Serum Zinc Levels in Apparently Healthy Children in Nigeria: Are They Acceptable

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Abstract

Introduction: Despite the importance of zinc in the human body, there is paucity of data on the zinc status of Nigerian children. The aim of this study was to determine the serum zinc levels of children attending the pediatric outpatient clinic of a tertiary hospital in South East Nigeria and to assess their need for routine zinc supplementation. **Materials and Methods:** One hundred children aged 5–60 months were recruited consecutively from the pediatric outpatient clinic. Their socioeconomic class (SEC) was assessed using the tool developed by Oyedeji. Physical examination was carried out to exclude malnutrition and/or liver disease. Samples were collected in the morning from nonfasting subjects and were analyzed using atomic absorption spectrophotometer. Serum zinc deficiency was defined as zinc level <80 µg/dl. **Results**: The overall median (range) serum zinc level was 83.3ug/dl (60–105 µg/dl) while the median (mean rank) serum zinc levels among male and female subjects were 83.4 µg/dl and 84.2ug/dl, respectively (U = 1071.00; P = 0.228). A total of 26 (26%) apparently healthy children had low serum zinc levels. There was no association between gender and serum zinc levels ($\chi^2 = 2.163$; $P = 0.141$). A significant positive but weak relationship was found between SEC and zinc levels (r = 0.208, P = 0.038) but not between serum zinc levels and age of the children (r = 0.185, P = 0.065). **Conclusion:** A significant proportion of Under-5s could have low serum zinc levels. Routine zinc supplementation may be necessary among this age group in Nigeria.

Keywords: Children, serum, zinc levels

Introduction

Zinc is an important micronutrient found in every cell of living organisms.[1] It plays important roles in several body functions. These roles include cell growth, wound healing, mucosal epithelization, deoxyribonucleic acid synthesis, and immune system development.[1,2] Zinc is also a component of various metalloenzymes, cell membranes, and proteins and plays important roles in immune regulation.[2,3] Zinc salts have been shown to have antiviral activity, either directly or through immune modulation against more than 40 viruses.[4]

Zinc is found in high concentration in the flesh of beef, pork, poultry, fish, and shellfish.[5] Lesser amounts are found in eggs and dairy products.[5] Consumption of these increases body zinc levels.[5] On the other hand, phytates in plants form complexes with zinc preventing its absorption from the jejunum.[6] Thus, consumption of gruels made from maize, sorghum, or millet which contains phytates reduce body zinc levels.[6] In addition, rapid turnover of cells and consequent rebuilding during acute and chronic illnesses also reduce zinc levels.[2]

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Zinc deficiency causes decreased lymphocyte response to mitogens,[2] T-cell lymphopenia, a specific CD4+ T-cell population depression, decreased thymic hormone activity, decreased natural killer cell activity[3] and depressed serum concentrations of albumin, pre-albumin, and transferrin.[2,3] Zinc deficiency produces reversible immune dysfunction,[2,3] and an increased risk of infection, particularly diarrhea and pneumonia.[7] A recent study in Ilorin Nigerian reported that zinc levels of under 5 children with diarrhea are lower than those children without.[8] Zinc supplementation for children in resource-poor countries can reduce the incidence and duration of diarrhea[9,10] and pneumonia[9,11] and may reduce malaria morbidity. [12]

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The global prevalence of zinc deficiency is reported to be 31% and zinc deficiency is said to be common in low-income countries of sub-Saharan Africa and South East Asia.[13,14] This is reported to be due to inadequate zinc intake which occurs in one-third of these populations.[15] Zinc deficiency has also been implicated in 4% of childhood deaths and 1% of childhood diseases in Asia, Latin America, and Africa.[14] In Nigeria, it is estimated that about 20% of children are zinc deficient.[16]

Despite the important role of zinc in the human body, there is paucity of data on the zinc status of Nigerian children aged 5–60 months. It is pertinent, therefore, to ask; what is the zinc level in apparently healthy children aged 5–60 months attending the pediatric outpatients clinic in a tertiary health facility in south east Nigeria? Do they routinely need zinc supplementation in our environment? This study, therefore, sought to determine the serum zinc levels in a cohort of apparently healthy children attending the pediatric outpatient clinic of The University of Nigeria Teaching Hospital (UNTH), a tertiary hospital in South East Nigeria and to assess their need for routine zinc supplementation.

Materials and Methods

Study site

This is a prospective and analytical study carried out at the UNTH, Ituku-Ozalla, Enugu. Enugu State is in the South East geopolitical zone of Nigeria. Enugu State is mainly inhabited by the Igbos with Igbo language being the main language used in the area although a large percentage of the inhabitants are able to communicate in English language. The Children Outpatient Clinic of the Paediatric Department of the UNTH, Ituku runs every weekday and sees about 840 patients within the age range of 1 month up to 18 years monthly.

Study population

The subjects were apparently healthy children aged 5–60 months who attend the pediatric outpatient clinic of the UNTH Ituku/Ozalla, Enugu. These children would have attended the clinic at least once prior to the follow-up visit at which enrolment took place.

Ethical considerations

Ethical clearance was obtained from the Health Research and Ethics Committee of the UNTH, Ituku/Ozalla Enugu (Protocol No: NHERC/UNTH/CSA/329/OL.5). All parents/caregivers of eligible children were informed of the purpose of the study, expected procedures, and potential risks and benefits following which a written consent was obtained before data collection. It was a cross-sectional study conducted at the pediatric out-patient clinic of the UNTH Ituku/Ozalla, Enugu state between August 2013 and May 2014.

Inclusion criteria

1. Children aged 5–60 months who are apparently healthy attending the Children Out-Patient Clinic of the UNTH, Ituku-Ozalla
2. Children whose parents/caregivers consented to the study after due counseling.

Exclusion criteria

1. Symptomatic (diarrhea, on-going febrile illness, severe malnutrition, or history suggesting liver disease), children
2. Children receiving zinc supplements at the time of recruitment or within the preceding four weeks as this is likely to influence their zinc levels
3. Children whose parents/caregivers did not consent to the study after due counseling.

Recruitment of subjects for the study

Children aged 5–60 months were recruited from the pediatric outpatient clinic. These were children who are being treated for mild ailments and had come for follow-up. They were recruited consecutively as they presented to the clinic till the sample size was reached. Their age was ascertained from the date of birth as seen from an official document, for example, health care, immunization card. Parents/caregivers who gave consent and whose children fell within the age bracket 5–60 months then were recruited. The pro forma designed for this study had sections on bio data and epidemiological data, detailed clinical history of the subjects (to exclude diarrhea, on going febrile illness, history suggesting liver disease and drug history) and a thorough physical examination. The physical examination was carried out to exclude possible signs of malnutrition and/or liver disease which are known to affect their serum zinc levels.

Socioeconomic class (SEC) was determined using the occupation and educational attainment of the caregivers of the study participants as described by Oyedeji.[17] The SEC was obtained by finding the mean score for the parents' educational attainment and occupation rounded off to the nearest whole number. Where any of the parents were dead, the social class of the child was assessed using that of the living parent. SEC I represent the highest SEC and class V the lowest**.**

The weight, length (for children <24 months), or height (for children ≥24 months of age) were measured. The weight of children < 24 months were obtained using an infant weighing scale (Waymaster Infant Spring weighing scale, Precision Engineering Co [Reading] Ltd, England. These measures weight to the nearest 0.1 kg. The average of three measurements was taken to minimize intraobserver error. The weight value obtained was plotted on the WHO standard growth chart,[18] and the z score was obtained as the intercept between the child's age and the measured weight. A z score <−2 was considered as undernourished and such patients were excluded.

For children ≥24 months, a stadiometer with weighing scale (RGZ 160® Healthmedical Equipment, England), was used. To obtain length for children <24 months of age, the Infantometer (Seca® Model 416 1721009 Secagmbh and co. kg. Hamburg, Germany) which measures length to the nearest 0.1 cm was used. The length was then measured and recorded. However, for children ≥24 months, the height was measured using the Stadiometer (RGZ-160® Healthmedical Equipment, England). The average of three measurements was taken to minimize intraobserver error. The length/height values obtained from these measurements were then plotted on the WHO standard growth chart[18] and the z score was obtained as the intercept between the child's age and the measured height. A z score <−2 was considered as undernourishment and such children were excluded. The obtained values were finally entered into the pro forma.

Sample collection for serum zinc level

Nonfasting blood samples were collected from the subjects in the morning (i.e., before 12 noon). The sample collection was done by the researcher fully assisted by the laboratory technicians. With the child well restrained by the parents/caregiver and a research assistant, the cubital fossa and the dorsum of the wrist were inspected for superficial veins. When visualized, a tourniquet was applied to the wrist or above the cubital fossa as applicable. The area over and around the visualized vein was then cleaned with 76% alcohol swab in a circular manner, from the center (the area to be punctured) outward.[19] This was then allowed to air dry for about 5–10 s.[19] Subsequently, a 23G veroject was inserted into the vein in the cephal direction at an angle of 30 degrees with the bevel of the needle facing superiorly.[19] When adequately situated inside the vein, 5 ml of blood was collected, and dispensed into one 5 ml trace element-free plain tubes (Silver Health Diagnostics), for zinc analyses. The sample bottles were then labeled accordingly. The samples were allowed to stand for about 90 min, (to allow for clotting and retraction),[20] after which it was spun with a centrifuge at 3000 revolutions per min for 10 min. Subsequently, the serum, (usually about 2.0 ml), was separated with a bulb pipette into trace element-free plain tubes. These were clearly labeled and placed in sealed plastic bags in a cardboard canister with absorbent cotton wool and immediately transported in an ice-packed cooler dedicated for this study to Projects Development Institute (PRODA), Enugu, for zinc analysis. These samples were placed in sample racks clearly labeled for the study and stored in a refrigerator at −20°C. The key for this refrigerator was in the sole custody of the laboratory technician.

Zinc analysis

The analysis was done at PRODA, Enugu, using Atomic Absorption Spectrophotometer (Model 210, manufactured by Buck Scientific Cooperation, Connecticut, USA). The machine operates at a wavelength range of 190–900 nm with typical sensitivity in the parts per million range.[20] The analysis was done using the principle of absorption spectrophotometry which assesses the concentration of an analyte in a sample.[21,22] The samples for serum zinc analysis were batched into groups of 20 and stored in a refrigerator (at −20°C) for later analysis of serum zinc. The analysis was done within 3 h of submission by the chief laboratory scientist assisted by the researcher. The instrument was zeroed using deionized water as blank. One milliliter of serum (obtained by allowing blood to coagulate)[20] was aspirated into an air-acetylene flame. This causes evaporation of the solution and vaporization of the free zinc atoms (atomization). A line source (hollow cathode lamp) operating in the ultraviolet (UV)-visible spectral region was then used to cause electronic excitation of the metal atoms, and the absorbance was measured with a UV-visible dispersive spectrometer with photomultiplier detector. The concentration of each sample in parts per million (ppm) was then extrapolated from the calibration curve.

The obtained values were finally entered into the pro forma. The Nigerian food consumption survey of 2003 used 80 μg/dl as the lowest limit of normal for zinc.[16] In this study, serum zinc deficiency was defined as zinc level <80 μg/dl.

Data analysis

Data were analyzed using IBM SPSS Statistics for Windows, Version 19.0. Armonk, NY: IBM Corp (2010). Descriptive statistics which includes frequency, percentage, means, standard deviation, and median were used to summarize categorical and continuous variables. Mann–Whitney (U) test was used to compare median of skewed continuous variables (serum zinc level). The test of association was done using Chi square, relationship between serum zinc levels and age was done using Pearson correlation while that between serum zinc level and SEC was done using Spearman's rho. The level of significance was set at $P < 0.05$. Results were presented in tables, charts, and prose.

Results

Of the 112 children approached for the study, parents/caregivers of seven refused blood sample collection from their children while five blood samples collected were insufficient. Thus, a total of 100 children were recruited into the study. Forty-nine percent of the children were within the 48–60 months of age group while 6% were within the 5–12 months' age group [Table 1]. Majority (99%) of the study population were Ibo. Eight of the children were in SEC I while 10 were in SEC V [Table 2].

Serum zinc levels were not normally distributed, but rather were skewed to the right, with majority of the subjects having serum zinc levels between 60 μg/dl and 105 μg/dl [Figure 1].

The overall median serum zinc level was 83.3 μg/dl while the median (mean rank) serum zinc levels among male and female subjects were 83.4 μg/dl and 84.2ug/dl, respectively ($U = 1071.00$; $P = 0.228$). A total of 26 (26%) of apparently healthy children had low serum zinc levels. Table 3 shows that there was no association between gender and serum

Table 1: Age and gender distribution

Age (months)	Male, *n* (%)	Female, *n* (%)	Total (%)
5-12	5 (9.4)	1 (2.1)	6
>12-24	11 (20.8)	5 (10.6)	16
>24-36	4 (7.5)	5 (10.6)	9
>36-48	8 (15.1)	12 (25.5)	20
>48-60	25 (47.2)	24 (51.1)	49
Total	53 (100)	47 (100)	

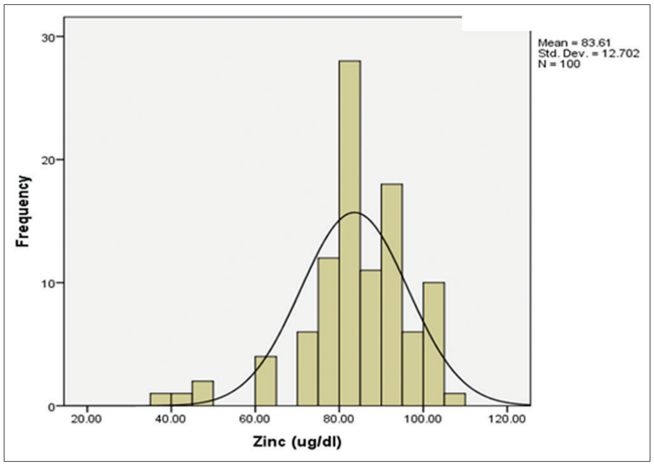

Figure 1: A curve on histogram of zinc levels among apparently healthy children

Table 2: The sociodemographic characteristics of the study population

	n
Tribe	
Ibo	99
Tivi	1
Total	100
Religion	
Christian	100
Muslim	0
Total	100
Socioeconomic class	
Class I	8
Class II	11
Class III	48
Class IV	23
Class V	10
Total	100

Table 3: Association between gender and serum zinc levels in the study population

	Zinc		χ^2	*P*
	Normal	Low		
Gender				
Male	36 (67.9)	17 (32.1)	2.163	0.141
Female	38 (80.9)	9 (19.1)		

zinc levels (χ^2 = 2.163; P = 0.141). A significant positive but weak relationship between SEC and zinc levels (r = 0.208, P = 0.038). No significant relationship was found between zinc levels and age (r = 0.185, P = 0.065) [Table 4].

Discussion

The prevalence of low serum zinc levels among apparently healthy children in the current study was 26%. This was lower than the prevalence of 72% observed in 2001, by Müller *et al.* [23] in rural Burkina Faso; 47.5% obtained in 2003, by Bilbis and coworkers[24] in Sokoto, North West Nigeria and more recently the 32% obtained in 2018 by Afolabi *et al.*,[8] in Ilorin North Central Nigeria. Müller *et al.*[23] used a subsample of a population, (30 small villages and 6 larger villages in Nouna district of North western Burkina Faso). The authors did not state the method of recruitment of this subsample. This may have resulted in some bias in favor of zinc deficiency. In addition, the children investigated were said to have consumed mainly cereals with little protein in their diet. This could account for the low zinc levels as zinc is known to be low in cereals.[5,7] Similar to this study, Bilbis *et al.*[24] in Sokoto, North West, Nigeria, evaluated serum zinc in children below 60 months. Studies[5,13,25,26] have shown that the leafy vegetable, Melochia corchorifolia, and cereals, which is relished among this population, though major sources of zinc,[25] contains significant amounts of phytate,[26] which reduces the bioavailability of zinc for absorbtion.[26,27] This could explain the higher prevalence of zinc deficiency in the sokoto study compared to our study. On the contrary, the prevalence of low serum zinc (26%) reported in our study was higher than the 11% reported in 2004, by Takyi[28] in Ghana as well as the 12.4% reported earlier this year (2020) by Abolurin *et al.*,[29] in Osun state Nigeria.

In the current study, serum zinc was used to assess zinc levels while Takyi[28] used hair zinc to assess zinc levels in their study. Hair zinc is prone to contamination from the use of zinc-containing shampoos[30,31] which increase the hair zinc content. Hair zinc levels also reflect chronic zinc nutrition status rather than acute.[30,31] This could explain the lower prevalence of zinc deficiency obtained in the Ghanaian study. On the other hand, although this current study and that of Abolurin *et al.*[29] were both on Under 5s and used the same method to analyze serum zinc levels, there was a huge difference in the prevalence of children with low serum zinc levels between the two study populations. A number of other factors could probably have been responsible such as the differences in their sample size; study location and differences in the diet of these children. While Abolurin *et al.* studied a total of 250 children; this current study population was 100. Again while this study was in Enugu south east Nigeria; the study by Abolurin *et al.*[29] was in Osun South west Nigeria. However, the dietary history of the children was not ascertained in both studies.

Earlier in 2004, Maziya-Dixon *et al.*[16] reported a slightly lower prevalence of 20% for serum zinc deficiency in a Nigerian food consumption and nutrition survey. An estimated global prevalence rate of 31% was obtained the same year.[5] These findings are comparable to those reported in the current study (26%). However, unlike the study carried out by Maziya-Dixon *et al.*[16] a larger population size was used in the current study and serum zinc levels were used to determine zinc deficiency state. On the other hand, the estimated global prevalence study pooled results from various populations (affluent and impoverished) with

Table 4: Relationship between age, socioeconomic class, and serum zinc levels in the study population

Variable	Statistics	Age	SEC
Zinc (µg/dl)	Correlation coefficient	0.185	0.208*
	P	0.065	0.038
	n	100	100

*Spearman's rho. SEC – Socioeconomic class

diverse cultures and feeding practices.[5] The study also used degrees of stunting in areas where serum zinc levels and zinc contents in various diets were not available to arrive at their results.[16] This may explain the relatively higher prevalence of 31% reported in the estimated global prevalence study compared to the 26 % reported in this study. Similarly, slightly higher prevalence of serum zinc level of 32% compared to what was found in this current study was reported in a more recent study by Afolabi *et al*.[8] The reasons for this could probably be due to the similarities in the age groups, sample size and the method of serum zinc analysis between this current study and that by Afolabi *et al*.[8]

There was no significant difference in serum zinc level based on gender and there was no relationship observed between serum zinc levels and age of the participants. No study to the best of my knowledge has compared zinc levels between male and female children. Similar comparisons among the older population showed that males had significantly higher serum zinc levels than females and that the younger population (male and female) had higher serum zinc levels than the older population (male and female).[32] The reasons for these differences are unclear but could be related to the age difference between the participants in the current study and that by Rea.[32] Further studies are needed to further explain this variation in serum zinc levels between males and females among children and the older age groups.

SEC had a significant relationship with serum zinc deficiency in the current study. Several studies show an association indicators of socioeconomic status (SES) and micronutrient intake although most of these studies are in adults and adolescents.[33-36] Similar findings were reported in 2014 by Yarhere *et al.*[37] in Port Harcourt Nigeria and Anyabolu *et al.*[38] in South Western Nigeria among children living with HIV. Children of low SEC have been found to have less access to food rich in zinc and consume diets rich in phytates thereby decreasing their serum zinc levels.[27]

Conclusion

The zinc levels are of children in Nigeria is unacceptably low and consistent with findings from other studies thus making a case for routine supplementation.

Recommendation

Routine zinc supplementation may be necessary among apparently healthy children aged 5–60 months in South Eastern Nigeria.

Limitations

The sample size appears small. Larger studies may be needed to further elucidate these findings

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Conflicts of interest

There are no conflicts of interest.

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