Effect of Consumption of Sourdough Pearl Millet (Pennisetum Glaucum) Snack on the Hemoglobin and Zinc status of School-Age Children in Odeda Local Government Area, Ogun State, Nigeria

Ilo Jumoke Georginaⁱ*, Onabanjo Oluseye Olusegunⁱⁱ, Oladoyinbo Catherine Adebukolaⁱⁱ, Sobukola Olajide P.ⁱⁱⁱ

- ii) Department of Human Nutrition and Dietetics, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria
- iii) Department of Food Science and Technology, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria

*Corresponding author: jumoke.famakinwa@yahoo.co.uk

ABSTRACT

Background: Micronutrients are required by the body in minute quantities, and their absence can have highly negative effects. A food-based approach is needed to increase the consumption of an adequate amount of underutilized micronutrient-rich foods.

Objectives: This study assessed the effect of consuming Sourdough excluded pearl millet snacks on the nutritional status of school-age children in Odeda Local Government Area of Ogun State. **Materials and Methods:** One hundred primary school age children were grouped into three study groups (sourdough pearl millet extruded snacks group, pearl millet extruded snack group, and ferrous supplement group) and monitored for eight weeks as part of the study's randomized block single-blind trial design. Blood samples from the subjects were taken at the start and end of the study, and hematological factors were examined (hemoglobin, packed cell volume, serum zinc). ANCOVA was used for statistical analysis. P<0.05 was considered significant

Results: Results revealed the average age of the children was 105.9 ± 27.2 months with 49.0% being female and 51.0% male. 54.9% of respondents were anaemic, while 11.4% tested low in zinc. Haemoglobin and packed cell volume at the endpoint had significantly improved in the three groups (p<0.05). Only the sourdough pearl millet extruded snacks, though, made a 10% improvement in the zinc status.

Conclusion: Sourdough pearl millet extruded snacks improved the hemoglobin status of anemic children and zinc status of children with low zinc levels. This study has demonstrated that sourdough pearl millet extruded snacks is a safe option for the treatment of anaemia in children, particularly those who do not enjoy taking medications.

Keyword: Food-based intervention, Micronutrient deficiency, Underutilized crop, school-age, Nutrition.

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INTRODUCTION:

More people are becoming aware of the numerous micronutrient deficiencies that exist in

underdeveloped nations (1). Strategies that are multifaceted, coordinated, and long-lasting are

I) Department of Home Science and Hospitality Management, (Nutrition and Dietetics option), Olabisi Onabanjo University, Ayetoro Campus, Ogun State, Nigeria

required to fight micronutrient deficiencies (2). The only sustainable and long-term approach to addressing micronutrient deficiencies is to increase access to and consumption of a nutritionally appropriate diet (3). Micronutrient deficiencies are a form of under nutrition that happen when vitamin and mineral intake or absorption is insufficient to support normal physical and cognitive development, health, and function. Because micronutrient deficiencies occur gradually over a long period of time, they are also known as hidden hunger (4).

A frequent micronutrient shortage in Nigeria and around the world is iron deficiency anemia. Children, expectant mothers, women in child bearing age, and adolescent girls are the main populations affected (5). As a result of their increased needs for iron, school-aged children are more likely to develop iron deficiency anemia, one of the main causes of anemia (6). Children in underdeveloped nations are particularly at risk due to increased growth requirements, large helminthes burdens, and diets with low iron bioavailability (3, 7). In Nigeria, it is estimated that 69% of preschoolers have iron deficiency anemia (IDA) (8, 9).

Diet is the main source of zinc intake, and when it is insufficient, biochemical problems and clinical indications appear (10,11). According to Penny (12), severe zinc deficiency causes stunting and failure to thrive in early children. Onyezili et al. (13) in their studyfound that a zinc shortage raises the risk of death from diarrhea, pneumonia, and malaria. Despite the widespread occurrence of zinc in everyday foods, dietary deficiencies are prevalent because meals with a vegetable source tend to contain inhibitory compounds such fiber and phytate, which lower the bioavailability of zinc (12). Ayoyu et al. (14) in Southeast Nigeria and Abolurin et al. (15) in Southwest Nigeria reported the prevalence of zinc deficiency at 43% and 42.8%, respectively. Abah et al. (16) reported that the prevalence of zinc deficiency is alarming among school-age children, with inadequate dietary zinc intake as an important contributing factor.

It is important to promote the adoption of foodbased tactics to meet everyone's nutritional needs and battle micronutrient deficiencies (17). These tactics include the use of nutritional supplements, food fortification, dietary variety and indigenous and traditional foods. Food-based approaches can simultaneously address numerous nutrients, such as dietary calories, proteins, and different micronutrients, without running the risk of adverse nutritional interactions or excess (3). Food-based solutions to combat iron and zinc insufficiency include a wide range of initiatives that seek to boost the production, accessibility, and consumption of micronutrient-rich foods as well as the bioavailability of these elements in the diet. Underutilized crops like pearl millet have been shown to have more nutritional value and potential health advantages than major cereals like wheat, rice, and maize. But the usage of these crops as food is still primarily restricted to rural communities at the household level (18). One of the earliest meals consumed by humans is millet, but as cities and industries developed, people began to use wheat and rice instead (19). Common processing methods result in a large loss of nutrients, which works against enhanced pearl millet usage. Food processing can be a crucial method for raising the nutritional value of foods that are available to the underprivileged in developing nations (20).

A mixture of flour and water that has been fermented by lactic acid bacteria is known as sourdough (21). Previous research has demonstrated that adding sourdough to bread can improve the nutritional value of the dough by improving mineral bioavailability and lowering the amount of Phytate (21). Despite the potential nutritional benefits of pearl millets and sourdough technologies, little is understood about how these nutrients affect people's nutritional and physical health. To encourage its usage as food, this study looked into how eating extruded pearl millet sourdough snacks affected the iron and zinc levels of school-age children in Odeda Local Government Area, Ogun State, Nigeria.

MATERIALS AND METHODS Study Design and Location

The study was a single-blind randomized blocked controlled intervention in which students were

followed for eight (8) weeks. The school-age children attended government-owned primary schools in Odeda Local government in Ogun State. Odeda is a local government area and town in Ogun State, Nigeria.

Sample or study Population

The population comprised of school-age children attending government-owned primary schools, 5-12 years, class one to six, in Odeda Local government in Ogun State.

Sample size determination-

The sample size selection was determined using the formula

 $N = \frac{Z^2 P(1-P)}{d^2}$

where: z = confidence level at 95 (standard value of 1.96%),

at d = 5% acceptable margin of error (standard value at 0.05)

p = prevalence of underweight children amongunder 5 children in Ogun state (32) = 17%

$$N = \frac{(1.96)^2 \times 0.17 \times 0.83}{(0.05)^2} = 216.8$$

plus 10 percent non-responsiveness is 227 children

Sample techniques and procedure

A multi stage sampling techniques was used.

Stage 1: Participating schools were selected from rural schools in Odeda Local government area which was known from previous studies that most of the children are either underweight or stunted and where high levels of micronutrient deficiencies exist (22). Odeda and Ilugun zones are the two main rural settlements in the Odeda local government area. However, Odeda zone was chosen using a simple ballot sampling.

Stage 2: Based on consents by Head teachers and executives' members of Parents Teachers Association. Only two (5) schools gave consents. All the 227 children whose parent gave consent were recruited for study inclusion assessment. Only those who met the inclusion criteria (105 children) were further recruited for the intervention. Stage 3: The study participants were stratified by grade and randomly allocated to the three (3) treatment groups; the intervention (children were fed with the products from sourdough pearl millet extruded snacks), control group 1 (children were fed with the products from Pearl millet extruded snack- not sourdough) and control group 2 (children were given ferrous supplement).

Inclusion Criteria: Children having haemoglobin levels between 8 and 11.5 g/dl, children present on the day of data collection, and children whose parents gave consent and assent to participation (mildly and moderately anaemic) were included in the study.

Exclusion Criteria: Children who refused to participate, whose parents or caregivers forbade them from participating, and kids whose haemoglobin levels were below 8 g/dl were excluded from the study, but not from the intervention itself. Children who were absent on the day that endpoint data were collected were not included in data analysis, although they were included for the intervention. Children with acute infection (clinical signs of fever or reportedly suffer from any infection) and severe micronutrient deficiency (severe anaemia, defined as haemoglobin (Hb) 8 g/dL, clinical signs of vitamin A deficiency, defined as night blindness, and clinical signs of iodine deficiency, defined as visible goitre) were also excluded from the study

Ethical Consideration

The Ogun State Hospital, Ijaye Abeokuta, Reference Number: SHA/RES/VOL.3/009, provided ethical clearance. Traditional leaders, the Ogun State Universal Basic Education Board (SUBEB HQ and SUBEB Zonal), the Primary Schools Authority, parents' teachers' association executives, individual parents, and the director of Primary Healthcare Odeda Local Government all gave their consent for the intervention study to be conducted. Only kids who consented and whose guardians or parents consented were included in the study. The participant had the option to refuse the request if they so desired. Following the signing of the consent form, the volunteer and willing subject took part. The children's caregivers signed on their behalf.

Data Collection

Two weeks before the start of the intervention, baseline data were collected from the five schools. Blood samples were collected from 227 children to determine the baseline haemoglobin status for the inclusion criteria selection. The 105 children, who met the criteria, were further selected for the feeding trial and de-wormed with oral de-worming drugs by the primary health care nurse. Data collection was at 2 interceptions (Baseline and end-point). Data collection took place at the schools.

Allocation of Study Groups

The three (3) study groups were randomly assigned to the participants in the study after stratification by grade (Figure 1). These study groups include the Intervention group, which included 33 children and fed them sourdough pearl millet extruded snacks, the Control group, which included 33 children but provided them non-sourdough pearl millet extruded snacks, and the Control group 2, which included 34 children (children were given 200mg dried ferrous sulphate). One hundred and five (105) children who were mildly to moderately anaemic were added to the study's participant pool. Two interceptions were used to gather data (Baseline and end-point). Data were gathered on the school's grounds. All names were kept private and apart from the information gathered. Personal information and the gathered data were stored anonymously, without identities. One child left the program because they didn't like the snack, and the other four abstained on the day of endpoint data collection. There were just 100 children accessible for the endpoint data collection.

Blood Samples Collection

Blood samples from the children in the five schools were taken as baseline data two weeks prior to the start of the intervention by the primary health care nurse and a laboratory technologist. Two hundred and twenty-seven (227) children's blood samples were obtained in order to assess the haemoglobin status for study recruitment. One syringe per each child was used to draw blood samples (10mls) in the early hours of the day from the children's arm while in sitting position.

The Study Snacks

The pearl millet snacks came in two varieties: sourdough and non-sourdough. Sourdough pearl millet extruded snacks underwent the sourdough preparation process, but non-sourdough pearl millet extruded snacks did not. Both snacks were produced and their iron and zinc concentrations were determined. The analysis's findings were applied to establish the study's snack weight distribution. With the exception of public and midterm vacations, the children received the extruded snacks and supplements every school day (5 days per week). While the supplement was purchased from the drug stores, the extruded snack was created by the Mboss project and distributed to study schools. During lunchtime at school, the teachers gave the kids the food and supplements. The weight $(40g \pm 2)$ of the intervention extruded food and the control snack were identical.

Biochemical Assessment

A Laboratory technologist and the local government-owned clinical nurse collected a venous blood sample (10 ml) from non-fasting patients into a 6 ml serum tube and a 4 ml EDTAcoated tube. The anticoagulant was fully combined with the obtained blood samples. After being clotted, the blood in the 6 ml serum tube was centrifuged for 10 minutes at room temperature at 3500 rpm. Samples were kept until analysis at -80°C. Samples were analyzed utilizing customized techniques (23). Using a heometer Rapid test kit, hemoglobin was tested within 4 hours of blood collection (Acon laboratory Inc.) Microhematocrit Centrifugation was used to calculate the packed cell volume (Model: JSH-120) The capillary tubes containing blood samples were centrifuged at a rate of about 12000g for 5 minutes. The height of the red cell column was measured, and its ratio to the height of the entire blood column was used to calculate packed cell volume. Using commercial control serum and a flame atomic absorption spectrophotometer (AAS), serum zinc was quantified. Hemoglobin (Hb) 11.5 g dL-111.9 g/dL (5-8 years and 12

years) was considered anemic in children (23). 31% packed cell volume (PCV) indicates anemia (24). 0.65 mg/dl of plasma zinc indicates a zinc insufficiency (25)

Data Analysis

Quantitative data were analyzed using descriptive and inferential techniques to provide frequency counts and percentages. The WHO cutoff thresholds were compared to the biomarkers. Statistical Package for Social Science (SPSS) 20.0 for Windows was used to conduct an analysis of covariance (ANCOVA) to identify differences between study groups. P-values lower than 0.05 were deemed significant.

RESULTS

Both the Sourdough Pearl Millet Extruded Snack and the Pearl Millet Extruded Snack have 25.29 mg and 36.51 mg of iron per 100g, respectively. Sour dough pearl millet extrude snacks and pearl millet extrude snacks both had 1.49 mg and 1.99 mg of zinc per 100g, respectively. The intervention snack provided iron by 101.1% of the RDA for children age 4 to 8 and 126.4% of the RDA for children ages 9 to 13. Control group 1 supplied iron with 146% of recommended dietary allowance for children aged 4 to 8 and 182% for children aged 9 to 13. The intervention snack provided zinc with 10% of the RDA for children ages 9 to 13 and 16% of the RDA for children ages 4 to 8. The control snack provided zinc with 10% of the RDA for children aged 9 to 13 and 15.9% for children aged 4 to 8 in Table 1

DISCUSSION

This study's major focus was on how sourdough pearl millet extruded snacks affected school-aged children's hemoglobin and zinc levels. Results revealed the average age of the children was 105.9 ± 27.2 months with 49.0% being female and 51.0% male. Anaemic individuals were more common than usual (Hb 11.5 g/dl; 11.9 g/dl for ages 5 to 11 years and 12 years, respectively). In contrast to data from other regions of the country, Kuku-Shittu et al. (26) found that iron insufficiency was more common in the Southwest regions of the country. In contrast, Quadri et al.

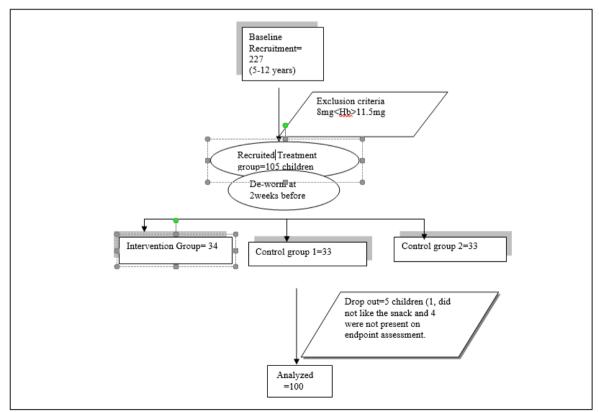


Figure 1: Study trial flow

	Age (Years)	RDA /day	Snack consumed per day	
			SPMS (%RDA)	PMS (% RDA)
Iron	4-8	10mg	101.1	146
	9-13	8mg	126.4	182.5
Zinc	4 -8	5mg	12	15.9
	9-13	8mg	7.5	10

Table 1: Iron and Zinc composition of treatments snacks

Weight of extruded snack; 40g±2, Treatments includes SPMS; sourdough pearl millet extruded snack, PMS; Pearl millet extruded snack (not sourdough) RDA; recommended dietary allowance

 Table 2: Percentage of children that showed improvement (at least 1g/dl) in hemoglobin, packed cell volume and zinc status for all the three study group

Variables	Intervention_Group %	Control group 1 %	Control group 2 %
Hemoglobin	44	48	37
Packed Cell Volume	56	60	48
Serum Zinc	10	-	-

stated that anemia in Ogun State was 16.3%. According to studies conducted in the Northwest of Nigeria by Bello-Manga et al. (28), the prevalence of anemia was also very high, at 40.3%. Worm infestation, hemoglobinopathies, and the consumption of diets high in cereals but poor in bioavailable iron were some of the contributing factors found (inherited blood disorders). The high consumption of tubers, which have been reported to have low iron contents, as well as poor dietary sources of iron or iron in a form that is not available for absorption (23) may be to blame for the prevalence of anemia in this study. Vegetables are a major source of iron, but they are also subject to chelation by oxalates, phytates, and other anti-nutrients, rendering them unavailable for absorption by the body (23). This is due to the fact that the school-age children come from poor socioeconomic status families, but they also engage in home gardening and have access to food sources that ought to boost their hemoglobin levels. The three study groups' impacts on the children's hemoglobin and packed cell volume status were assessed by the study. For the three groups, there was a noticeable

improvement, with control group 1 showing the greatest improvement. When the three study groups were compared to one another, there were no noteworthy differences. The baseline values led to the notable discrepancies that were seen. According to a study by Ayogu and Onah (2019), eating cookies enriched with cowpeas had a substantial favorable impact on haemoglobin and serum ferritin. However, it was anticipated that when compared to other study groups that received snacks made from pearl millet extruded, the kids in control group 2 (the ferrous supplement group) would have a higher haemoglobin status. Sourdough technology and the extrusion cooking process lower the millets' phytate amounts and boost the bio-accessibility of their minerals (30, 21, 31). In this study, the prevalence of zinc deficiency (zinc 65 g/dl) is low (11.4%), but only the children in the intervention group showed a 10 percent improvement. The relatively high blood zinc level of the children at baseline and/or the insufficient zinc content in the pearl millet extruded snack may have contributed to the modest effect of the pearl millet extruded snacks on serum zinc concentrations. The

sourdough technology's impact of zinc could be credited for the 10% improvement seen in the intervention group. A major limitation of the study was that the children were not categorized by gender. This is the first study to investigate the effects of sourdough pearl millet extruded snacks in a school feeding trial without fortification and compared them to control groups. Since biscuits and chin-chin were the two snacks that the children generally consumed, the millet sourdough snack because it is also a form of snack (29) can be enjoyed by this age group. The study only included children who were mildly to moderately anemic. However, the study found improvements in haemoglobin and packed cell volume status, for all the three groups. The three groups (intervention group, control group 1, control group 2) underwent analysis of covariance to see whether baseline values had a significant impact on the endpoint values. According to the findings (p > 0.05), there was no discernible difference between the endpoint values for any of the three groups. The strong impact of the covariate (baseline status) on the endpoint values for serum haemoglobin and packed cell volume explained the observed discrepancy (that is baseline significantly adjusted endpoint). Only the group given the sourdough pearl millet extruded snack saw a 10% increase in serum zinc.

CONCLUSION

This study has demonstrated that sourdough pearl millet extruded snacks is a safe option for the treatment of anaemia in children, particularly those who do not enjoy taking medications. Additionally, governments can employ and use the sourdough pearl millet extruded snacks for a variety of micronutrient deficiencies intervention programs that target children who are zinc- and anaemia-deficient.

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Authorship Contribution Statement

ILO J.G conceptualized, collected data, analyzed data, interpret data and developed the

manuscript. Onabanjo O.O was the main supervisor while Oladoyinbo C.A and Sobukola O.P a were co-supervisors. They guided conceptualization, data collection and data analysis.

Conflicts of interest

The authors declare that there is no competing interests.

Publication Ethics

"Ethical approval for the involvement of human subjects in this study was granted by the Ogun State Hospital, Ijaye Abeokuta, Reference Number: SHA/RES/VOL.3/009 19-07-2018.

The purpose of the study was explained to the participants who gave their consents and also dully filled the informed consent forms. Participants were informed of their freedom to withdraw, or refuse to take part in the study without prejudice

Traditional leaders, the Ogun State Universal Basic Education Board (SUBEB HQ and SUBEB Zonal), the Primary Schools Authority, parents' teachers' association executives, individual parents, and the director of Primary Healthcare Odeda Local Government all gave their consent for the intervention study to be conducted. Only kids who consented and whose guardians or parents consented were included in the study. The participant had the option to refuse the request if they so desired. Following the signing of the consent form, the volunteer and willing subject took part. The children's caregivers signed on their behalf.

Consent for Publication

Consent for publication is not necessary because this manuscript does not have personal data like individual details, images or videos.

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