Zinc Deficiency among Adolescence in Stunting Locus Area, Lombok, Indonesia

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ABSTRACT

Adolescence is at risk for zinc deficiency due to their high acceleration of growth, with bone mineral deposition reaching 80-90% by the adolescent phase. In zinc deficiency, adjustments in zinc pools were made, including the release of zinc retention in bones. Severe zinc deficiency could lead to growth retardation. This study aimed to determine the occurrence of zinc deficiency in the early adolescent stage (10 - 12 years) in the stunting locus area, Lombok. In the first stage, a cross-sectional design was conducted among children aged 10 - 12 years in Mataram City and East Lombok. In the second stage, subjects were assigned to the stunting and non-stunting group with a case-control design. Zinc intake assessment of 3-day 24 hours recall and concentration of zinc serum were used to determine zinc status. Approximately 34% of the subjects were stunting (n=774). Further analysis showed that 100% of the subjects (n=207) had a low zinc intake, with the average zinc intake below 45% of the recommended dietary allowance for zinc. The occurrence of zinc deficiency according to zinc serum concentration was only 2% of subjects. No significant difference in zinc intake and zinc serum was found between the stunted and non-stunted groups. Low zinc intake was observed among children in stunting locus districts. However, the proportion of zinc deficiency based on zinc serum concentration was very small. Zinc serum concentration may not capture zinc deficiency in the population, especially the mild or moderate ones.

Keywords: Zinc Deficiency, Zinc Intake, Zinc Serum, Stunted

1. INTRODUCTION

Zinc is required in almost all biological systems and plays vital roles in the catalytic process, structural and regulatory function. More than 300 enzymes in the human body require zinc for their catalytic function [1]. Zinc also facilitates the structural integrity of approximately 2500 transcription factors, counting for 8% of the human genome. Through its structural component (zinc fingers), zinc involves in signal transduction, cell differentiation, adhesion and transcription [1,2]. As a regulatory function, zinc-dependent promoters stimulate translocation and transcription of numerous genes [3]. Given the diverse function of zinc, it is not surprising that depletion of zinc may affect physical growth, reproductive maturity, immunological competence and gastrointestinal function, such as turnover of gastrointestinal mucosa [4,5].

Zinc depletion may arise from low zinc intake, poor bioavailability and increased zinc losses. Substances such as phytate in rice, cereal, bean and fibre could influence zinc absorption [6]. In an agricultural country such as Indonesia, rice and legumes are the most common staple food and dominate the food variety in a household. It is estimated that 71.2% population in the Southeast Asia region are at risk for low zinc consumption [7]. Twenty-three out of 25 low-middle income countries exhibited a prevalence of zinc deficiency of more than 20% [8].

Zinc deficiency is common not only in women and children but also in adolescence. Adolescence is at risk for zinc deficiency due to several factors, i.e. 1) they are at greater demand for energy, protein, vitamin and mineral to fulfil the increase of physical growth rate and development; 2) stunted adolescent are likely to become a short stature mother that might affect their reproductive development and their offsprings; and 3) the change of lifestyle and food habits of adolescents. In addition, adolescence is at its highest growth rate. During this period, bone mineral deposition is at its peak [9]. It reaches 80-90% by the end of the adolescent phase [10].

Zinc homeostasis is a highly regulated and very complex system. Alteration in one compartment causes

a cascade of homeostasis responses. Based on kinetic studies, muscle and bone were some of the main zinc pools in the human body. In zinc deficiency, adjustments in zinc pools were made, including the release of zinc retention in bones [11]. The present study aims to determine the occurrence of zinc deficiency among children in the early adolescent period (age 10 – 12 years) in stunting locus areas. According to demographic statistics in West Nusa Tenggara 2017, 1 in 3 children aged 5-12 years old was stunted [12]. The concentration of zinc serum, daily zinc consumption and anthropometry were assessed to estimate the prevalence of zinc deficiency.

2. METHOD

2.1 Study Design

The study consisted of 2 stage research. In the first stage, a cross-sectional design was conducted among children aged 10 - 12 years in Mataram City and East Lombok to identify the prevalence of stunted children. In the second stage, randomised subjects were assigned to the stunted and non-stunted groups with a case-control design. They were assessed for daily zinc intake and concentration of zinc serum.

2.2 Subjects

The study was carried out in Mataram city (Mataram District) and Dasan Lekong (East Lombok), West Nusa Tenggara, Indonesia. All children in grade 5 and grade 6 from the selected school were included in the study. The elementary schools were selected by random cluster sampling. Inclusion criteria were: 1) registered as students in the selected school; 2) age 10 – 12 years during data collection; 3) agree to participate in this study.

Sample size calculation indicated that 746 children for the first stage and 100 subjects in each group (stunting and non-stunting groups) for the second stage should be included in the study. This calculation was based on the population of school children grade 5 and 6 at elementary school in Mataram and East Lombok (49,500), 37% proportion of stunted children in Indonesia from the previous study, 80% power, and type 1 error rate of 5%. Informed consent was obtained from all subjects (the parents), and ethical approval was given by the ethical committee of the Faculty of Medicine, University of Mataram (Ethical approval number 1846/H18.8/TU/2010).

2.3 Data Collection

Zinc serum was analysed in SEAMEO-Tropical Medicine, University of Indonesia, Jakarta, using the *Flame Atomic Absorption Spectrophotometry* (AAS) method. Zinc serum concentration < 10.7 umol/L (> 70 µg/dL) for boys more than 10 years old and < 10.1 umol/L (> 66 µg/d)L for girls more than 10 years old

was categorised low [13,14]. Blood collections were conducted by certified laboratory analysts.

Daily zinc intake was assessed with 3-day 24-hour food recalls consecutively by trained nutritional enumerators. Recommended daily intake for zinc in adolescence aged 10-12 years old follows the recommendation given by Indonesian Health Ministry [15].

Bodyweight and height were measured by a body scale with an accuracy of 0.1 kg and a microtoise stadiometer with an accuracy of 0.1 cm. Anthropometry measurements were conducted by trained health enumerators. Nutritional status was assessed by Z score based on WHO MGRS data using WHO Anthro-plus software.

2.4 Study Procedure

In the first stage, subjects were measured for their height to identify the prevalence of stunting. Additional data such as socio-economic background were also taken into account. Then, subjects were systematically randomised again to enter the second stage. In the second stage, randomised subjects were assigned to the stunted and non-stunted groups. Data on daily zinc intake and zinc serum were collected.

2.5 Statistical Analysis

Data were expressed as mean±SD (Z-score, zinc serum, age) and proportion (stunted, wasting, zinc deficiency). Differences between groups were analysed by an independent T-test. Data that were not normally distributed were log-transformed. Correlation between variables was assessed by the Pearson correlation method and presented in scatter plots.

3. RESULT

3.1 First Stage Study

3.1.1 Subject Characteristics

In the first stage of research, 841 students were screened for their eligibility, and 774 students participated in the study, which consisted of 377 (48.7%) boys and 397 (51.3%) girls. Sixty-seven students were not included in the study due to unwillingness to participate in the study and not being present on the data collection day. Of 774 subjects, 361 (46.6%) students lived in the rural area, and 413 (53.4%) lived in the urban area. Subjects' socio-economic backgrounds are illustrated in Table 1. Most parents had educational backgrounds until elementary level, 64.8% of fathers worked in the informal sector, and 41.4% of mothers were unemployed.

Tab1e 1. Subjects' Socio-Economic Characteristics

Characteristics	Frequency (n)	Proportion (%)
Educational level		

Fathers: • Elementary school	313	50.0
Mothers: • Elementary school	383	58.0
Occupation Fathers: • Agriculture • Casual labour • Employee	256 83 258	34.9 11.3 35.2
Mothers: • Unemployed • Agriculture • Casual labour • Employee	304 283 8 104	41.4 38.6 1.1 14.2

3.1.2 Prevalence of Stunted Children

As shown in Table 2, 33.6% of subjects were stunted, and 20.2% of subjects were classified as wasting. The average body mass index (BMI) was 16.3±3.0 (mean±SD) with a negative height-for-age Z score (HAZ) and BMI-for-age Z score.

Table 2. Anthropometry Variables of Subjects

Parameters	Value
Mean±SD • Age (years) • Weight (kg) • Height (cm) • HAZ • BMI • BAZ	 11.2±0.7 30.5±8.5 135.9±8.2 -1.4±1.2 16.3±3.0 -0.8±1.4

Proportion (%)(frequency)
• Stunted (HAZ < -2 SD)
  - Boys
  - Girls
• Wasting (BAZ < -2 SD)
  - Boys
  - Girls
	
	33.6 (260)
	31.3 (118)
	35.8 (142)
	20.2 (156)
	23.1 (87)
	17.4 (69)

HAZ: Z score height-for-age; BMI: body mass index; BAZ:Z score BMI-for-age.

3.2 Second Stage Study

3.2.1 Subject characteristics

In the second stage of research, 207 students participated in the study: 100 subjects were assigned to the stunted group and 107 subjects to the normal height group. Again, age stratification was conducted due to the potential of bias in age between stunted and non-stunted groups (p=0.003). It is shown that children in the stunted group had less body weight and was thinner than their non-stunted counterparts, as indicated by significantly lower mean of body weight and Z score for BMI-for-age (Table 3).

Table 3. Subject Characteristics in The Second Stage of Research

Characteristics	Stunted Group (mean±SD)	Non-Stunted Group (mean±SD)
Age (years)	11.4±0.1	11.2±0.1
Height (cm)	129.2±0.5*	138.6±0.6*
Body weight (kg)	25.4±0.4*	32.8±0.8*
BMI • 10 - <11 years • 11 - <12 years • 12 - <13 years	15.1±0.2* 15.1±1.7 15.0±1.9 15.3±1.5	16.9±0.3* 16.2±2.8 17.4±3.5 17.1±2.4
HAZ • 10 - <11 years • 11 - <12 years • 12 - <13 years	-2.6±0.1* -2.5±0.4 -2.6±0.5 -2.7±0.6	-1±0.1* -0.8±0.8 -1.1±0.9 -1.3±0.6
BAZ • 10 - <11 years • 11 - <12 years • 12 - <13 years	-1.4±0.1* -1.1±1.0 -1.6±1.2 -1.6±0.9	-0.5±0.1* -0.6±1.2 -0.3±1.5 -0.6±1.0

3.2.2 Zinc Intake

Zinc intake was assessed with three days of 24-hour food recalls conducted consecutively by trained nutritional enumerators. The mean zinc intake of subjects from the stunted group was significantly lower than those in the non-stunted group (5.3±1.7 mg/day versus 5.9±1.8 mg/day) (Table 4). After age stratification, a significant difference in zinc intake between groups was found in the age group 10 - < 11 years. In the stunted group, both genders consumed less zinc intake compared to the non-stunted group (5.3±1.8 mg/day in stunted boys and 5.2±1.6 in stunted girls versus 6.0±1.8 mg/day in non-stunted boys and 5.8±1.8 mg/day in non-stunted girls). A similar pattern was found in the energy intake. Boys and girls in the stunted group had lower energy intake compared to those in the non-stunted group (1499.3±393.2 kcal/day and 1403.8±305.0 kcal/day versus 1537.9±309.0 kcal/day and 1486.0±297.9 kcal/day), although it was not statistically significant. Given that zinc intake was also influenced by energy intake, zinc intake was adjusted for energy intake. No significant difference in zinc intake between groups was found in all age groups.

Table 4. Daily Zinc Intake in Stunted (n=100) and Normal Groups (n=107)

Characteristics	Stunted Group (mean±SD) (median)	Non-Stunted Group (mean±SD) (median)
Daily zinc intake (mg/day) • 10 - <11 years • 11 - <12 years • 12 - <13 years	5.3±1.7 (4.9)* 5.0±1.7* 5.6±1.8 5.3±1.6	5.9±1.8 (5.6)* 5.8±1.6* 6.0±1.9 5.5±2.0
Daily animal protein (g/day	21.0±1.2 (17.0)	22.4±1.0 (21.6)

Phytate zinc ratio	16.5:1 (16.4:1)	15.6:1 (14.7:1)
Daily energy intake (kcal/day	1438.2±340.6 (1371.6)	1507.4±302.2 (1454.0)
Zinc bioavailability (proportion/%)		
• Low (15%)	62.0	47.7
• Medium (30-35%)	7.0	12.1
Proportion of zinc RDA (%)	40.6*	44.5*
Proportion of energy EAR (2050 kcal/day) (%)	70.2	73.5
Proportion of protein RDA (50 g/day) (%)	84.3	89.5
Absorbed zinc (mg/day)	1.5±1.2*	1.9±1.2*

*Significantly different ($p<0.05$). RDA: recommended dietary allowance; EAR: estimated average requirement.

All subjects in both groups had zinc intake below the recommended allowance for zinc. The mean intake for zinc, protein and energy in both groups are low compared to the Indonesian Recommended Dietary Allowance. Although the energy intake was lower in the stunted group, no significant difference was found between groups. Consumption of animal protein as an enhancer for zinc absorption was significantly different between groups in the age group 10 - <11 years. The Phytate zinc ratio was comparable between groups. Approximately 62% of subjects in the stunted group and 47.7% of subjects in the non-stunted group consumed low bioavailability of zinc.

As shown in Figure 1, the correlation coefficient between height-for-age Z score and zinc intake, height-for-age Z score and energy intake, and height-for-age Z score and protein intake were 0.2 ($p=0.03$), 0.16 ($p=0.02$) and 0.2 ($p=0.005$), respectively. This indicated that there was an increasing trend in zinc intake, energy intake and protein intake, along with the increase of Z score height-for-age.
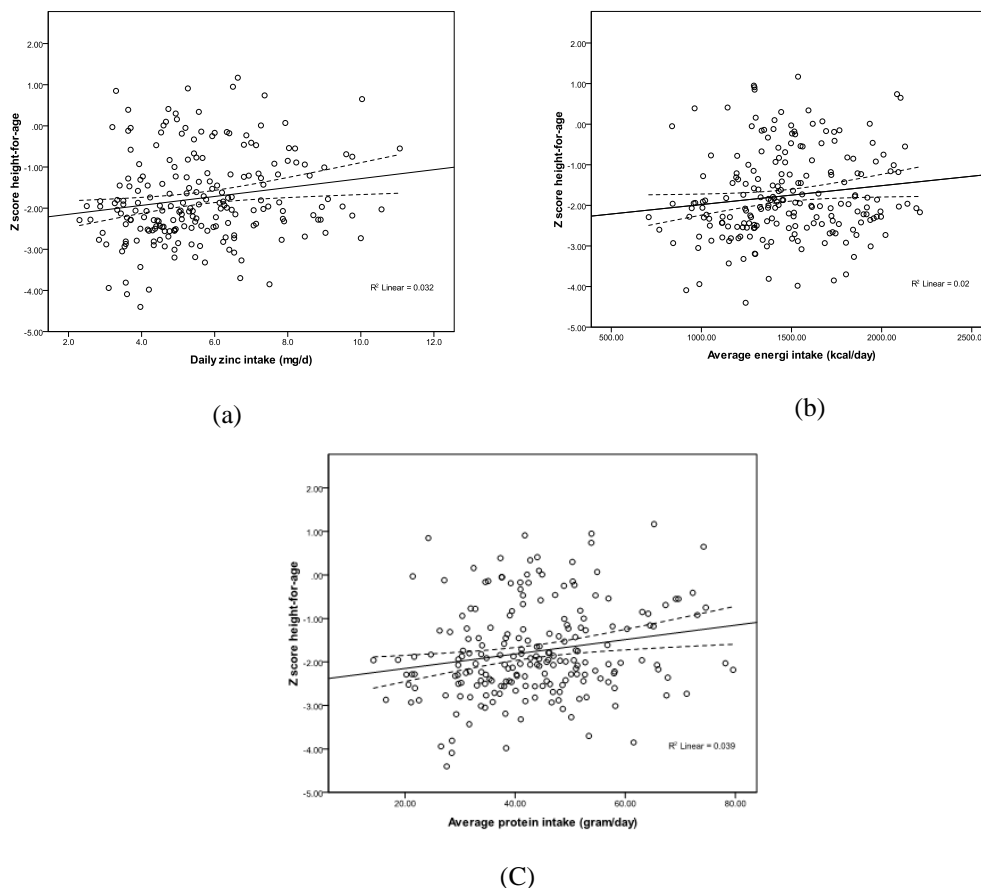

(a)

(b)

(C)

Figure 1 Scatter plot graphic of height-for-age Z score, zinc intake, energy intake and protein intake among subjects (n=207) participated in the second stage of research. (a) Scatter plot between height-for-age Z score and daily zinc intake; (b) Scatter plot between height-for-age Z score and daily energy intake; (c) Scatter plot between height-for-age Z score and daily protein intake (line: mean; dashed line: 95% confidence interval)

3.2.3 Zinc Serum

In this study, non-fasting zinc serum concentration was used as a biomarker for screening zinc deficiency. Two per cent of the subjects had low zinc serum concentrations. No significant difference in zinc serum concentration was found between the stunted and non-stunted groups (13.6 ± 1.9 umol/L versus 14.0 ± 2.3 umol/L). Those values were above the cut-off point for zinc deficiency (age > ten years: boys < 10.7 umol/L dan girls < 10.1 umol/L) [14]. Male children in both groups had a lower mean of zinc serum concentration than the female students.

3.2.4 Screening for Zinc Deficiency

Given no available gold standard for screening zinc deficiency, the present study used three indicators for assessing zinc deficiency: zinc intake, zinc serum concentration, and the presence of stunting as a functional indicator. Based on zinc intake, 100% of subjects in the stunted and non-stunted groups were at risk for zinc inadequacy, with the average proportion of zinc intake less than 40.6% and 44.5% of zinc RDA, respectively. As for zinc serum concentration, it was found that only 2% of subjects in the stunted group and 2% of subjects in the non-stunted group had low zinc serum concentration. No significant difference was found in zinc serum concentration between groups.

4. DISCUSSION

The present study showed that 1 in 3 subjects was stunted. Subjects were assigned to the stunted and non-stunted groups, and then they were observed retrospectively for their daily zinc intake and serum zinc concentration. The results showed that 100% of subjects in both groups consumed zinc less than the recommended daily intake. The daily mean intake of zinc was significantly lower in the stunted group.

Zinc intake in the present finding was higher than the study conducted by Indriastuti Y.A. (2005). They found that daily zinc intake among students aged 10 – 12 years in Tangerang district, Indonesia, was 24.8% of zinc RDA [16]. Their subjects also exhibited anaemia. Zinc and iron tend to be available in the same food product. Besides that, both minerals' absorption was also influenced by similar food substrates such as animal protein and phytate content [5]. The phytate zinc ratio in the present study was 16.5:1 and 15.6:1 in the stunted and non-stunted groups, respectively. Phytate or myo-inositol hexaphosphate is a potent chelator of minerals, including zinc, mainly in seeds of cereal grains, nuts, and legumes. Since phytate cannot be digested and absorbed in the human intestinal tract, zinc bound to phytate would pass through the intestinal lumen unabsorbed [5]. When the phytate zinc ratio is 15, zinc absorption decreases by almost 50% resulting in a negative zinc balance [17].

In the present study, only 2% of subjects had zinc serum concentration below normal. This finding is contradicted with the average daily zinc intake level of the subjects, which was less than 45% of zinc RDA. However, if the predicted zinc absorbed in the present study is taken into account (1.7 ± 1.3 mg/day), it is above the estimated physiologic requirement for adolescent females and males, 1.26 mg/day for and 1.4 for boys, respectively [18]. This implies that zinc intake in the present study can meet the daily physiological requirements sufficiently. Another possibility is that zinc serum concentration might have a limitation in identifying mild to moderate zinc deficiency [1]. In kinetic studies, muscle and bone were some of the significant zinc pools in the human body. However, during zinc deficiency, zinc retention in the skeletal system was released into the plasma compartment in order to supply zinc for other main organs [11]. Therefore, it is possible that the zinc serum/plasma assessment may give a normal result. Further study is warranted.

Serum zinc concentration in the stunted and the non-stunted groups was 13.6 ± 1.9 umol/L versus 14.0 ± 2.3 umol/L. No significant difference was found between the stunted and non-stunted groups. This was above the cut off point for zinc deficiency which was < 10.7 umol/L for boys dan < 10.1 umol/L for girls (14). In relation to gender, the present study found that male children had lower zinc serum concentrations than the females in both stunting and non-stunting groups. Several studies have reported that males are more vulnerable than females [19,20]. They argued that boys had higher requirements because they had a higher growth rate and a greater proportion of muscle per kilogram body weight. Since zinc is primarily located in fat-free mass tissue, bone and skeletal muscle, therefore it is likely that zinc in the plasma compartment is mobilised to the main zinc pool to fulfil the zinc requirement for a higher growth rate in boys [5].

In the present study, children in the stunted group had less body weight and were thinner than their non-stunted counterparts, as indicated by a significantly lower mean of body weight and Z score for BMI-for-age. The mean energy requirements in the stunted group were significantly lower than those in the non-stunted group. Even the daily energy requirement in the non-stunted group was below the FAO/WHO/UNU total energy requirement [21]. Many studies have shown that stunting commonly occurs in young children; however, there has been growing evidence that the growth in school-age children could not catch up or even remain stable. Instead, their growth progression declines further from the international growth standard [22]. As shown in Table 3, subjects in both groups experienced a progressive decline in height-for-age Z score. If this condition continued, they might continue to falter and become short-statured

adults. Growth faltering is a significant public health concern because attained height has important implications for adult work performance [23] and reproductive outcomes [24,25].

In the present study, chronic malnutrition is likely responsible for stunting among these subjects instead of genetic factors. Amigo et al. (2001) conducted a study to identify growth deficit among primary school-age children in the district of Santiago, Chile, whose parents were of short stature compared with those of not short stature [26]. That study found that the most critical factors for short stature children whose parents had very low stature were a history of malnutrition (OR 5.26; 95% CI 2.68 – 10.34) and short length at birth (OR 4.87; 95% CI 2.18 – 10.92), as for the crucial factors for short stature children whose parents had normal stature were a history of malnutrition (OR 4.58; 95% CI 2.20 – 9.53) and unhygienic housing condition (OR 4.29; 95% CI 1.76 – 10.48).

There are several limitations to this study that should be addressed. First, the utility of zinc serum concentration in assessing zinc status could be compromised by several factors, including time of previous meal consumed, the time gap between blood collection and centrifugation, the presence of systemic inflammation and administration of supplement or other medication. The present study did not control for time of previous meal consumed, systemic inflammation and history of medication. Second, the children gave the information of daily zinc intake only, instead of cross-checking with the mothers. The argument for this is that children of age 10 – 12 years old usually choose their own food and can memorise them. In addition, the interview was conducted by a trained nutritional surveyor aided with a food model and zinc intake was assessed with 24-hour food recalls three days consecutively. Before data collection, the research team had surveyed and documented food and snacks sold in the school cafeteria. Therefore, the chance for random error could be minimised.

In conclusion, the present study revealed that low zinc intake was observed among children in stunting locus districts. However, the proportion of zinc deficiency based on zinc serum concentration was very small. This discrepancy raised a question on the usage of zinc serum concentration, especially in screening for mild zinc deficiency in the population. Further research is needed to identify mild zinc deficiency with the larger sample size and take into account other confounding variables such as anaemia, infection status and other nutrient intake related to zinc absorption.

5. CONCLUSION

The present study revealed that low zinc intake was observed among children in stunting locus districts. However, the proportion of zinc deficiency based on zinc serum concentration was very small. This discrepancy raised a question on the usage of zinc serum concentration, especially in screening for mild zinc deficiency in the population. Further research is needed to identify mild zinc deficiency with the larger sample size and take into account other confounding variables such as anaemia, infection status and other nutrient intake related to zinc absorption.

AUTHORS' CONTRIBUTIONS

The authors' responsibilities were as follows: DI – designed the research, data collection, data analysis and drafting the manuscript; AE – designed the research and data collection; LN – data collection. All authors reviewed the manuscript and approved the final manuscript.

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