

The Effect of Red Bean Cookies' Administration the Blood Calcium and Zinc Levels of Stunting Children Hair

Bernike Doloksaribu¹, Efendi S. Nainggolan¹, Rumida¹, Ginta Siahaan^{1*}

Lecturer at the Politeknik kesehatan kementrian kesehatan medan, Department of Nutrition¹

Corresponding Author: 1*



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ABSTRACT

Stunting is a growth disorder indicator of nutritional deficiency over a long time. Low intake of protein, calcium, and zinc are identified by calcium and zinc in hair. Calcium and zinc minerals have an essential role in growth and development during the first 1000 days of life. One factor that affects stunting is the low intake of bone-forming nutrients such as calcium and zinc consumed, causing the calcium content in the blood to decrease and children's hair growth. Calcium and zinc minerals have an essential role in human growth. This study aims to determine the effect of red bean cookies on blood serum calcium and zinc levels in toddler's hair (13-36 months) who experienced stunting in the work area of Pantai Labu Community Health Center. This study was a Quasi Experiment with pre- and post- test design. The population were stunted toddlers in 13 - 36 months, divided in two groups, with control and treatment of 26 persons in each group. The administration administration of cookies with and without red beans is provided every day, as many as five pieces (20 gr/piece) weighing 100 gr for 12 weeks. There were differences in the administration of cookies in the control and treatment group by independent t-test method of blood serum calcium with $p=0.027$. While for biomarkers, hair zinc level was $p=0.001$. Cookies with the addition of red beans are able to increase blood serum calcium and hair zinc levels in stunted toddlers.



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1. Introduction

Stunting is a chronic problem of nutrition for toddlers. This situation is influenced by the nutritional status of the pregnant mother, infection disease in the toddler period, and intake of nutrition. [1]. About a quarter of toddlers of 5 worldwide are stunted [2]. Stunting were identified when children measured based on body height/age categorized according to WHO Antroplus 2005, if the z-score of body height/age < -2 SD.

The World Health Organization (WHO) has set the prevalence rate of stunting events $\leq 20\%$, while the stunting incidence rate based on Riskesdas' 2018 in Indonesia was 30.8%, there was 32.14% case in North Sumatra province of Deli Serdang regency which is a district in North Sumatra and where the study was

conducted which reported at 27.44%. This data showed that the prevalence of stunting in Indonesia at the provincial and district levels is still above the prevalence rate established by WHO [3].

The occurrence of stunting in toddlers, can start from the first 1000 days of life. Insufficient supply of nutrients in the first 1000 days of life can be caused by low intake of macro-and micronutrients, especially minerals, calcium, and zinc, which are needed for growth and development [4].

Calcium is a bone formation for the matrix formation of bone tissue. Deficiency of calcium can inhibit cell division, repair of tissue and may effect weight and height [5]. Serum Calcium should be maintained for the function as a regulator of cell processes, bone mineralization, muscle contraction, enzyme activity, and internal cell hormone [6], [7].

Zinc is an essential trace element that functions in the synthesis, secretion, and action of growth hormone, in this case is Insulin Growth Factor 1 (IGF-1) [8]. The presence of zinc in the body can be measured based on hair zinc levels, because some findings stated that zinc concentration in the hair is assumed to be a low intake of zinc consumed [9]. Hair zinc can also provide information about zinc mineral metabolism in cells that reflect mineral accumulation from zinc intake over 2-3 months [10].

To fulfill the needs of nutrients that are important in the growth and development of toddler, it is necessary to add foodstuffs that contain essential nutrients to snack foods, other foods, and daily consumed foods. Cookies are one type of biscuit that is loved by the public because it tastes crispy, savory, and suitable as a snack that contributes to meeting daily nutritional needs [11].

Cookies with the addition of red bean flour are popular in all circles from children to adults [12]. Red bean cookies help overcome the problems of stunting, osteoporosis, and infectious diseases because they contain nutrients such as protein, calcium, zinc, phosphorus, and iron. In 100 grams of red bean cookies that have been checked in the laboratory, FMIPA UNIBRAW obtained a carbohydrate content of 28 gr, fat 24.15 gr, protein 10.3 grams, zinc 69.24 mg, calcium 39.15 mg and energy 172 kcal. [15] gave the same condition in the form of cookies for 30 days, but made from soy beans was able to increase the level of albumin and blood hemoglobin of pregnant women needed for fetal growth and development. This illustrates that cookies are preferred and able to help the first 1000 days of life regulatory process run well, if this first 1000 days of life period is problematic, it can result in children born stunted. This study aims to determine the effect of red bean cookies administration on blood serum calcium levels and zinc levels in hair in toddlers (13-36 months) who are stunted in the work area of the Pantai Labu community Health Center.

2. Materials and Methods

The design of this study was Quasi Experiment with a pre-and post test and 26 persons in each group. The control group was in Sentang Dome and the treatment group was in Durian Village. This study was held in Pantai Labu Community Health Centre's work area. In Durian and Kuba Sentang village were found stunted toddlers 28,9% and 29,01%. These data show that the two villages have stunting prevalence of Deli Serdang 27.44%.

The sample magnitude was determined by the formula (Rachmat, 2016):

$$n1 = n2 = \left(\frac{Z\alpha^2 + Z\beta}{X1 - X} \right)^2$$

$$= 25,6 \sim 26 \text{ people}$$

Reason for choosing the determination of the sample, the formula above is based on a paired numerical test.

The provision of cookies with red beans is given to stunted children in Durian Village as much as five pieces (100 gr / day), on the grounds that red bean cookies contribute 50% of the RDA, while calcium contributes 6% of the RDA and zinc minerals exceed the needs of the RDA [13]. Cookies are given for 12 weeks because the process of accumulation of hair zinc from the results of metabolic zinc intake occurs for 2-3 months [9]. Meanwhile, feeding high calcium for 12 weeks can increase blood serum calcium so that bone mineralization occurs [14]. The provision of cookies with and without red beans began in early July 1 to September 30, 2018. Meanwhile, the control group in Cuban and Sentang villages was given cookies without adding red beans with a nutrient protection of carbohydrate 20gr, fat, 7.25 gr, protein, gr, zinc 7.12 mg, calcium 15.07 mg, energy 160 kc. A total of five pieces (100 gr/day) for 12 weeks. Snack consumption is controlled by researchers and directly supervised by health cadres who have been briefed by researchers, so it is certain that the cookies given are consumed by stunted toddlers. To obtain the effectiveness of giving cookies to stunted children in the treatment and control group, the authors recalled 24 hours for 2 days before and after the giving of cookies.

Data on stunting toddlers were first screened by taking body height measurements using microtoic measuring instruments with an accuracy of 0.1 cm and then compared with age so that the number of stunted children was obtained as many as 26 people in each village. The collected data includes data on average age, blood calcium levels, and zinc levels in the hair. The data that has been collected is then processed manually through the stages of the process, which begins with editing, coding, cleaning, tabulating the data. Then it is entered and processed with the help of computer programs.

Blood calcium level examination were carried out by taking the blood of the left arm of the toddler as much as 2-2.5 ml. Blood collection were carried out by health analysts who have been referred to by researchers. Furthermore, it was examined through 3 procedures including taking venous blood, making serum, and calcium examination using the Autoanalyzer Architect Plus 8200 tool, which was carried out at the Paramitha Medan laboratory.

The hair zinc examination were carried out at the Chemical Laboratory of FMIPA Universitas Brawijaya using the Anatomic Absorbance Spectrophotometry (AAS) method which is to weigh 50 mg of hair with analytical scales and then put it into a plastic tube that has been washed and labeled with a number and date and then added 15 ml triton X 1% to the sampel then in a mixer using a mechanical mixer for 30 minutes then pour it into a plastic filter rinse by deionized by means of sprayed \pm 100 cc. Then dried in the oven with a temperature of 110o C for 12 hours. Transfer the original bottle according to the label and number. The lid uses parafilm and is ready to be checked on the Atomic Absorbance Spectrophotometer (AAS) with the brand shimadzu / AA-6200 [15].

Analysis data of univariate and bivariate data normality test was carried out using t-dependent test. In this study, all data were distributed normally and the test carried out was a t-dependent test to see changes in each group, and then continued t- independent to see the differences between the groups. Furthermore, the results of the blood serum calcium and hair zinc examiners were categorized by comparing with normal

values of blood serum calcium and hair zinc. This categorization is intended to see changes in blood serum calcium and hair zinc towards a normal direction. This research has received Ethics Eligibility approval from the Ethics Committee No: 036/KEPK/POLTEKKES KEMENKES MEDAN/2017.

3. Results

3.1 Sample Characteristics.

Table 1. Frequency Distribution of Sample Characteristics.

Sample Characteristics	Treatment		Control	
	n	%	n	%
Gender				
- Male	16	61,5	17	65,4
- Female	10	38,5	9	34,6
Age (months)				
13 - 24 months	14	53,8	9	34,6
25 - 36 months	12	46,2	17	65,4

Table 1 explained that most samples in the intervention and comparison groups were male, 61.5% and 65.4%, respectively. Meanwhile, based on age, in the intervention group most were aged 13-24 months (53.8%) and in the comparison group most were aged 25-36 months.

3.2 Nutrients intake before and after giving cookies with and without red beans.

Table 2, Distribution of nutrient intake before and after giving cookies with and without red beans by the 24-hour recall method

Indicator	RDA (2019)	Treatment Group						P-Value
		Before			After			
		Mean	SD	% RDA	Mean	SD	%RDA	
Energy (kcal)	1350	1128	2.19	83.6	1185	2.04	87.8	0.085
Carbohydrate(g)	215	201	10.7	93.5	210	9.63	97.7	0.124
Protein (g)	20	11.7	3.52	58.5	12.3	3.3	61.5	0.067
Fat (g)	45	42.4	4.19	93.3	43.2	4.57	96.0	0.124
Zinc (mg)	3	0.91	0.15	30.3	1.05	0.37	35.0	0.078
Vit C (mg)	40	26.3	2.15	65.0	27.1	3.09	67.8	0.192
Fe (mg)	7	3.5	1.08	42.9	3.9	2.16	57.1	0.075
Calsium (mg)	650	378	2.01	58.2	415	1.82	63.8	0.120
Indicator	RDA (2019)	Control Group						P-Value
		Before			After			
		Mean	SD	%RDA	Mean	SD	%RDA	
Energy (kcal)	1350	1197	4.7	88.7	1231	5.02	91.2	0.072
Carbohydrate (g)	215	195	7.34	90.7	204	6.58	94.9	0.059
Protein (g)	20	10.2	2.61	51.0	10.9	2.73	54.5	0.078
Fat (g)	45	41.0	3.87	91.1	41.7	4.20	92.7	0.153
Zinc (mg)	3	0.89	0.07	29.7	0.92	0.37	30.7	0.092
Vitamin C (mg)	40	25.07	2.37	62.7	25.43	2.41	63.6	0.210
Fe (mg)	7	2.7	1.5	38.6	3.1	2.13	44.3	0.081
Calsium (mg)	650	3.53	1.7	54.3	382	1,98	58.8	0.112

From Table 2 showed that the recall results obtained for 2 consecutive days carried out before and after the administration of cookies with and without red beans, it can be seen that the data showed homogeneity, where $p > 0.05$ method used to see homogeneity in the results of the 24-hour recall, using t-dependent test. This table of recall results showed that there was no difference in nutrient intake before and after the intervention, so that the provision of cookies can be used as an indicator to see changes in blood calcium and zinc levels in the hair of stunted children.

3.3 Blood Calcium and Hair Zinc Levels.

Table 3. Distribution average values of blood calcium and hair zinc levels before and after intervention.

Variabel	Treatment		Control	
	Mean ± SD	p value	Mean ± SD	p value
blood calsium level (mg) before	7.72 ± 5.32		7.66 ± 4.91	
blood calsium level (mg) after	9.08 ± 3.54	0,001	8.68 ± 3.72	0,002
blood calsium level (mg) difference	1.36 ± 2.78		1.02 ± 10.69	
Hair zink (ppm) before	115.37 ± 127.47		112.76 ± 117.47	
Hair zink (ppm) after	144.07 ± 89.98	0,027	121.47 ± 99.08	0,058
Hair zink (ppm) difference	28.7 ± 1.62		8.71 ± 35.06	

Table 3 showed that there was a difference between serum calcium before and after the administration of cookies for both the control group and the treatment group using a t-dependent statistical test found $p < 0.05$. The difference in blood calcium for the control group was 1.02 mg, while for the treatment group it was 1.36 mg. Meanwhile, in hair zinc, there were differences before and after the administration of cookies for both the control group and the treatment group using a t-dependent statistical test found $p < 0.05$. The difference in hair zinc for the control group was 8.71 ppm, while for the treatment group it was 28.7 ppm.

3.4 The Difference between Blood Calcium Levels and Hair zinc Lenels.

Table 4. The Difference between Blood Calcium Levels and Hair Zinc Levels

Variabel	Treatment	Control	p value
	Mean ± SD	Mean ± SD	
Blood calsium level (mg) difference	1.36 ± 2.78	1.02 ± 10.69	0,027
Hair zink (ppm) difference	28.7 ± 1.62	8.71 ± 35.06	0,001

Table 4 showed that there were differences in the difference between blood serum calcium and hair zinc after intervention in the treatment and control groups. The statistical test method used to see the difference was t- independent and found $p < 0.05$.

Status Distribution of Blood Calcium Levels and Hair Zinc Levels. Tabel 5. Status Distribution of Blood Calcium Levels and Hair Zinc Levels

	Treatment
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Indicator	Before		After		p Value
	n	%	n	%	
Low Blood Calcium Levels	18	69	10	38	0,001
Normal Blood Calcium Levels	8	31	16	62	
Low Hair Zinc Levels	17	65	6	23	0,027
Normal Hair Zinc Levels	9	35	20	77	
Indicator	Control				p Value
	Before		After		
	n	%	n	%	
Low Blood Calcium Levels	20	77	13	50	0,027
Normal Blood Calcium Levels	6	23	13	50	
Low Hair Zinc Levels	16	62	14	54	0,001
Normal Hair Zinc Levels	10	38	12	46	

Table 5 showed that the administration of cookies with and without red beans for 12 weeks saw changes based on normal categories, where the percentage change in blood calcium was greater in the treatment group, which was 62% while for the control group it was only 50%. As for the hair zinc indicators, the percentage change for the normal category in hair zinc was greater in the treatment group, which was 77%, while for the control group it was only 46%.

Table 5 also shows that red bean cookies are able to change the indicators of blood calcium and hair zinc towards normal to be greater.

4. Discussion

Sample Characteristics. In this study and carried out in the working area of the Pantai Labu Community Health Center, it includes Durian Village and Dome Sentang Village to see stunted toddlers. Based on the age classification in the treatment group, it was dominated by the age group of 13-24 months (53.8%), while the control group was dominated by the age group of 25-36 months (65.4%). The stunting toddler group in the treatment group is an age group that falls into the category of 1000 HPK, where the growth and development process during the 1000 HPK period must be really considered because the nutritional deficiencies that occur will cause stunted children, low Aq, psychometric development is disturbed [16].

The stunting toddlers in the treatment and control group were dominated by the male sex, for the treatment group it was 61.5%, while for the control group it was 65.4%. This is in line with the research conducted by [17], [18]. which found that the percentage of men who experience stunting is higher than women. However, But it is different from the research conducted by [10]. which found the percentage of men is smaller than women for children who are stunted.

Nutrient Intake Before and After Intervention. The effectiveness of giving cookies with and without red beans to stunted toddlers we first carried out data recall data collection using the method 24 hours before and after the intervention. It is intended to avoid bias due to the daily food consumption of toddlers. From the results of the 24-hour recall, the intake of nutrients (energy, carbohydrates, protein, fat, zinc, vitamin C, Fe, and calcium) using the t-dependent test, it turned out that the results did not differ markedly ($P>0.05$). These data show the homogeneity of the nutritional intake of toddlers before and after the intervention.

The Effect of Cookies With and Without Red Beans on Blood Calcium. The provision of cookies carried out for 12 weeks (3 months) turned out to have an impact on increasing blood serum calcium. Serum calcium in the blood where there was an increase from the average value before by 7.72 to 9.08 after administration in the treatment group. Likewise, the control group increased serum calcium in the blood from an average value of 7.66 to 8.68 after administration. The average blood serum calcium showed that both the control group and the average treatment of blood serum calcium in stunted children were in the normal range of 8.8-10.8 mg/dl. [19].

The results of statistical tests using t- dependent test showed significant differences in the control group (cookies without red beans) $p=0.002$ and the treatment group (cookies with the addition of red beans) $p=0.001$. When compared between the treatment and control groups after the administration of cookies with and without red beans, changes in blood calcium levels using the t- independent test and found a significant difference of $p: 0.027$.

Cookies are cookies made from wheat flour with the addition of skim milk and butter to the manufacture. In the treatment group, cookies were also added with red bean flour of 50 gr as a substitute for wheat flour. Calcium content will increase after the addition of red beans examination in the chemical laboratory of FMIPA Universitas Brawijaya and the addition of calcium content from 15.07 mg to 39.15 mg. The concentration of calcium in plasma, especially free calcium ions, is very carefully maintained in such a way for the transmission of neural implus and muscle concentration, as well as as a catalyst for various biologic reactions. The integrated hormonal system regulates calcium homeostasis through the process of controlling the transport of calcium in the intestines, kidneys, and bone. Serum calcium homeostasis develops to maintain calcium ion levels in the extracellular within the normal range by flowing calcium by reciprocal processes in and out of reserves in the bones, which can result in low mineralization of the new bone deposit matrix and osteoblast dysfunction [20].

Calcium deficiency that causes low blood serum will cause metabolic disorders including the regulatory system, inflammatory cytokines. This affects chondrocytes directly and has an impact on the bone formation process, because the absence of bone mineralization ultimately leads to stunting [21], [22]. The risk of stunting is 3.93 times greater in toddlers with low calcium intake [23]. Adequate calcium intake during the growth period will affect serum levels in the blood, which is a predictor of the growth and development of a toddler child who will affect the concentration of calcium in the blood to remain in a normal state (8,8-10,8 mg/dl). Calcium can also affect the work of growth hormone and can also affect the work of IGF1 so that the growth of toddlers becomes maximum [24].

This calcium intake can be obtained from the daily diet consumed such as milk, fish, green vegetables, as well as legumes. Food products that are made attractive in the form of cookies that are loved by toddlers, are added with various ingredients that are high in calcium content such as beans including green beans, peanuts, soybeans, and kidney beans which are high in calcium content, so that the calcium needs of toddlers can be met. Cookies can be used as an additional food for toddlers that can be consumed every

day [21], [25].

Several studies that used local foodstuffs made from soybeans to be used as biscuits / cookies [26], soybeans, and moringa leaves are used as biscuits/cookies [20], and using green beans as porridge [1] can increase calcium intake so that blood serum calcium also increases. These studies found that there was a change in height in stunted children by providing local foodstuffs. The increase in height will affect all stunted children who will avoid stunting. In this study, researchers also used local foodstuffs derived from beans (kidney beans), which are also high in calcium, easy to get, and affordable prices.

The Influence of Giving Cookies With and Without Red Beans Toward Hair Zinc Levels. The provision of cookies given every day is five pieces, where each piece weighs of 20 gr. Cookies were given for 12 weeks (3 months) in the control and treatment groups. Cookies have an impact on increasing zinc levels in the hair. Zinc levels in the hair where there was an increase from the average value before by 115.37 to 144.07 after administration in the treatment group. Likewise, the control group increased zinc levels in the hair from an average value of 112.76 to 121.47 after administration. However, the results of statistical tests using the t- dependent test showed that there was no difference before and after the provision of cookies in the control group, $p= 0.058$. Meanwhile, in the treatment group, there were significant differences before and after the provision of cookies with the addition of red beans, $p = 0.027$. When compared between the treatment and control groups after the administration of cookies with and without kidney beans, there was a change in hair zinc levels using the t-independent test found a significant difference $p: 0.001$.

Cookies without the addition of red beans contain 7.12 mg of zinc, while cookies with the addition of red beans contain 69.24 mg of zinc. Zinc in the hair is used to see the nutritional status in the body over a long period of time [10], [9], [27]. Zinc examination of the hair is also often used for children who have a risk at the time of blood draw to rebel accidentally due to the disorder of the disease they suffer from, such as children suffering from autism. In addition, hair samples are easier to obtain and more stable in metabolic processes [15]. Daily intake of zinc has contributed to varying levels of serum zinc. However, zinc concentrations remain constant in hair, skin, heart, and skeletal muscles [10]. The zinc metabolism of hair and its alternation in the brain is slower than in peripheral tissues. This suggests that zinc homeostasis in the hair is affected by chronic zinc deficiency [27], [28].

Red beans that have been used as flour and then substituted as much as 50 gr in making cookies make a significant contribution in increasing zinc levels in the hair of stunted toddlers. Zinc intake in the treatment group with red bean substitution contributed to the zinc levels of stunted children's hair. Hair zinc levels are a biomarker to determine the presence of zinc deficiency, where hair zinc will be taken as endogenous zinc to meet zinc needs [9]. analysis of zinc minerals in hair indicates the accumulation of zinc minerals over a long period of time when compared to blood serum zinc. The presence of zinc serum in the blood is heavily influenced by its accumulation by albumin, which helps the process of binding and transphore zinc [17]. Zinc is known to play a major role in regulating cellular activity by acting as a co-factor, stimulating the synthesis of proteins needed for matrix formation. Zinc can increase alkaline phosphate associated with DNA synthesis and the result stimulates bone growth needed for the growth and development of toddlers [29], [30].

This result is in accordance with Budiastuti's research in 2011 which provides zinc sulfate supplementation and biscuits, and it turns out that it can increase the hair zinc levels of stunting toddlers by giving for 3 months. Another study conducted by [15] on children with autism treatment local snacks containing zinc turned out to be able to change zinc levels in children with autism.

The advantages in this study used hair zinc measurements which turned out to be higher accuracy compared to blood serum zinc. The measurement parameters of the presence of zinc are rarely carried out on the hair, but it is easy to carry it out. The weakness in this study concerns the control of cookie consumption on holidays even though its implementation is assisted by health cadres. The location of the homes of stunted children who are used as samples is far from one another so that it requires more enumerator energy.

5. Conclusion

There is no effect of administration cookies with and without red beans on blood calcium dan, only administration cookies with red beans give an effect on hair zinc levels.

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