The benefit of rebon shrimp-based supplementary feeding on serum albumin level in children who have undergone stunting

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ABSTRACT

Stunting is still an unresolved global health problem caused by inadequate nutritional intake, significantly affecting a person's future development. Rebon-shrimp is high protein and inexpensive local food, but still underutilized. This quasi-experimental study aimed to determine the effect of supplementary feeding from Rebon-shrimp on serum albumin levels in stunting children aged 24-60 months. The intervention group (n=44) received rebon shrimp-based supplementary food for 90 days, while the control group (n=44) received a placebo. Measurement of serum albumin was carried out by the ELISA method using blood samples. The results showed a statistical difference (p<0.001) in serum albumin levels in the intervention group, while the control group did not differ statistically (p=0.363). The intervention group experienced an increase in albumin levels of 15.55 g/L, while the control group tended to experience a decrease in serum albumin levels of -1.92 g/L. There was no significant difference in serum albumin levels before the intervention in the two groups (p=0.180). Still, after the administration of rebon products, there was a significant difference in serum albumin levels between the two groups (p<0.001). Supplementary food made from rebon shrimp was beneficial for increasing the serum albumin level of stunting children.

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1. INTRODUCTION

Malnutrition, including stunting, is still an unresolved global health problem. Stunting has long-term effects on both individuals and societies. Short adult height and less than ideal function later in life are directly related to stunting. It is also an essential indicator of the early-life mechanisms at play in stunted growth and other undesirable outcomes. Stunting increases the chance of mortality and lowers productivity, learning ability, and adult and childhood health [1], [2].

Stunting-related dietary patterns result in significantly lower levels of circulating essential amino acids than do non-stunted children. These inadequate intakes of necessary amino acids may hinder growth by interfering with the growth regulatory pathway, which is incredibly sensitive to the availability of amino acids.

Lack of protein consumption impairs the function of growth regulatory mechanisms that control growth hormones and energy to control the development of the chondral plate, skeletal muscle, small intestinal cellular growth and differentiation, hematopoiesis, and iron metabolism. These problems, including as anemia, diminished cognition, environmental intestinal dysfunction, and immunity to infectious infections, are pertinent to childhood stunting and its related morbidities [3], [4]. The most objective and quantitative information on nutritional status is provided by biochemical tests. According to some theories, biochemical alterations are the earliest sign of nutritional deficiency, followed by cell or organ dysfunction, before clinical malnutrition is proven [5]. Albumin is one biochemical indicator of undernutrition in children. An important protein called albumin is present in the blood plasma that the liver produces. Low protein consumption, inadequate protein digestion, or inadequate protein absorption are the causes of the drop in plasma albumin [6]. According to research conducted in Nepal, children with stunted growth had considerably lower serum albumin levels than children with normal height [5].

Serum albumin is one of the markers of nutritional status. Albumin indicates the status of the protein in the body. Several studies have stated that serum albumin is an important prognostic factor for malnourished patients, especially hospitalized patients. Many malnourished hospitalized patients have low albumin levels, which can worsen the prognosis of their disease. Serum albumin is a nutritional index widely used to examine the population because it is easy to measure and is associated with the risk of mortality in various diseases. In hospitalized patients, low albumin levels are linked to an increased risk of morbidity and mortality. Malnourished children generally have a decrease in the synthesis and breakdown of total body protein. The body's natural defense system consists of proteins that can be broken down and bound to bacterial products. Circulating these proteins is essential for recognizing bacterial products by leukocytes that function for phagocytosis and killing bacteria. Malnourished children have an average decrease in total protein synthesis and an increase in the breakdown, which causes a decrease in albumin levels in the body. An increased risk of infection is linked to the body's decreased albumin levels [7].

Albumin is a serum protein reserve in the body produced by the liver. Typically, albumin constitutes 55% of all plasma proteins. A protein called albumin that is plentiful in plasma, is simple to detect, and is frequently used to assess nutritional status. Patients are categorized as normal with an albumin value of 3.5 g/dL, mild with 3.0-3.49 g/dL, moderate with 2.5-2.9 g/dL, and severe with 2.5 g/dL [8].

The nutritional intake of toddlers can be obtained from the family food menu and supplementary feeding. Supplementary Feeding is intended to help meet the needs of malnourished under-fives. Rebon shrimp, as local food in coastal areas, has good nutritional potential. Rebon shrimp has the potential to be an alternative source of animal protein, which is cheap and can be used as a natural protein for toddlers [9].

Fresh rebon shrimp contains 12.26% protein, 83.55% water, 0.6% fat, and 2.24% ash [10], while dried rebon shrimp contains 19.00% water, crude protein 48.29, ash 16.05%, and crude fat 3.62% [11]. The number of rebon shrimp is abundant, but the utilization of rebon shrimp still needs to be higher. Research on frozen food products made from rebon shrimp in 2022 showed that processed products made from rebon shrimp had good protein content to help meet children's daily nutritional needs [9]. This study aimed to determine the effect of supplementary feeding from rebon shrimp on serum albumin levels in stunting children aged 24-60 months.

2. METHOD

2.1. Study design

The research design used in this study was a quasi-experiment with control group design. It using pre-test and post-test as evaluation tools. All the sample in this study was stunting children (height-for-age z-score <-2 standard deviations). This study involved 88 stunting children, and it was divided into two groups, the intervention group (n=44) and the control group (n=44). The intervention group will receive rebon shrimp-based supplementary food, including nuggets, fish sticks, and fried otak-otak, with a serving size of three pieces/day (75 gr/day). The control group will receive a placebo made from low-protein flour. Both groups received the product for 90 days. Rebon-Based supplementary food products are frozen products made by researchers and have been tested for proximate and minerals tests to determine the product's nutritional content and portion sizes [9]. Rebon shrimp-based supplementary food was given every 30 days for 90 days (Days 0, 30, and 60).

2.2. Study location

This study was conducted from June to December 2021 in The Ma'rang District, Pangkep Regency, South Sulawesi Province, Indonesia. The location of this research was choosen because this area is one of the stunting loci areas in South Sulawesi Province, Indonesia. Ma'rang sub-district is a coastal area rich in marine resources, including rebon shrimp. However, the incidence of stunting in this district is still relatively high.

2.3. Study population

Children boys and girls aged 24 to 59 months, height for age (HFA) z-score <-2 standard deviations, Indonesian citizens living in Ma'rang district for at least six months, and the willingness to sign the informed consent form as the inclusion criteria for this study. The exclusion criteria were children who were ill at the time of data collection and children who had an allergy to shrimp products. The dropout criteria were children who are unwilling to have their blood samples taken and do not consume the rebon-shrimp products that are given following the prescribed recommendations.

2.4. Sample size

The determination of the sample size in this study refers to the minimum number of samples proposed by Gay and Diehl, which states that for experimental research, a minimum of 15 samples in 1 group is required [12]. The sample size formula to anticipate the possibility that the selected sample will drop out is as follows:

$$n' = \frac{n}{1-f}$$

Here, n'=number of research samples, n=calculated sample size, f = approximate proportion of drop outs, approx. 10% (f=0.1). The number of samples in this study is (n')=15 / (1-0,1)=17 samples. Although then minimum sample size was calculated to be 17 children each group, a total 88 samples were collected which divided into 2 groups, the intervention group (n=44 children) and the control group (n=44 children).

2.5. Sampling methods

The sampling technique used in this research is purposive sampling, where the research sample meets the criteria of the research subject set by the researcher. The research subjects were divided into two groups using the stratified sampling method. Research subjects were grouped into three groups according to age range (group 1: age 24-35 months; group 2: age 36-47 months; and group 3: age 48-60 months). After the subjects were divided into two groups, each group would be subjected to a Mann-Whitney test to see differences in the baseline on characteristic data, demographic data, eating behavior data, and parents' socioeconomic data.

2.6. Data collection

Before the screening, the research team was trained to measure nutritional status, interview, and questionnaire-filling techniques. The Indonesian Republic of Health Minister's Regulation No. 2 of 2020 Concerning Child Anthropometric Standards is referenced for determining a child's nutritional status. By carrying out anthropometric measurements, the research team screened to determine the sample according to the inclusion criteria. A trained team of 10 people with educational backgrounds in Nutrition, Nursing, and Midwifery measured weight and height. The researcher then visited the family home of the child who met the research inclusion criteria. The researcher will explain the research procedure to be carried out. If the family agrees, it will be followed by signing the informed consent. The research team will then conduct interviews to obtain demographic data for children, filling out the questionnaires using direct interview techniques.

Furthermore, the procedure for taking the child's venous blood is as much as 3 cc. Venous blood sampling will be carried out by trained laboratory medical personnel. The intervention group was given intervention through additional processed food products based on rebon shrimp. At the same time, the control group will be given a placebo made from low-protein flour. Both groups will receive the product for 90 days. To ensure compliance with the consumption of rebon shrimp products for children, the research team, in collaboration with cadres of the integrated health service post (*posyandu*), will monitor the consumption of rebon shrimp products for children, either directly, call by telephone, or contacted them by WhatsApp both through call and messages. The research team also provided a checklist sheet to parents to ensure their children consume the products provided. During the intervention, the research team took anthropometric measurements of children every 30 days to monitor the development of children's weight, height, and nutritional status. After 90 days of intervention, the research team will again take the child's venous blood, followed by anthropometric measurements, to assess the child's nutritional status.

2.7. Tools and instruments

Demographic data for children was filled in using a research questionnaire that the researcher had prepared to find out the data on the characteristics of children. The anthropometric examination used digital scales, a microtoise stature meter, and a body length measuring board. Determination of the child's z-score is done using the WHO Anthroplus application. Blood samples were taken on day 0 and day 90 using a 3 cc sterile syringe. The child's blood is put into a red vacutainer tube and stored in an ice gel box. After all the blood samples were collected, the serum was separated in the Ma'rang Health Centre laboratory. The serum was then put into a microcentrifuge tube. The serum samples were then taken to the Hasanuddin University

Medical Research Centre (HUM-RC) Laboratory of Hasanuddin University Hospital. Examine serum albumin levels using the Human ALB/Serum Albumin ELISA Kit RAB 0603-1KT Sigma-Aldrich at the HUM-RC Laboratory of Hasanuddin University Hospital.

2.8. Data analysis

Statistical analysis was performed using SPSS version 24.0. Respondent characteristics, demographic data, weight, height, eating behavior data, parent socioeconomic data, and serum albumin data were presented as mean±standard deviation (SD) or percentage. The Wilcoxon and Mann-Whitney tests determined the differences in respondent characteristics data, demographic data, eating behavior data, parent's socioeconomic data, nutritional intake, and serum albumin data. The level of significance (p-value) used was p<0.05.

2.9. Ethical consideration

The Research Ethics Committees (RECs) No. 271/UN4.6.4.5.31/pp36/2021 have given their approval for this study. The informed consent form that was signed by both parents and approved by the RECs was used to acquire informed consent. Researchers give close attention to moral problems, such as upholding privacy and secrecy, upholding fairness and inclusivity, and weighing the dangers and benefits.

3. RESULTS AND DISCUSSION

The results in Table 1 show that that there is no significant difference in body weight (p=0.736), height (p=0.497), and nutritional status based on HFA (p=0.054) between the intervention group and the control group. The data in Table 2 shows that there are significant differences in children's nutritional intake, which includes energy (p<0.001), protein (p<0.001), fat (p<0.001), and carbohydrates (p<0.001) between the two groups. Intake of energy (1,153.98 Kcal), protein (36.39 gr), fat (59.80 gr), and carbohydrates (161.68 gr) in the intervention group was higher than energy (998.44 Kcal), protein (19.09 gr), fat (44.74 gr), and carbohydrates (133.40 gr) in the control group. This result shows that children in the intervention group who received rebon shrimp-based food had better daily nutritional intake than children in the control group.

Tabel 1. Data on weight, height, and nutritional status based on HFA between intervention group and control group

Variables	Intervention group (n=44)				Control group (n=44)				p-value*
	Min	Max	Mean	SD	Min	Max	Mean	SD	
Weight (kg)	8.00	15.20	11.065	1.668	8.20	14.90	10.947	1.602	0.736
Height (cm)	76.00	100.50	88.584	6.494	79.00	98.50	87.715	5.404	0.497
Height for age (z-score)	-4.30	-2.01	-2.4968	0.588	-4.62	-2.06	-2.7382	0.570	0.054

Note: *Independent T-test

Tabel 2. Children's daily nutrition intake in the 3rd month of Intervention

Nutrition Intake	Intervention group (n=44)				Control group (n=44)				p-value*
	Min	Max	Mean	SD	Min	Max	Mean	SD	
Energy (Kcal)	1011.90	1319.60	1153.98	49.662	903.50	1145.80	998.44	52.352	<0.001
Protein (gr)	25.80	42.70	36.39	3.14	14.20	26.60	19.09	2.628	<0.001
Fat (gr)	43.50	77.30	59.80	6.32	27.10	60.40	44.74	6.78	<0.001
Carbohydrate (gr)	118.70	227.50	161.68	20.55	93.80	181.30	133.40	16.77	<0.001

Note: *Mann-whitney test

The results in Table 3 show that the intervention group had a p-value <0.001, indicating a significant difference in pre and post-intervention. In contrast, the control group had a p-value of 0.363, which indicates no significant difference. The result in Table 3 also shows that the children in the intervention group had an increase in serum albumin. In contrast, children in the control group tend to experience a decrease in serum albumin. Observation on day 0 in the intervention group in Table 3 showed the mean value (mean±SD) of children's serum albumin was 32.30±7.67 and increased on day 90 (47.86±12.24). Different results were seen in the control group, which experienced a decrease in the mean (mean±SD) of children's serum albumin levels by 31.54±9.91 on day 0, decreased to 33.22±11.48 on day 90.

Figure 1 compares changes in serum albumin levels of the intervention and the control group. It shows an increase in children's mean serum albumin level in the intervention group (15.55±11.04), while the control group experienced a decrease in the average albumin level of children (-1.91±16.80). Figure 2 shows that most children in the intervention group (n=30) and the control group (n=26) were in the low serum albumin category on day 0. After day 90, the children in the group that received the rebon product intervention experienced changes in the category where most children had normal albumin levels (n=35).

Tabel 3. Comparison of albumin serum (g/L) on days 0 and 90 in the intervention group and the control group

Comparison of albumin serum between Group	Min	Max	Mean	Std. Dev.	p-value
Intervention group					
Albumin serum day 0	22.20	48.62	32.30	7.67	<0.001*
Albumin serum day 90	22.32	66.02	47.86	12.24	
Control group					
Albumin serum day 0	21.10	64.15	35.14	9.91	0.363*
Albumin serum day 90	22.30	65.12	33.22	11.48	

Note: *Wilcoxon test

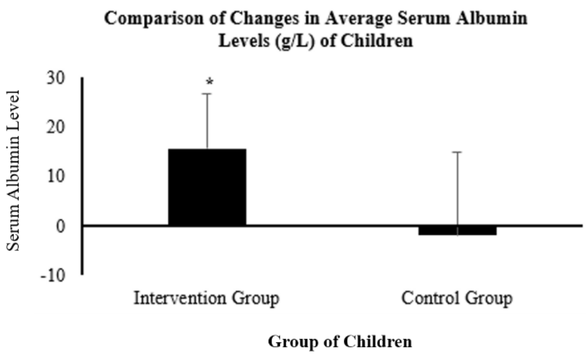

Figure 1. Comparison of changes in Albumin Serum Levels of the Intervention (n=44) and Control (n=44) Group. There was a significant change in the Intervention Group (p<0.001)

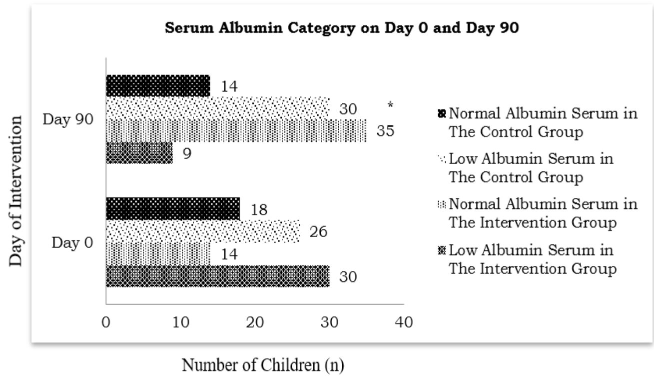

Figure 2. Categories of Albumin Serum in the Intervention Group (n=44) and Control Group (n=44) on day 0 and day 90. There was a significant change in serum albumin levels after the intervention between the two groups on day 90 (p<0.001)

In contrast, in the control group, the children who had normal serum albumin levels decreased to 14 people. Table 4 shows no difference in albumin levels in the two groups on day 0 before the intervention (p=0.180). However, after 90 days of intervention, there was a significant difference in albumin levels in the two groups (p<0.001). This study shows a significant change in serum albumin levels in the children who received rebon products.

Tabel 4. Comparison of albumin serum (g/l) on days 0 and 90 between the intervention group and the control group

Comparison of albumin serum between Group	Mean	Std. Dev.	p-value
Albumin serum day 0			0.180*
Intervention group (n=44)	35.14	9.91	
Control group (n=44)	33.22	11.48	
Albumin serum day 90			<0.001*
Intervention group (n=44)	35.14	9.91	
Control group (n=44)	33.22	11.48	

Note: *Mann-whitney test

4. DISCUSSION

The results of this study indicate that there was a statistical difference ($p<0.001$) in serum albumin levels in the intervention group, while the control group did not differ statistically ($p=0.363$). Children in the intervention group experienced an increase (mean±SD) of serum albumin levels of 15.55±11.04, while the results of serum albumin levels in the control group decreased by -1.91±16.80. There was no significant difference in serum albumin levels before the intervention in the two groups ($p=0.180$). However, after the administration of rebon shrimp products, there was a significant difference in serum albumin levels between the two groups ($p<0.001$).

According to the Republic of Indonesia's Ministry of Health's criteria for children's nutritional adequacy, children aged 1-3 years must consume 20 grams of protein per day and children aged 4-6 years must consume 25 grams per day of protein [13]. The results of this study indicate that the sample in the intervention group received a higher protein intake (36.39 gr) than the protein intake (19.09 gr) in the control group. These data indicate that supplementary feeding based on rebon shrimp can help increase the adequacy of children's daily protein needs. Rebon shrimp is a high-protein food. Amino acids combine to generate the nitrogen-containing compounds known as proteins. The primary structural element of muscles and other tissues in the body is protein. Furthermore, protein aids in the synthesis of hormones, enzymes, and hemoglobin. Energy can also be obtained from protein. Protein, however, is not the preferred option for an energy source. A protein must be broken down into its most basic form, amino acids, before the body can use it. About 20 types of amino acids are needed for human growth and metabolism. These amino acids consist of non-essential amino acids that can be synthesized by the body and essential amino acids that the body cannot synthesize. These essential amino acids need to be met through daily nutritional intake. Protein is available in various food sources, including animal and plant foods [14]. Animal protein is better than vegetable protein sources [15], [16]. Rebon is a source of animal protein, so supplementary food made from rebon shrimp can help meet the protein needs of children [9], [11].

Albumin is a protein component, making up more than half of plasma proteins. The liver synthesizes albumin and increases osmotic pressure, vital in maintaining vascular fluid [17]. Albumin is a serum protein reserve in the body produced by the liver. Typically, albumin constitutes 55% of all plasma proteins. The albumin amount for nutritional status assessment is average at >3.5 g/dL [18]. The liver produces albumin, which is the most prevalent plasma protein, and releases it into the blood. About 10% of the liver's total protein synthesis, or 10-15 g of albumin each day, is produced by a healthy adult, mostly in hepatocyte polysomes [19].

Albumin is a protein transporter in the body that monitors protein levels in response to dietary intake, particularly from protein sources [20]. The results of the proximate test of rebon products used in this study contained good protein. The protein content in nuggets is around 20.41 gr/100 gr; fish sticks are 26.35 gr/100 gr, and fried otak-otak is 25.11 gr/100 gr [9]. This study showed that the intervention of rebon products with a good protein content could help increase children's serum albumin levels by 15.55 g/L.

According to Prentice and Bates, Protein contributes amino acids needed to create bone matrix and has an impact on bone formation due to its ability to alter the release and activity of the osteotropic hormone insulin growth factor I (IGF-I). Proteins can therefore alter a person's genetic capacity for reaching their optimum bone mass. Animal protein food sources contain the necessary amino acids to satisfy the body's protein requirements. Growth issues may result from the body obtaining insufficient amounts of the amino acids it needs from meals [21], [22].

Inadequate intake of energy and nutrients, as well as infectious diseases, are factors that significantly contribute to the problem of stunting. A study in Jakarta in 2021 showed that children with a deficient intake of energy, macronutrients, and micronutrients were more prone to stunting. Children with energy deficiency have a six times greater risk of experiencing stunting. In comparison, children with protein deficiency have a four times greater risk of experiencing stunting compared to children who do not experience energy and protein deficiency [23]. Plasma levels of IGF-I and bone matrix proteins and growth factors, which are crucial for bone production, are influenced by the quantity and quality of protein consumed [24].

Because protein modulates the release and function of the osteotropic hormone IGF-I, protein consumption supplies the amino acids the body needs to construct bone matrix and influence bone development. Thus, protein consumption can influence a person's genetic potential for reaching their optimum bone mass. According to a study, protein supplementation can raise levels of IGF-1 and there is a positive relationship between protein intake and IGF-1 [25]. It has been demonstrated that a low protein diet reduces IGF-I synthesis and its consequences, which in turn reduces bone mineral acquisition. IGF-I has an impact on bone formation by influencing osteoblasts directly and promoting chondrocyte proliferation and differentiation in the epiphyseal growth plate [26]–[28]. Additionally, IGF-I boosts 25 hydroxyvitamin D3 renal conversion to the active hormone 1.25-dihydroxyvitamin D3, which promotes enhanced intestinal absorption of calcium and phosphorus [29].

Indicators of the adequacy of protein intake can be determined by the level of albumin as stored protein in the body and are related to changes in nutritional status. Serum total protein and albumin levels were significantly decreased under conditions of protein-energy deficiency [30]. Measurement of albumin levels is so far considered the standard in evaluating nutritional status conditions, and its indicative value increases when combined with prealbumin or retinol binding protein (RBP) [18].

Albumin synthesis and plasma levels are very sensitive to protein intake. Where can decrease drastically when a food shortage occurs and increase when the shortage is corrected. In this study, the provision of supplementary food made from rebon shrimp, which is high in protein, positively impacts the increase in serum albumin in children. Dietary protein and amino acids promote albumin production [31]. In the body, albumin is the main transport protein for micronutrients, so these nutrients are bound to albumin in the blood. If micronutrients are deficient, it will disrupt protein, fat, and carbohydrate metabolism so that growth is stunted. In addition, it can cause disturbances in the sense of taste and a decreased immune system against infection [7], [32]. To prevent this, efforts are needed to increase the intake of macronutrients and micronutrients. Rebon is a high-protein food ingredient. Foods high in protein will supply amino acids that are useful in albumin synthesis, which is essential for someone with low albumin levels.

Although the provision of rebon-based supplementary food has been able to help meet children's protein intake, there are still children who are still in the low albumin category. This occurs due to the continuous change of protein in the body (protein hydrolysis and catabolism in malnourished children and co-morbidities) into amino acid components used for growth and repair of body tissues (protein turnover) which lasts a short time, causing little body protein reserves obtained from the intake of high protein foods [33]. With a half-life of 19 days, albumin is the most prevalent protein in the body and has the best chance of being broken down. Other studies have shown that in malnourished children, there is a decrease in the synthesis and breakdown of total body protein. This is due to the adaptation to a lack of energy in malnourished children [32]. Albumin formation decreases relatively early in conditions of protein malnutrition [34].

5. CONCLUSION

Based on the results of this study, it was concluded that there was an increase in serum albumin after the intervention of additional food products made from rebon shrimp in stunting children aged 24-60 months. Rebon shrimp is a local food that is high in protein, inexpensive, and helpful in increasing albumin levels in the blood. It is hoped that rebon shrimp as an affordable local food for the community can be an alternative nutritional intervention that can be carried out in Indonesia.

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