coefficient of correlation between AFM₁ in urine and AFB₁ in grains was not statistically significant in either the intervention or the control group, indicating that correlation between aflatoxin contamination in food ingredients and aflatoxin exposure in children was not established with our data. Likewise, the Spearman's coefficient of correlation between AFM₁ in urine and the Z-Scores for undernutrition was not statistically significant (Table 4).

Among the three indicators of undernutrition, stunting was particularly prevalent among the sampled children with the average Z-Score at -1.23 (Table 4). The rates of undernutrition were 7%, 31%, 20% for wasting,

Table 3. AFB₁ contamination in maize and corresponding AFM₁ in urine samples at baseline, central Tanzania, 2015.

Group	Variable	<i>n</i>	Mean	SD	Min	Max	CC ^b
Intervention	AFB ₁ in maize (µg/kg)	57	38.3	65.4	1	271.0	0.210
	AFM ₁ in urine (pg/ml)	61	57.1	110.4	ND ^a	614.0	(<i>p</i> = .147)
Control	AFB ₁ in maize (µg/kg)	16	34.0	65.3	1	245.0	-0.064
	AFM ₁ in urine (pg/ml)	16	60.3	126.9	ND ^a	431.0	(<i>p</i> = .815)

^aNot detectable.

^bSpearman's coefficient of correlation.

Table 4. Undernutrition status in sampled children at baseline, central Tanzania, 2015.

	Z-Score				% of children with Z-Score		CC* with AFM ₁ in urine
	Mean	SD	Min	Max	> - 2	< -2	
Wasting	0.04	1.44	-3.62	4.11	93%	7%	0.029 ($p = .802$)
Stunting	-1.23	1.69	-6.06	2.28	69%	31%	0.067 ($p = .548$)
Underweight	-0.61	1.55	-4.35	3.68	80%	20%	0.022 ($p = .848$)

NB: $n = 87$; 6–23 months old.

*Spearman's coefficient of correlation.

stunting, and underweight, respectively, implying that in the studied villages, underweight was mainly attributed to stunting rather than wasting.

Impacts of intervention

Table 5 shows the estimated DID coefficients which represent the effects of the training for mothers on wasting, stunting, and underweight in their children³. On wasting, the effects of the training were found to be positive and statistically significant for all measurement days (Day 7, Day 14, and Day 21). Conversely, the outcomes on stunting were not as striking as on wasting. The effect was not significant until Day 14, turning significant on Day 21, according to the OLS robust estimation and weakly significant according to the panel regression models. The result is comprehensible since height is much less variable than weight even for children in the growth phase. The quantitative interpretation is that, on average, receiving the continued training raises the Z-Score for stunting by 0.252 within 3 weeks. On underweight, the impacts were similar to the case of wasting. Continued training of mothers raised the Z-Score for underweight by 0.493 on average, over the 21-day intervention period.

On the whole, high degrees of consistency were observed between the three estimation models, indicating that the estimated effects of the intervention on the growth outcomes were robust to altering assumptions behind the models.

The mean AFM₁ level decreased from 57.1 pg/ml to 20.3 pg/ml (by 64%) in the intervention group while it decreased from 60.3 pg/ml to 53.6 pg/ml (by 11%) in the control group, albeit not shown in the table. Moreover, dietary diversity increased from three food groups to five food groups with adequacy in the consumption of food groups. Lastly and importantly, the recipe formula was well received by mothers and children with a high acceptability rate (>90%). Mothers were keen to feed their children with a variety of legumes, cereals, vegetables rich in vitamin A, and other green leafy vegetables as long as they were beneficial for children's growth and health outcomes.

Table 5. Effects of training on wasting, stunting, and overweight in central Tanzania, 2015: Difference-in-difference estimations.

Dependent variable	Wasting Z-Score			Stunting Z-Score			Overweight Z-Score		
	Fixed effect	Random effect	OLS robust	Fixed effect	Random effect	OLS robust	Fixed effect	Random effect	OLS robust
Treatment Dummy	na	0.3225 (0.375)	0.3225 (0.447)	na	-0.7351 (-0.102)	0.7351 (0.103)	na	-0.1556 (-0.704)	0.1556 (0.705)
Day 7 Dummy	-0.1794 (0.302)	-0.1794 (0.301)	-0.1794 (0.607)	0.0444 (0.776)	0.0444 (0.775)	0.0444 (0.166)	-0.1475 (0.253)	-0.1475 (0.252)	-0.1475 (0.1475)
Day 14 Dummy	-0.1125 (0.517)	-0.1125 (0.517)	-0.1125 (0.732)	-0.0138 (0.930)	-0.0138 (0.930)	-0.0138 (0.834)	-0.0956 (0.459)	-0.0956 (0.458)	-0.0956 (0.713)
Day 21 Dummy	-0.2181 (0.210)	-0.2181 (0.209)	-0.2181 (0.528)	-0.0938 (0.547)	-0.0938 (0.547)	-0.0938 (0.152)	-0.2156* (0.095)	-0.2156* (0.094)	-0.2156 (0.429)
DID Day 7	0.3344* (0.083)	0.3344* (0.082)	0.3344 (0.346)	-0.0227 (0.895)	-0.0227 (0.895)	-0.0227 (0.785)	0.2683* (0.061)	0.2683* (0.060)	0.2683 (0.326)
DID Day 14	0.3924** (0.042)	0.3924** (0.041)	0.3924 (0.247)	-0.0213 (0.902)	-0.0213 (0.901)	-0.0213 (0.839)	0.2990** (0.037)	0.2990** (0.036)	0.2990 (0.263)
DID Day 21	0.4594** (0.017)	0.4594** (0.017)	0.4594 (0.199)	0.2523 (0.144)	0.2523 (0.143)	0.2523** (0.038)	0.4932*** (0.001)	0.4932*** (0.001)	0.4932** (0.080)
Constant	0.0438 (0.406)	-0.2194 (0.504)	-0.2194 (0.572)	-1.2306*** (0.000)	-0.6306 (0.121)	-0.6306 (0.116)	-0.6070*** (0.000)	-0.4800 (0.194)	-0.4800 (0.190)
F-statistic (p-value)	F = 2.58** (0.019)	F = 2.58** (0.008)	F = 2.97*** (0.008)	F = 1.46 (0.193)	F = 1.46 (0.193)	F = 3.97*** (0.001)	F = 4.29 (0.000)	F = 5.23*** (0.000)	F = 5.23*** (0.000)
Wald test χ^2 (p-value)		$\chi^2 = 18.71$ *** (0.009)			$\chi^2 = 11.19$ *** (0.131)			$\chi^2 = 25.79$ *** (0.001)	

NB: The estimated coefficients are shown along with the p-values in parentheses. ***, **, and * indicate the statistical significance level of 0.01, 0.05, and 0.10, respectively. The number of observations covering the four periods was 348 in each model (i.e., 87 household times 4 periods). The time-invariant control variables in the Random Effect and OLS Robust regressions are dispensed with from the table, which are mother's age, household head sex, household head age, household head education, household size, and asset holding.

Discussion

The rates of undernutrition in the sampled children from Kongwa district were higher than the WHO standard for a wealthy and healthy economy, as a consequence of low diversity in diets, lack of AFB₁ control in farm produce, and inappropriate hygiene practices. Despite producing different crops, dietary diversity among children was limited as their diet was dominated by a single crop of maize, the traditional staple food for consumption, and in lean months they sold other crop produce to buy maize for their consumption. As a result, all the children were fed with watery maize porridge and/or breast milk, resulting in insufficient dietary diversity.

The farmers' inappropriate pre- and post-harvest crop handling practices aggravated AFB₁ contamination in crop produce and corresponding AFM₁ levels in urine. The previous study by Chen et al. (2018b) shows that AFM₁ levels in Kongwa district were higher in Kilimanjaro and Iringa districts since the samples in Kongwa were collected from stored produce. Crop produce tends to be contaminated particularly during prolonged storage with fungal growth increasing significantly after 5 months especially under warm humid conditions as is common in the tropics (Chen et al. 2018b; Turner 2013).

In the current study, exposure to aflatoxins was detected in the form of AFM₁ levels in urine samples collected from children. A similar study conducted in other regions of Tanzania and Malawi revealed the presence of aflatoxin-albumin (AF-alb) adduct in blood samples of 67% of the children with the mean concentration level of 4.7 pg/mg and 20.5 pg/mg of albumin, respectively (Shirima et al. 2015; Seetha et al. 2018a). In our study, the urine biomarker was estimated, which is an indicator of short-time exposure to AFB₁ whilst the estimation of AF-alb is an indicator of exposure over several months (Chen et al. 2018b), though both of these biomarkers are considered efficient for risk assessment studies. Our data failed to support correlation between the AFB₁ contamination in food ingredients and AFM₁ exposure in children's urine samples, which may be due to the relatively small sample size and only maize and groundnut were tested which left out other possible sources of exposure such as cassava flour in complementary food, breast milk and food ingredients of mother's diet. Likewise, correlation between AFM₁ levels in urine samples and extent of undernutrition among children was not supported by our data. Although correlation was not found between AFB₁ in food and AFM₁ in urine, aflatoxin contamination was detected in both the food and urine samples with 81% of the food samples tested positive and 83% of the urine samples tested positive. It is noteworthy that 44% of the AFB₁ positive samples had AFB₁ levels higher than 20ppb. The vast majority of the population in eastern and southern Africa consume maize as their staple, which can presumably pose the risk of chronic exposure to aflatoxin (Ngoma

et al. 2016). Arguably, the risk associated high AFB₁ contamination in food should not be neglected.

The multiple-period panel regression analysis indicated that the immediate effects of the comprehensive training were generally positive and statistically significant on the indicators of undernutrition among children, particularly of wasting and underweight. The three-week training enhanced the Z-scores for wasting by 0.459 and underweight by 0.493. The result for stunting also suggested a weakly significant and positive change arising from continuous feeding of safe and balanced diets with improved nutrient content especially protein, calcium, and iron compared to the baseline, which would underpin a linear growth in a longer time frame which is out of the scope of this study. Moreover, hygiene training effectively reduced the incidence of diarrhea. In addition, training on grain sorting prior to porridge preparation lowered the levels of AFM₁ in urine to less than detectable level. Furthermore, the intervention contributed to swiftly reducing the incidence of diarrhea suggesting the effectiveness of the improved hygiene practices covered by training. On the whole, the outcome of the intervention was positive toward addressing undernutrition, given the high acceptability of the recipe, which suggests promising potential for disseminating capacity building programs of a similar kind.

There are four major limitations in this study. First, since this study focused on the effects of the comprehensive training, there was one treatment group that received the entire package of intervention. In other words, the effect of a particular subject of the training was not distinguished from that of another subject. Hence, there remains a question of attribution in respect of which subject of the training was effective and what synergy the combination of the subjects brought forth. Second, since our focus was on the real-time effects of the three-week program, longer term effects such as stunting were beyond the scope of this study. The sustainability of the effects of the training would require further investigation. Third, confounding effects of possible aflatoxin contamination in mothers' breast milk and adult food including cassava were not quantified, which could be a factor behind stunting at the initial stage of infants' life. Fourth, mothers were advised to collect only the early morning urine sample. Moreover, considering the number of children and time of sample collection, it was not practical to collect the urine sample from each child by the research team which is a major limitation that the study could not double-check on sample collection procedure.

Conclusions

Undernutrition in children is detrimental to the economic and social development in low-income countries, in particular, sub-Saharan Africa because it affects the future populations of the continent. This study investigated the integrated

effects of three-pronged training on rural agrarian populations of central Tanzania on the undernutrition indicators, focusing on the immediate impacts. The results unequivocally suggest that comprehensive training on improved practices leads to positive outcomes on children's undernutrition, suggesting behavioral changes among their mothers. Direct observation of the immediate outcome helped convince mothers to maintain good practices that were affordable in the communities. Further elicitation and incorporation of local preferences and tastes would help guarantee sustainable adoption. In all likelihood, the result implies the need for policies and institutions to incentivize development agencies and governments to invest in upscaling intervention programs of this sort targeted at relevant mothers in resource poor rural communities.

Notes

1. It is important to note that this study is not a clinical trial but socioeconomic intervention research.
2. Ethical Approval Number: NIMR/HQ/R.8a/Vol.IX/2072.
3. The Hausman test pointed to the random effects model for all the four dependent variables.

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Declaration of Conflicting Interests

The authors declared no potential conflict of interest with respect to the research, authorship and/or publication of the article.

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