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Covid-19 Era in the Coastal Areas: The Cookies Formulation Gonad of *Diadema setosum* and its Impacts on Malnutrition During Infection

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Abstract

Infection with *Salmonella typhi* bacteria during the COVID-19 era has the potential to worsen malnutrition in children in low- and middle-income nations, particularly around the coast. Recent studies have demonstrated that one of the best strategies for preventing malnutrition is consuming local food sources from the sea, which are easy to acquire, inexpensive, and high in nutrients. In this study, 15 male mice were used as the test subjects. They were split into two groups: the intervention group and the control group, both before and after intraperitoneal injection of *Salmonella typhi*. The intervention group was given dosages of cookies containing *Diadema setosum* gonad (0.40 mg/kg of body weight), whereas the control group received only a natural diet. The blood samples were then analyzed to measure their hemoglobin levels using the Sahli haemometer method, and serum albumin concentrations were determined using the bromocresol green method. The results show that the treatment with *Diadema setosum* gonad cookies had a significant effect on the levels of albumin and hemoglobin (Hb), as well as the body weights of the intervened mice, with a statistically significant increase ($p<0.05$). Conversely, BALB/c mice in the control group demonstrated a decrease in their initial food intakes,

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resulting in a significant reduction in body weight, albumin, and hemoglobin (Hb). This finding implies that the cookie formula with *Diadema setosum* gonad as the principal ingredient has potential benefits in lowering the incidence of malnutrition for children.

Introduction

Typhoid fever due to infection with *Salmonella typhi* bacteria is still high in low-income countries, especially in coastal areas.[1,2] The impact of *Salmonella typhi* in the COVID-19 Era has the potential to increase cases of malnutrition in children in low-income and middle-income countries.[3,4] Decline in income and reduced access to appropriate nutritional foods, as well as interruptions in nutrition and health services all of which lead to a decrease in child nutritional status.[5] Meanwhile, it is predicted that more than 2 million children are at a high risk of malnutrition.[6,7] Malnutrition has been a challenging public health issue leading to 45% of deaths in children under the age of 5, resulting in 17 million wasted children and 149 million stunted children all over the world.[8] On a national basis, Indonesia has the rate of malnourished children at 17.7%, while the prevalence of stunted children is still significant, reaching approximately 30.8 %, with the prevalence of stunted growth in the Southeast Sulawesi Province alone at 28.7%.[9] Hidden hunger is a risk factor for many different health problems, including stunting, which is one of the most common signs of malnutrition found all over the world.10 Globally, a rising trend of these malnutrition cases is predicted to occur during COVID-19 pandemic.

Nutritional assessments and malnutrition screening are necessary to be implemented to identify individuals with the risk of malnutrition.[11] Including body mass index (BMI), hemoglobin level, low albumin level, and low body weight.[12] Low albumin level is related to a higher mortality rate of COVID-19 pneumonia, therefore, nutritional evaluation should be a priority, especially for patients who are unable to meet their recommended protein and energy demand.[13,14]

One of the alternative approaches to serve certain population groups due to the detrimental impacts of COVID-19 pandemic is the utilization of local wisdom-based food sources that affordable with high nutrient contents. *Diadema setosum* is one of the sea urchin species that disperse in abundance populations in the Indonesian sea waters, mainly in Southeast Sulawesi Province. *Diadema setosum* gonad has quality nutrient contents (n-3 Fatty Acid, protein, albumin, vitamin A, vitamin E, minerals including Zn, Fe, and selenium) and able to improve the body's immune system as demonstrated from various studies.[15,16] Our previous study, BALB/c mice infected with the *Salmonella typhi* strain and treated with *Diadema setosum* gonad extract (200 mg/kg BW) for 17 days demonstrated an increase in body weight (1.7g) relative to the control group. Significant weight reduction ($p<0.05$).[17] Concerning to its potential nutritional benefits, we have been developing a formula of gonad cookies made of the primary ingredient of *Diadema setosum* gonad for dietary consumption among children who are vulnerable to malnutrition related to the risk of *Salmonella typhi* infection. However, the effects of this formula on human nutritional status have not been elucidated yet that prompt us to investigate its nutritional effects on body weight, albumin level and hemoglobin (Hb) level on experimental animals as the objective of our basic research.

Materials and Methods

Animals and Treatments

In this research, 12 weeks old BALB/c mice strain, free of pathogens, reared in stainless-steel cages at the Molecular Biology and Immunology Laboratory for Disease Infection, Faculty of Medicine, Halu Oleo University, Kendari. All mice were fed by standard diets ad libitum and were kept under a controlled temperature of 23° C, 50% room humidity, on a 12-hour light ray per day. This research used a total of 15 BALB/c mice with body weights ranging from 30 to 40g.

Ethical Approval

The Health Medical Research Ethics Committee at the Faculty of Medicine, Halu Oleo University (Kendari, Indonesia) approved this research with registration number: 165/UN29.17.1.3/ Kometik/2021. This protocol was approved

under the Animal Welfare Act and the Regulation of Animal Experiments (approval number: 2014/6979). All applicable international, national, and institutional guidelines for animal care and use were followed.

Animals and Diets

The experimental mice (as shown Figure 1) were divided into two groups, BALB/c mice in the first group were not fed with *Diadema setosum* gonad cookies (control group) and those in the second group (intervention group) were fed with *Diadema setosum* gonad cookies (0.4 g/ kg body weight). For each group type, observation was conducted over 21 days. On day 0, day 7, day 14, and day 21, each mouse was weighed using a Triple Beam Balance (OHAUS). Blood samples of BALB/c mice were collected from the two groups 7 days later, during the intervention period (no LPS), and they were then allowed to rest for 3 hours. The two groups of BALB/c mice were afterward injected intraperitoneally with *Salmonella typhi* lipopolysaccharide (LPS; 0.2 ml x 10^3 ml / CFU) as outlined in the previous technique.[15] After drawing blood samples, BALB/c mice in the intervention group were orally treated with the *Diadema setosum* gonad cookies (0.4 g/ kg body weight) using oral feeding cannulas. The contents of nutrient in cookies *Diadema setosum* gonad were analyzed by The First Indonesian Molecular Biotechnology Company, (PT. Saraswati Indo Genetech, Indonesian, Bogor), and are presented in Table 1.[18]

Table 1: Nutrient content in cookies of *Diadema setosum gonad* (100 g)[18]

Type of Nutrient	Unit	Content	Methods
Vitamin E	mg/100 g	2.62	18-5-1/MU/SMM-SIG, HPLC
Vitamin A	mcg/100 g	29.74	18-5-1/MU/SMM-SIG, HPLC
Magnesium (Mg)	mg/100 g	106.33	18-13-1/MU/SMM-SIG, ICP-OES
Iron (Fe)	mg/100 g	2.63	18-13-1/MU/SMM-SIG, ICP-OES
Zinc (Zn)	mg/100 g	2.29	18-13-1/MU/SMM-SIG, ICP-OES
Protein	%	8.70	18-8-31/MU/SMM-SIG, Kjeltec
Total Fat	%	17.27	18-8-5/MU/SMM-SIG. Weilbull
Total Energy	Kcal/100 g	458.03	Calculation

Analyses of Hb and serum albumin

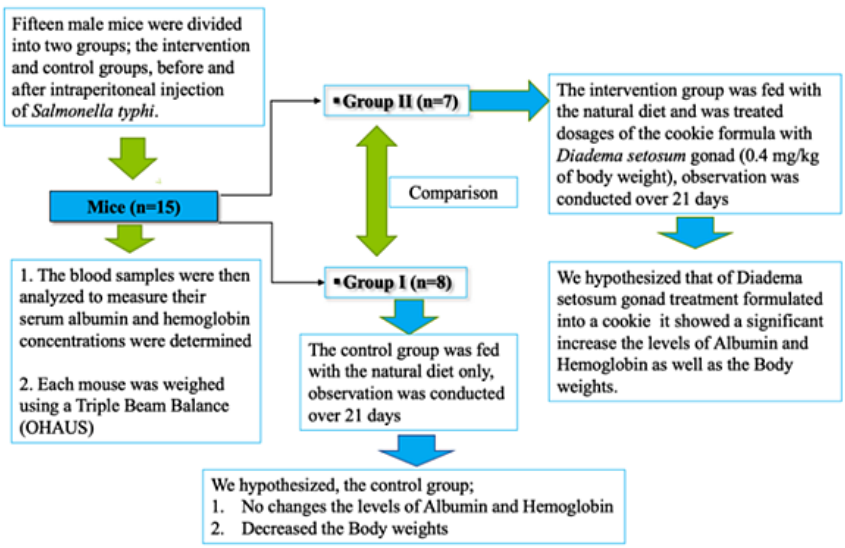

Fig. 1: a flowchart representation of the research design

Blood samples from mice were taken again 14 days after the commencement of pasca-LPS induction, a 1-ml insulin syringe was used to draw the blood samples with a concentration of 0.2 mL and these samples were obtained from the caudal part of each mouse. The blood samples were then analyzed to measure their Hb levels using the Sahli haemometer method which was referred to in a previous study.[19] However, serum albumin concentrations were determined using the *Bromocresol green* method and their absorbance values were measured *spectrophotometrically* at 628 nm.[20]

Statistical Analysis

All recorded data were expressed as a mean standard deviation (±SD). Repeated measurements were used to compare the levels of albumin, hemoglobin, and body weights of BALB/c mice between the control and treatment groups. *Diadema setosum* gonad includes more n-3 Fatty Acid, protein albumin, vitamin A and vitamin E, and minerals such as Zn, Fe, and selenium. An independent sample t-test was utilized to compare albumin, hemoglobin, and body weight levels between the two doses. Statistical significance was considered as $p < 0.05$.

Results

After 21 days of observation, the experimental results revealed the following differences in the dynamic changes of the body weight of BALB/c mice between the two groups: The body weight of mice in the intervention group of *Diadema setosum* gonad cookies (dose 0.4 g/kg body weight) increased within seven days compared to those in the control group, indicating that the body weight of mice in the intervention group of *Diadema setosum* gonad cookies (dose 0.4 g/kg body weight) increased within seven days. Meanwhile, there was a substantial difference in body weight between the control and intervention groups of BALB/c mice 14 days after the onset of post-LPS-induced (p-value = 0.001). The body weight of mice in the intervention group was significantly reduced within seven days from the beginning of post-LPS-induced, but it showed a significant increase when compared with those in the control group, which had a drastically reduced body weight ($p<0.05$) within 14 days from the onset of post-LPS induction.

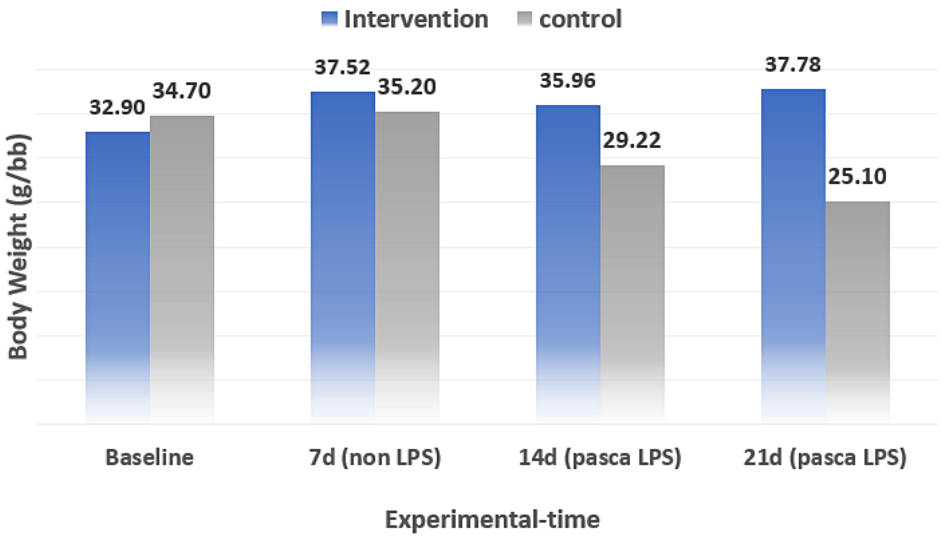

Fig. 2: Dynamics of change in body weights of mice

Results of the experiment as shown in Figure 3 represent the clinical effects of the treatment of *Diadema setosum* gonad cookies on both hemoglobin level and serum albumin level of BALB/c mice. Within 14 days of pasca-LPS induction, the hemoglobin level of BALB/c mice in the intervention group was substantially higher than those in the control group, whereas the serum albumin level of

BALB/c mice in the intervention group was relatively stable with a constant line, compared to those in the control group with significantly lower levels. This suggests that BALB/c mice in the control group failed to control infections, and as a result of inflammation and a decrease in initial appetite, albumin and hemoglobin production were likely suppressed.

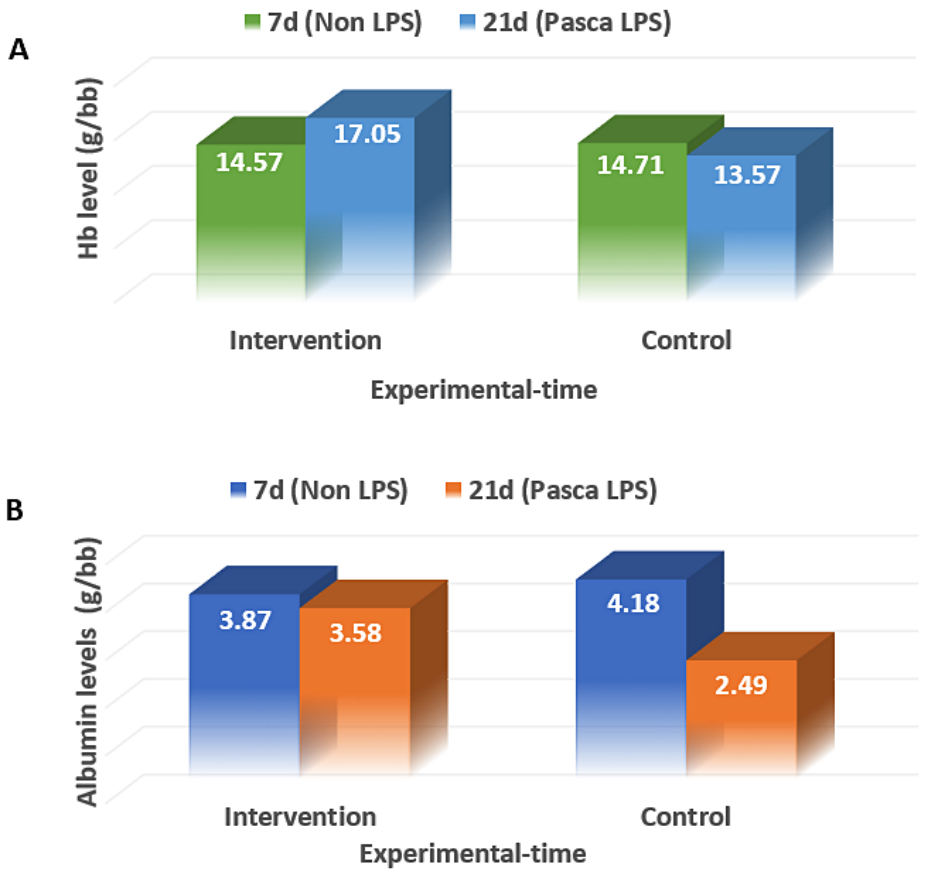

Fig. 3: Graph of the alterations of (A) Hb level and (B) Albumin level of mice

Discussion

Malnutrition, inflammation, and insufficient protein intake can contribute to decrease in albumin levels in the blood.[21] This is supported by previous research in which a decrease in serum albumin in a mouse model showed a strong correlation between a later phase of inadequate protein-energy requirement and body weight.[22] A classic example also demonstrated a strong link between hypoalbuminemia and malnutrition, which leads to the kwashiorkor syndrome in infants and children, caused by a diet low in protein and calories compared to normal limits.[23]

Malnutrition-related risk factors are likely to occur as a result of inadequate requirements of energy protein intake and infections, as seen in BALB/c mice in the control group which demonstrated a decrease in their initial food intakes, resulting in a significant reduction in body weight. Conversely, BALB/c mice in the intervention group increased weight ($p<0.05$) within 14 days of observation from the onset of post-LPS induction, and treatment using the *Diadema setosum* gonad on the albumin level, showing an improved clinical effects and increase in hemoglobin level. Nutrient components in the gonad of *Diadema setosum* might have a higher

ability in sustaining albumin level and increasing hemoglobin level of BALB/c mice which may help with early defense mechanisms from infection and promote appetite recovery.

Our findings are comparable with earlier research in that a lower mouse body weight is only associated with a lower serum albumin concentration ($r = 0.57$, $p<0.05$) and that mouse body weight remained positively correlated with serum albumin on the ninth day ($r = 0.78$, $p<0.05$).[20] A proportionate reduction in total food consumption, such as in 25% or 50% of normal, is insufficient to maintain a normal growth rate.[24] Nutritional risks, as well as low levels of albumin, prealbumin, protein, and zinc, were positively linked with the severity ($p<0.01$).[25] Serum albumin and hemoglobin concentrations have been found to be strong predictors of survival in infected and malnourished humans.[26]

Foods that contain n-3 fatty acids and micronutrients can affect hemoglobin levels. Low n-3 Fatty Acid status can increase anemia in the infectious mice model compared to the group of mice that received sufficient n-3 Fatty Acid treatment.[27,28] Human studies have shown that food or supplements containing n-3 fatty acids and micronutrients can be an effective, safe, and affordable therapy for anemic patients.[29] The use of cookies or biscuits in clinical trials shows good prospects in improving children's diet by providing bioactive compounds such as n-3 Fatty Acid, proteins, phenolic compounds, vitamins and minerals.[30] Our recommendation of this study, *Diadema setosum* gonad could have potential effects for preventing child malnutrition related to the risk of *Salmonella typhi* in the COVID-19 Era within a long duration of time and consistent with continuing growth.

Mitigation measures and adaptation in the adverse situation due to the COVID-19 pandemic by empowering regional potentiality scale is one of the alternative strategies to serve certain population groups and provide health benefits by exploring local-based food sources.[31] Innovations based on the use of locally sourced foods have the potential to improve the body's defensive system against infection.[32] It is of important interest to know that *Diadema setosum* gonad contains higher n-3 Fatty Acid, protein albumin, vitamin A and vitamin E, and minerals including Zn, Fe, selenium.[16] Referring to this present study and other studies, nutritious foods that are safe and affordable are necessary to be developed for the prevention of infection in the COVID-19 Era.[33] It is critical to safeguard future generations by bolstering community health systems and prioritizing access to proteins, n-3 PUFAs, and micronutrients from sea biota sources which are affordable and low cost for the improvement of sustainable child nutritional status. This study has limitations, so the applicability of these findings to humans must be examined further, as there are still substantial differences between humans and rats.

Conclusions

Nutritional quality and innovation can be derived from the utilization of sea-based food sources such as the cookie formula made with the primary ingredient of *Diadema setosum* gonad, which could have implications on the improvement of child nutritional status given its potential benefits in reducing the burden of malnutrition efficiently and cost-effectively for the management of nutritional policies in the COVID-19 pandemic era.

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Conflict of Interest

All authors of this work declare no conflict of interest

References

1. Haggi A., Khurram M., Din M.S.U., Aftab M.N., Ali M., Ahmed H., Afzal M.S. COVID-19 and *Salmonella Typhi* co-epidemics in Pakistan: A real problem. J Med Virol. 2021; 93(1): 184–186
2. Siegel F.R. Disease Protection in Sea

Coast (and Inland) Cities: Problems in Dense Populations with Shantytowns/Slums. Adaptations of Coastal Cities to Global Warming, Sea Level Rise, Climate Change and Endemic Hazards. 2020; 49–62.
3. Mehta S. Nutritional status and COVID-19: an opportunity for lasting change? *Clin Med (Lond)*. 2020; 20(3): 270–273.
4. Menendez C., Gonzalez R ., Donnay F., Leke R.G.F. Avoiding indirect effects of COVID-19 on maternal and child health. Lancet Glob Health. 2020; 8(7): e863–e864.
5. Akseer N., Kandru G., Keats E.C., Bhutta Z.A. COVID-19 pandemic and mitigation strategies: implications for maternal and child health and nutrition. *Am J Clin Nutr.* 2020; 112(2):251-256
6. Kabir M., Saqib M.A.N., Zaid M. COVID-19, economic impact and child mortality: A global concern. *Clin Nutr.* 2020; 39(7): 2322–2323.
7. UNICEF Situation tracking for COVID-19 socio-economic impacts. June, 2020. https://data.unicef.org/resources/rapid-situation-tracking-covid-19-socioeconomic-impacts-data-viz/, Accessed 13 June 2021
8. Titaley C.R., Ariawan I., Hapsari D., Muasyaroh A., Dibley M.J. Determinants of the Stunting of Children Under Two Years Old in Indonesia: A Multilevel Analysis of the 2013 Indonesia Basic Health Survey. *Nutrients.* 2019; 11(5): 1106.
9. Kementerian Kesehatan Repoblik Indonesia, Laporan Riskesda 2018. https://www.kemkes.go.id/resources/download/info-terkini/hasil-riskesdas-2018.pdf. Accessed 15 July 2021
10. Hussein A. M. S., Fouda K., Abozed S. S., Mohamed R. S., Salama M. F., Hussein M. M. Improvement of Growth Retardation and Related Immunodeficiency by Dietary Intervention with Crackers Containing Animal Source Ingredients in Malnourished Rats. *Curr Res Nutr Food Sci.* 2021; 9(3): 875-889.
11. Hamdu D., Moloney L., Rozga M., Cheng F.W. Malnutrition Care During the COVID-19 Pandemic: Considerations for Registered Dietitian Nutritionists. *J Acad Nutr Diet.* 2021; 121(5) 979-987
12. Li T., Zhang Y., Gong C., Wang J., Liu B., Shi L., Duan J. Prevalence of malnutrition and analysis of related factors in elderly patients with COVID-19 in Wuhan, China. Eur J *Clin Nutr.* 2020;74(6):871-875.
13. Zhou F., Yu T., Du R., Fan G., Liu Y., Liu Z., et al. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. *Lancet.* 2020;395:1054–1062.
14. Stachowska E., Folwarski M., Jamioł-Milc D., Maciejewska D., Skonieczna-Żydecka K. Nutritional Support in Coronavirus 2019 Disease. *Medicina* (Kaunas). 2020; 56(6): 289.
15. Salma W.O., Wahyuni S., Nursin K., Yasir Haya L.O.M., Irawan Y., As´ad S. The Effect of Sea urchin (*Diadema setosum*) gonad Extract on gene ekspression of FOXP3 and the production cytokine on Salmonella typhi. *Pharmaceutical Science.* 2018;8 (12): 140-146.
16. Salma W.O., Wahyuni S.,Yusuf I, Yasir Haya L.O.M., Yusuf I, Asad S. Immune nutrient content of sea urchin (*Diadema Setosum*) gonads. *Int J Nutr Food Sc*. 2016; 5 (5):330–336.
17. Salma W.O., Hafizah I., Nurtamin T., Yasir Haya L.O.M., As´ad S. The Potential Gonad of *Diadema setosum* as a Healthy Food to Improve the Nutritional Status of Coastal´s Children. *Int. J. Sci.* Basic Appl. Research. 2016, 27 (2):189-193.
18. Salma W.O., Yasir Haya L.O.M., Binekada I.M.C., Fristiohady A., Alifariki L.O. Buku Referensi Potret Masyarakat Pesisir; Konsep Inovasi Gizi & Kesehatan.Yogyakarta-Indonesia: Deepublish. 2021.: p.192
19. Delianis P., Ana A., Indriatmoko., Sri S., Dwi H. The Potency of Sea Urchin (*Diadema setosum*) Gonad on Brain Cells of White Rats (Rattus norvegicus). *Asian Journal of Pharmaceutics.* 2016; 10 (2):100-7
20. Alaverdashvili M., Li X., Paterson P.G. Protein-Energy Malnutrition Causes Deficits in Motor Function in Adult Male Rats. *J Nutr.* 2015; 145(11): 2503–2511.
21. Cray C., Zaias J., Altman N.H. Acute phase response in animals: a review. Comp Med. 2009;59(6):517–26.
22. Don B.R., Kaysen G. Serum albumin: relationship to inflammation and nutrition. *Semin Dial.* 2004;17(6):432–7.
23. Levitt D.G., Levitt M.D. Human serum albumin homeostasis: a new look at the

roles of synthesis, catabolism, renal and gastrointestinal excretion, and the clinical value of serum albumin measurements. *Int J Gen Med.* 2016; 9: 229–255.

24. Ling P.R., Bistrian B.R. Comparison of the effects of food versus protein restriction on selected nutritional and inflammatory markers in rats. *Metabolism.* 2009; 58(6): 835–842.
25. Allard L., Ouedraogo E., Molleville J., Bihan H., Giroux-Leprieur B., Sutton A., Baudry C. Malnutrition: Percentage and Association with Prognosis in Patients Hospitalized for Coronavirus Disease 2019. *Nutrients.* 2020; 12:3679. https://doi.org/10.3390/nu12123679
26. Gupta K.B., Gupta R., Atreja A., Verma M., Vishvkarma S. Tuberculosis and *Nutrition.* Lung India. 2009; 26(1): 9-16
27. Hayford F.E.A., Dolman R.C., Ozturk M., Nienaber A., Ricci C., Loots D.T., Brombacher F., et al. Adjunct n-3 Long-Chain Polyunsaturated Fatty Acid Treatment in Tuberculosis Reduces Inflammation and Improves Anemia of Infection More in C3HeB/FeJ Mice With Low n-3 Fatty Acid Status Than Sufficient n-3 Fatty Acid Status. *Front Nutr.* 2021; 8: 695452. https://doi.org/10.3389/fnut.2021.695452
28. Nienaber A., Baumgartner J., Dolman R.C., Ozturk M., Zandberg L., Hayford F.E.A., et al. Omega-3 Fatty Acid and Iron Supplementation Alone, but Not in Combination, Lower Inflammation and Anemia of Infection in Mycobacterium tuberculosis-Infected Mice. *Nutrients.* 2020; 12(9): 2897
29. Daak A.A., Ghebremeskel K., Hassan Z., Attallah B., Azan H.H., Elbashir M.I., Crawford M. Effect of omega-3 (n-3) fatty acid supplementation in patients with sickle cell anemia: randomized, double-blind, placebo-controlled trial. *Am J Clin Nutr,*. 2013; 97(1):37-44.
30. Goubgou M., Songré-Ouattara L.T., Bationo F., Lingani-Sawadogo H., Traoré Y., Savadogo A. Biscuits: a systematic review and meta-analysis of improving the nutritional quality and health benefits. *Food Prod Process and Nutr.* 2021; 3(1): 26.
31. Farrell P., Thow A.M., Wate J.T, Nonga N., Vatucawaqa P., Brewer T., Sharp M.K., Farmery A., et al. COVID-19 and Pacific food system resilience: opportunities to build a robust response. *Food Secur.* 2020; 12(4): 783–791
32. Orhan I. E., Deniz F.S.S. Natural products as potential leads against coronaviruses: could they be encouraging structural models against SARS-CoV-2?. *Natural Products and Bioprospecting*. 2020;10(4):171–186.
33. Fore H.H., Dongyu Q., Beasley D.M., Ghebreyesus T.A. Child malnutrition and COVID-19: the time to act is now. *Lancet.* 2020; 396(10250): 517–518.