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The acute supplementation of combination juice of yellow watermelon *(citrullus lanatus thunb.)* - plantain (*musa paradisiacal* var. *Sapientum l.*) suppress post-exercise blood lactic acid production in rats

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ABSTRACT

Background: Yellow watermelon contains citrulline, which can suppress lactic acid production, while plantains contain potassium which is important for muscle performance. The yellow watermelon and plantain combination juice potential to be a natural sports drink that delays muscle fatigue by suppressing lactic acid production after exercise.
Objectives: To determine the effect of yellow watermelon-plantain juice on lactic acid in rats after swimming test.
Materials and Methods: This true experimental study used a post-test-only with controlled group design. Thirty Sprague Dawley rats, eight-week-old, male, were divided into five groups, namely positive control (C+), negative control (C-), dose 1 (P1), dose 2 (P2), and dose 3 (P3). The C (+) group received no juice and was not tested swimming, the C (-) group received no juice but was tested swimming, P1 received combined juice up to 1.8 g and tested swimming, P2 received combined juice up to 3, 6 g and tested swimming, P3 received combined juice up to 1.8 g with the addition of 0.27 g granulated sugar and tested swimming. The juice is given 30 minutes before the test. The swim test was performed for three minutes; after that, the blood was taken to test the lactic acid levels. The data were analyzed using the one-way ANOVA and the advanced post-hoc with the least significant difference test.
Results: The lactic acid levels in C (+), in C (-), P1, P2, and P3 groups after swimming test were 1.38 mMol / L; 7.14 mMol / L; 3.74 mMol / L; 1.66 mMol; and 2.91 mMol/L. There were differences in levels of lactic acid ($p < 0.05$) in each group after the combination juice intervention was given.
Conclusion: Combination juice of yellow watermelon-plantain has an effect on lactic acid levels after swimming test. Dose 2 (3.6 g) was the best because it produces the lowest lactic acid after the swimming test.

Keywords: Yellow watermelon; Plantain; Lactic acid; Swimming test

BACKGROUND

Energy metabolism during anaerobic exercise is accentuated exclusively from muscular strength with high explosive power.[1] This process begins with the glucose breakdown process (glycolysis) as well as the glycogen breakdown process (glycogenolysis) and is independent of oxygen availability during ATP (Adenosine Tri Phosphate) formation.[2] Thus, it is important to acknowledge glucose availability in the muscle since its metabolic advantage is providing speed and strength in a short period.

Low performance in exercise can be caused by lactic acid accumulation which faster than it should be. However, exercising has an unavoidable side effect that is the production of lactic acid, which might induce muscle fatigue. L-citrulline is one of the most popular types of nutritional supplements that are legal ergogenic aids. L-citrulline has beneficial functions such as accelerating metabolite waste removal like lactic

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acid, improving endurance performance, and promoting faster recovery after exercise.[3] L-citrulline also acts as a source of energy during exercise.[4] A findings by Pérez-Guisado and Jakeman reported that supplementation of 8 g of citrulline malate can reduce muscle soreness at 24 and 48 hours following anaerobic exercise.[5] Supplementation with 8 g of citrulline 1 hour before exercise also can increase the number of reps lifting weights.[6]

Previous studies related to nutritional ergogenic have investigated that some fruits potentially be a source of nutritional ergogenic. Watermelon is one type of fruit that naturally rich in amino acids including L-citrulline. Watermelon also contains carbohydrates, potassium, phytonutrients such as carotenoids (lycopene and beta carotene), polyphenolics, and vitamins.[7] The rind watermelon juice contains 45.02 mg/g L-citrulline and flesh watermelon juice contains 43.81 mg/g.[8] Rind watermelon juice contains a higher L-citrulline content compared to flesh watermelon, and L-citrulline in yellow watermelons is higher than red watermelons.[9] Nevertheless, the findings may suggest that both flesh and rind watermelon juices potentially offer similar benefits to pure L-citrulline for improving exercise performance.[10] A study by Ridwan, *et al*., showed that supplementation with 100% flesh watermelon juice improves endurance in swimming performance in rats.[8]

Another type of fruit that has a beneficial effect as nutritional ergogenics is plantain. Plantain is one type of banana fruit, contains about 31.15 g of carbohydrates and 564 mg potassium.[11] Potassium is an electrolyte that acts as a body fluid balancer, to deliver nerve impulses and muscle contractions. Potassium plays role in muscle relaxation and also promoting muscle fatigue delay.[12] A previous study by Ustafia, *et al*., showed that bananas were more effective than banana milkshakes in removing fatigue after exercise.[13] A findings by Faturochman also showed that banana was effective to prevent muscle fatigue in an anaerobic sprint.[14] The carbohydrates both in watermelon and plantain are a good source of energy during exercise, so they can be promoting fatigue delay. Increasing the amount of glycogen storage by 25-100% can be done by consuming the carbohydrates before the exercise. It can delay fatigue during exercise up to 20%.[15] Many evidence suggest that L-citrulline give a beneficial effect for improving exercise performance. However, there is little evidence to support that watermelon juices could provide such improvements. Cutrufello, *et al*., reported that acute watermelon juice supplementation appears to be ineffective in improving exercise performance.[16] Ridwan *et al*., who reported that supplementation with 100% flesh watermelon juice improves endurance in swimming performance did the experiment in the longer term to avert such drawbacks.[8] Nevertheless, this study design used acute term supplementation by considering that the nutritional ergogenics from both the fruits will give the effect in a short period. This study used rats as subjects not in athletes directly, because the formula with the best effect has not been found. This study needs many subjects to investigate which the best formula with optimal effect.

MATERIALS AND METHODS

Design, location, and time

This study was a true experimental with a post-test-only randomized controlled group design. The experiment was conducted in Food and Nutrition Research Centre Inter-Laboratory Universitas Gadjah Mada (UGM) in May 2019. Ethical research for this study was approved by the research ethics committee with reference number 2190/KEPK/V/2019.

Materials and juices preparation

The yellow watermelon was classified as Black Orange type (*Citrullus lanatus 'Black Orange'*), which was obtained from yellow watermelon plantation, Nusawungu, Cilacap, Central Java. with approximately weighing between 4.0 – 5.0 kg. The plantain banana used in this study was classified as *Musa paradisiaca Linn,* which was obtained from plantain plantation, Kalimanah, Purbalingga, Central Java. The plantain was characterized as yellow-colored, weighing 150 – 200 grams each fruit. Total glucose and potassium content from yellow watermelon and plantain in this study were measured in Food and Nutrition Research Centre

Inter-Laboratory UGM with analysis certificate number PS/157/V/2019. The total glucose contents for yellow watermelon and plantain were 5.19% and 15.40%, and the potassium was 111.921 mg/kg and 438.910 mg/kg respectively. The watermelons and plantains peeled to obtain the yellow flesh and white rind watermelons and the flesh of plantains. The 100 g watermelons (flesh and rind) and 100 g flesh plantains processed using a commercial blender to obtained combination juice. The juice is prepared freshly.

Animal models and treatment doses

Thirty-five, healthy, eight-week-old male Sprague-Dawley rats were obtained from the Food and Nutrition Research Centre Inter-Laboratory UGM. The animals were acclimatized for three days with a normal pellet diet and filtered tap water *ad libitum*. Rats were assigned into 5 groups of 7 rats: negative control (C-) group (not treated with combination juice before exercise), positive control (C+) group (no combination juice administration and no exercise), single-dose (P1) group (treated with single-dose of combination juice, 1.8 grams of solid forms diluted with water, before exercise), double dose (P2) group (treated with double-dose of combination juice, 3.6 grams of solid form diluted with water, before exercise), and single-dose with sugar (P3) group (treated with single-dose of combination juice 1.8 grams of solid form diluted with water and added with 0.27 grams of granulated sugar, before exercise). Determination of the combination juice doses administered to the rats was based on the calculation of mean fruit consumption servings number in humans. In humans, these fruits can be consumed separately as a fruit or as a juice with the addition of water. The details for determining the dose of combination juice in this study are shown in Table 1 below.

Table 1. Determination of The Combination Juice Doses

Groups	Treatment Dose	Explanation
Treatment 1 (P1)	1.8 g combination juice	In humans, equal with consumption of 50 g yellow watermelon + 50 g plantain
Treatment 2 (P2)	3.6 g combination juice	In humans, equal with consumption of 100 g yellow watermelon + 100 g plantain
Treatment 3 (P3)	1.8 g combination juice + 0.27 g granulated sugar	In humans, equal with consumption of 50 g yellow watermelon + 50 g plantain + 13 g granulated sugar

Exercise treatment

The rats were fed with combination juice as the source of energy, L-citrulline, and potassium before exercising. Thirty minutes after the combination juice was administered, the exercise was conducted. The anaerobic exercise was set as a swimming test, where the rats were drowned in a water pool and were left to swim for three minutes. Three minutes is the maximum duration of a lactic acid system in anaerobic exercise.[17] After three minutes of swimming, the rats were pulled up from the pool, and the post-exercise blood sample was taken.

Serum Analysis

The blood sample was collected immediately after exercise. Blood serum was attained through rats' orbital sinus to obtain lactic acid levels. Blood serum analysis was required to analyze lactic acid level after exercise resulted from a different dose of yellow watermelon-plantain juice administration specifically in treatment groups, and compared between treatment groups and control groups.

Data Analysis

Data obtained were analyzed using SPSS 20 for Windows (SPSS Inc., Chicago, IL). The data were presented as mean ± standard deviation (SD). One-way analysis of variance (ANOVA) followed with post-hoc LSD test were applied to identify statistical differences between groups. Statistically, a significant result was considered at a p-value < 0.05.

RESULTS

The mean body weight of rats in this study was 209 g. Since the study design only used a post-test, data from the positive control group were used as a comparison standard. Rats are considered to be due to factors that can affect lactic acid production, such as training volume or physical activity, as homogeneous. Figure 1 shows the mean lactic acid levels in the blood of rats after swimming for three minutes. As a normal standard, the results of the C (+) group lactic acid test were used when the mice received no treatment, and their blood lactic acid levels were monitored simultaneously with the other treated groups. The normal blood lactic acid levels in this study were 1.38 mmol / L, which corresponds to the normal blood lactic acid concentrations (at rest) in humans which are < 2 mmol / L.

The highest lactic acid levels were in the C (-) group, which received no juice but did a swim test. The blood lactic acid level of group C (-) was five times higher than C (+). Meanwhile, in the treated group, the highest lactic acid levels were produced by the P1 group, almost three times higher than normal levels. The P2 group scored was the lowest of the treatment groups, almost equal to the normal group. The P3 group, with the same juice dose as P1 with added sugar, produced lower lactic acid levels than P1. The addition of sugar affects lactic acid production after swimming. In general, however, the results of statistical tests showed that there were significant differences between groups of dose variations $p < 0.05$.

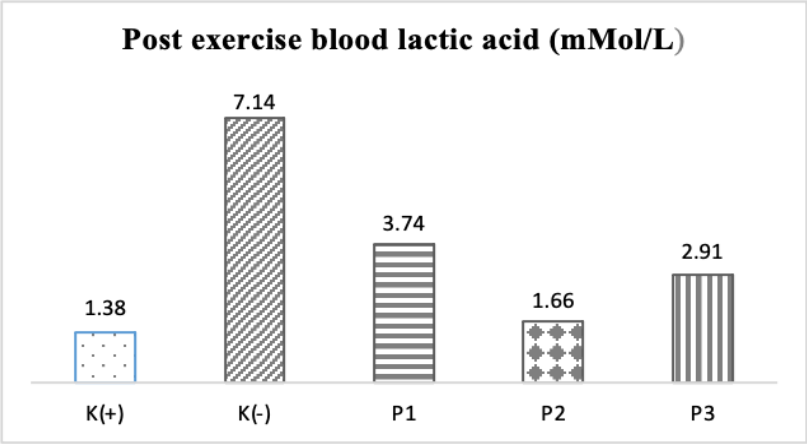

Figure 1. Comparison of mean lactic acid levels

Table 2. Mean difference lactic acid levels

Groups	Mean Difference (mMol/L)	IK 95%		*p**
		Minimum (mMol/L)	Maximum (mMol/L)	
C (+) vs C (-)	-5.76	-6.03	-5.49	0.00
C (+) vs P1	-2.35	-2.62	-2.08	0.00
C (+) vs P2	-0.28	-0.54	-0.01	0.04
C (+) vs P3	-1.53	-1.80	-1.26	0.00
C (-) vs P1	3.40	3.13	3.67	0.00
C (-) vs P2	5.48	5.21	5.75	0.00
C (-) vs P3	4.23	3.96	4.50	0.00
P1 vs P2	2.08	1.80	2.34	0.00
P1 vs P3	0.83	0.55	1.09	0.00
P2 vs P3	-1.25	-1.52	-0.98	0.00

* *Post hoc LSD test* (p<0,05)

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The largest mean difference between the groups was between C (+) and C (-), with both standard comparisons being under opposite conditions, resting and receiving a swim test. The largest inter-group difference between treatment groups was between P1 and P2. The P2 juice dose was twice the juice dose on P1 and the result was posted swimming lactic acid production was suppressed by more than 50%. The smallest difference was between groups P1 and P3. With the addition of 0.27 g of sugar, the P3 group produced less lactic acid than P1 after swimming. The addition of 0.27 g of granulated sugar has a protective effect on lactic acid production after swimming, as the addition of sugar increases the availability of glucose for energy. However, the effect was not as good as a double dose of juice.

DISCUSSION

Muscle fatigue occurs more quickly in high-intensity and short-duration sports that require rapid energy over a short period, while oxygen uptake is very limited. This condition causes the metabolism to produce a byproduct called lactic acid. Lactic acid, which accumulates, inhibits muscle contraction and causes pain. This condition is known as muscle fatigue.[18]

The results of this study show that the administration of a combination juice at different doses to Sprague Dawley rats caused a significant difference in the mean blood lactic acid levels. The main difference was in C (+) and C (-) groups, both of which were the same control group that did not receive juice, but C (-) got three minutes of swimming activity while C (+) rested. The C (+) groups as the normal inactivity standard had a blood lactic acid level of 1.38 mmol / L. This value corresponds to the average lactic acid level of normal human blood under inactivity conditions of 1-2 mmol / L.[19] Meanwhile, C (-) group as the standard for full activity, which has the three-minute swim test without prior consumption, had blood lactic acid levels of 7.14 mmol / L.

In the treatment group, the P2 group with a juice dose twice the dose of P1 had the lowest blood lactic acid levels after exercise of all treatment groups. The lactic acid levels in the P2 group were even close to normal lactic acid levels such as C (+) under resting conditions and when the person during physical inactivity. This mean was not met by P1 and P3 groups, which means that the other two treatment groups have standard blood lactic acid levels as if they were active. Thus, it can be said that the dose of P2 has a very good protective effect in suppressing the production of lactic acid, a byproduct in the formation of energy in exercise with high intensity and short time where O2 supply for the metabolism is very limited. The P2 which administered with a combined juice of up to 3.6 g / 200 g body weight of rats had higher citrulline values than the P1 dose, which only administered of 1.8 g / 200 g body weight of rats or P3 dose that the combination juice was given 1.8 g / 200 g body weight rats with the addition 0.27 g sugar.

Citrulline is a type of amino acid that plays a role in improving athlete performance by suppressing lactic acid production. Citrulline is classified as an ergogenic non-essential amino acid involved in three metabolic pathways: detoxification of ammonia in the urea cycle, synthesis of glutamine to arginine in the gut and kidneys, and synthesis of nitric oxide.[20] During the formation of ATP for energy production in intense exercise, dehydrates AMP (adenosine monophosphate) produces ammonia. Ammonia activates phosphofructokinase and facilitates the production of lactic acid.[21] Citrulline detoxifies ammonia so that lactic acid production can be controlled.[22] Citrulline accelerates the process of elimination of ammonium and lactate in plasma and other muscle metabolites.[23] Watermelon is known to have a fairly high content of citrulline, especially in the mesocarp. Seedless yellow watermelon varieties have a higher citrulline content than the other varieties. Administering 2 g of yellow watermelon mesocarp extract can reduce lactic acid production.[24]

In P1 and P3 groups, citrulline contained in the combined juice was relatively equal, as the groups which received a juice dose of 1.8 g per 200 g of rat body weight respectively. The difference was 0.27 g of sugar was added to the P3 group. The results of blood lactic acid levels of both groups after exercise showed that P3 was better than P1. It because 0.27 g of sugar was

added to the P3 group, which means that sugar absorption was increased. The addition of a simple sugar intake increases the energy source that is ready to be used in the body for activity. The availability of more ready-to-use energy sources increases the ability to perform longer activities so that muscle fatigue does not occur in a short period.[25] Previous studies have also shown a significant difference in the lactic acid levels in the blood of football players before and after given brown sugar. The treatment group showed a decrease in blood lactic acid level before and after administration of brown sugar from 11.5 mmol / L to 7 mmol / L. In the control group, there was an increase in blood lactic acid levels from 9 mmol / L to 10.8. mmol / L.[26]

This study used a combination of yellow watermelon and plantain. Juicing with a combination of two types of fruits was intended to increase nutritional value. The role of plantains in contributing to carbohydrates and high potassium. While the role of the yellow watermelon is to contribute carbohydrates and citrulline. The function of potassium as an electrolyte regulates the pH balance, the cofactor of the enzyme pyruvate kinase, Na + K + -ATPase which plays a role in energy production, synthesis of glycogen. In addition, potassium can also help train muscles and prevent muscle spasms and cramps.[12] Giving bananas before exercise greatly prevents muscle fatigue in the anaerobic phase.[27] Meanwhile, citrulline have beneficial functions accelerating metabolite waste removals such as ammonium and lactate from the plasma and the other results of muscle metabolism so the muscle fatigue does not occur quickly.[3]

The dose of combination juice was supplemented varied in each group. The doses were given according to watermelon and banana household standards consumption, 100 g each fruit. The dose was converted into mice with a conversion number of 0.018. In this study, the P2 group which received the juice dose of 3.6 g / 200 g body weight showed the lowest blood lactic acid production. It showed that the dose of combination juice made from 100 g of yellow watermelon and 100 g of plantain is most effective in suppressing lactic acid production in the blood during high-intensity short-term activities.

CONCLUSION

The combination juice of yellow watermelon-plantain affected suppressing the production of post-exercise blood lactic acid. The best dose was 3.6 g per 200 g rats body weight. It produced optimal suppression of post-exercise lactic acid production with the lowest blood lactic acid levels as if there was no activity.

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REFERENCES

1. Kemenkes RI. Pedoman gizi olahraga prestasi. Kementrian Kesehatan RI: Jakarta. 2014.
2. Fink HH, Mikesky AE. Practical applications in sports nutrition. Jones & Bartlett Learning; 2017.
3. Meneguello MO, Mendonca JR, Lancha Jr AH, Costa Rosa L. Effect of arginine, ornithine and citrulline supplementation upon performance and metabolism of trained rats. Cell Biochem Funct. 2003;21(1):85–91.
4. Curis E, Nicolis I, Moinard C, Osowska S, Zerrouk N, Bénazeth S, et al. Almost all about citrulline in mammals. Amino Acids. 2005;29(3):177.
5. Pérez-Guisado J, Jakeman PM. Citrulline malate enhances athletic anaerobic performance and relieves muscle soreness. J Strength Cond Res. 2010;24(5):1215–22.
6. Glenn JM, Gray M, Wethington LN, Stone MS, Stewart RW, Moyen NE. Acute citrulline malate supplementation improves upper-and lower-body submaximal weightlifting exercise performance in resistance-trained females. Eur J Nutr. 2017;56(2):775–84.
7. Jayaprakasha GK, Patil BS. A metabolomics approach to identify and quantify the phytochemicals in

watermelons by quantitative 1HNMR. Talanta. 2016;153:268–77.
8. Ridwan R, Abdul Razak HR, Adenan MI, Md Saad WM. Supplementation of 100% flesh watermelon [Citrullus lanatus (Thunb.) mats. And nakai] juice improves swimming performance in rats. Prev Nutr Food Sci. 2019;24(1):41–8.
9. Rimando AM, Perkins-Veazie PM. Determination of citrulline in watermelon rind. J Chromatogr A. 2005;1078(1–2):196–200.
10. Tarazona-Díaz MP, Alacid F, Carrasco M, Martínez I, Aguayo E. Watermelon juice: potential functional drink for sore muscle relief in athletes. J Agric Food Chem. 2013;61(31):7522–8.
11. Kemenkes RI. Daftar Komposisi Bahan Makanan. 2008.
12. McKenna MJ, Bangsbo J, Renaud J-M. Muscle K+, Na+, and Cl− disturbances and Na+-K+ pump inactivation: implications for fatigue. J Appl Physiol. 2008;104(1):288–95.
13. Ustafia L, Tursilowati S, Noviardhi A. Pemberian Milkshake Pisang Sebelum Latihan terhadap Kelelahan dan Tekanan Darah Atlet Sepak Bola. J Ris Gizi. 2017;5(2):41–7.
14. Faturochman, Said Junaidi AS. Journal of Sport Sciences and Fitness DI SEMARANG. 2020;5(2):104.
15. Irawan MA. Nutrisi, Energi & Performa Olahraga. Polt Sport Sci Perform Lab. 2007;1:1–13.
16. Cutrufello PT, Gadomski SJ, Zavorsky GS. The effect of l-citrulline and watermelon juice supplementation on anaerobic and aerobic exercise performance. J Sports Sci. 2015;33(14):1459–66.
17. Mooren F, Völker K. Molecular and cellular exercise physiology. Human Kinetics Publishers; 2005.
18. Williams MH. Nutrition for health, fitness, and sport. WCB/McGraw-Hill; 1999.
19. Janssen Peter GJM. Training lactate pulse rate. Oule Finl Polar ElectroOy. 1989;
20. Rabier D, Kamoun P. Metabolism of citrulline in man. Amino Acids. 1995;9(4):299–316.
21. Tomlinson C, Rafii M, Ball RO, Pencharz P. Arginine synthesis from enteral glutamine in healthy adults in the fed state. Am J Physiol Metab. 2011;301(2):E267–73.
22. Takeda K, Machida M, Kohara A, Omi N, Takemasa T. Effects of citrulline supplementation on fatigue and exercise performance in mice. J Nutr Sci Vitaminol (Tokyo). 2011;57(3):246–50.
23. Aranta Galuh Fridintya AGF. Perbedaan Efektivitas Pemberian Jus Pisang Ambon dan Jus Pisang Raja dalam Mengatasi Kelelahan Otot pada Tikus Wistar. Diponegoro University; 2011.
24. Adnyana IK, Arlinda ND, Safitri D. EFEK ANTILELAH EKSTRAK AIR MESOKARP SEMANGKA KUNING (Citrullus lanatus Thunb.) TANPA BIJI. Kartika J Ilm Farm. 2014;2(2):1–6.
25. Hairy J. Fisiologi Olahraga. Jakarta: Dirjen Dikti; 1989.
26. Abdurahman MN, Ray HRD, Ruhayati Y. Potensi Gula Merah dalam Meningkatkan Penampilan Olahraga. J Terap Ilmu Keolahragaan. 2018;3(1):24–9.
27. Kumairoh S, Syauqy A. Pengaruh pemberian pisang (Musa paradisiaca) terhadap kelelahan otot anaerob pada atlet sepak takraw. Diponegoro University; 2014.

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Effect of tomato and red guava juice on blood glucose level in overweight woman

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ABSTRACT

Background: Based on the results of Riskesdas 2018 that the prevalence of diabetes mellitus in Indonesia showed an increase from 6.9% to 8.5%. Overweight is closely related to impaired blood glucose, insulin resistance, and decreased insulin secretion. Preventive efforts that have been made using non-pharmacological treatments, such as increasing the intake of fiber and lycopene from fruits.
Objectives: This study aimed to determine the effect of tomato juice and guava juice on blood glucose levels in overweight women.
Materials and Methods: The study design is a true experiment conducted in 11subjects in treatment group and 11 subjects in control group. The subject of this study were overweight adult women 45-55 years. Blood sampling was taken in the morning, then glucose levels were measured using the GOD-PAP method (Glucose Para Amino Phenazone). We gave 600 ml of tomato juice and red guava every day for 21 days. To find the effect of juice on fasting blood glucose levels controlled by nutrient intake and physical exercise using the Repeated Measure ANOVA test.
Results: In the treatment group, there was a decrease of 3.24 mg/dl in blood glucose levels; while, in the control group, the decrease in blood glucose levels was only 0.26 mg/dl. However, we found no statistically significant differences in both groups.
Conclusions: Consumption of tomato juice and red guava reduced fasting blood glucose.

Keyword: Blood glucose; Tomato; Red guava; Women; Overweight

BACKGROUND

At the age of 45, people experience an increased risk of developing diabetes and glucose intolerance due to decreased ability of beta cells to produce insulin whose function is to metabolize glucose[1]. Females are more prone to diabetes due to estrogen. As estrogen decreases, insulin resistance begins to arise which causes an increase in blood glucose[2]. Overweight is closely related to several conditions, including impaired blood glucose, disrupted balance of glucose and insulin, and decreased insulin metabolism. Obesity causes an increase in fatty acids or Free Fatty Acid (FFA) in cells and this could lead to insulin resistance[3].

People suffering from DM will depend on drugs throughout their life. Anti-hyperglycemia drugs cause side effects such as nausea, weight gain, diarrhea, and bloated stomach which lead to non-adherence medication[4]. Therefore, we need an alternative that does not have side effects, which is by utilizing fruit with a low glycemic index. Fruit is a safe alternative because it is part of the diet.

Tomato contains an active substance called lycopene. Lycopene is an antioxidant whose ability is to resist free radicals. Lycopene can lower blood glucose by reducing insulin resistance, leading to cell tolerance to glucose increases, and excess blood sugar levels can be overcome[2].

Red guava is a fruit with high vitamin C content and pectin-type fiber. Pectin is hypoglycemic which can reduce blood glucose levels as pectin plays a role in the formation of gels in the gastrointestinal tract. Vitamin C plays a role in reducing glucose toxicity, thereby preventing a decrease in β cell mass and insulin level.

In this research, 300 grams of tomatoes and 300 grams of red guavas were processed into juice. This combination is expected to reduce blood glucose levels in overweight females aged 45 - 55 years.

MATERIALS AND METHODS

This research implemented a true experimental design with randomized pre-test and

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post-test control group design. It was conducted in RW 02 Pedurungan Tengah, Semarang. The research subjects were grouped randomly. The sample size was calculated using the hypothesis test formula on two independent groups, obtaining 11 subjects in the treatment group and 11 subjects in the control group. The subjects involved were females aged 45-55 years with a BMI ≥ of 23 kg/m^2, not taking medication, and had never been diagnosed by a doctor with diabetes mellitus. This study has been approved by Health Research Ethics Committee of Health Polytechnic of Semarang Number 113/EA/KEPK2019.

The subjects in the treatment group had their blood glucose levels measured before and after giving tomato and red guava juices. While the subjects in the control group had their blood glucose levels measured, but not receiving tomato and red guava juices. Blood sampling was carried out in the morning after the subjects were asked to fast for 8 hours. Measurement of fasting blood glucose levels was done using the GOD-PAP method (Glucose Oxidase Para Amino Phenazone). The research variables included the independent variable of tomato and red guava juice provision. Tomatoes and red guavas, each at 300 grams, were juiced together by adding 150 ml of water. The treatment group was then given the juice to consume 2 times a day, in the morning and evening, with each administration of 300 ml for 21 days. The dependent variable was fasting blood glucose.

The instruments used consisted of sample identity form, data collection form to record food intakes, product acceptability test form, juice consumption adherence form, and physical exercise form. To measure food intakes, the 2 x 24-hour food recall method was used to determine energy and carbohydrate intakes. Meanwhile, a food frequency questionnaire (FFQ) was used to determine fiber, vitamin C, and lycopene intakes. Juice consumption adherence was measured by asking the subjects how much juice was consumed. Microtoice was used for height measurement and digital weighing was used for weight measurement. To determine the effect of tomato and guava juice on fasting blood glucose levels before and after the research, and analysis was performed using the Repeated Measure ANOVA test, with $\alpha = 0.05$.

RESULTS

Table 1 shows that the research subjects were aged between 45 - 55 years. The average BMI in the treatment group was 28.26 kg/m^2 and the control group was 29.2 kg/m^2. The average fasting blood glucose levels in the treatment and control groups before treatments were 84.77 mg/dl and 85.66 mg/dl, respectively. The average fasting blood glucose levels after treatments in the treatment and control groups were 81.53 mg/dl and 85.4 mg/dl, respectively. The average energy adequacy levels in the treatment and control groups were 69.12% and 52.81%, carbohydrate intakes were 54.53% and 42.2%, fiber intakes were 10.55 g and 8.08 g, lycopene intakes were 2.42 μg and 1.21 μg, and vitamin C intakes were 57.92 mg and 45.98 mg.

Table 1. Research Subject Characteristics

Variable	*Treatment*			Control			
	Mean±SD	*Min*	*Max*	*Mean±SD*	*Min*	*Max*	*P-value*
Age	49.67±3.24	45	55	50.67±3.96	45	55	0.29
BMI	28.26±3.76	23.1	34.7	29.22±4.3	23.1	35.3	0.57
Initial GDP	84.77±13.11	63.6	103.5	85.66±13.67	66.3	114.0	0.86
Final GDP	81.53±11.46	63.6	104.0	85.4±6.66	72.5	92.6	0.31
Energy (%)	69.12±25.69	26.6	117.8	52.81±19.1	28.5	91.2	0.47
Carbohydrate (%)	54.53±20.32	25.2	90.2	42.2±13.1	28.5	91.2	0.41
Fiber (g)	10.55±2.98	4.8	14.1	8.08±2.25	4.8	11.8	0.47
Lycopene (μg)	2.42±1.04	0.8	4.2	1.21±0.51	0.8	2.5	0.10
Vit C (mg)	57.92±15.4	34.8	78.9	45.98±9.55	34.7	60.9	0.10

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Table 2. Differences in Blood Glucose Levels between Treatment and Control Groups

	Between Treatment *Mean ±SD*	After Treatment *Mean ±SD*	Difference	P-value
Control group	85.66 ± 13.67	85.40 ± 6.66	-0.26	0.533
Treatment group	84.77 ± 13.11	81.53 ± 11.46	-3.24 ± 6.85	0.438

Based on the results of analysis presented in table 2, there was a decrease in fasting blood glucose levels for subjects in the treatment group by 3.24 mg/dl and in the control group by 0.26 mg/dl. The statistical test on the difference in decreased fasting blood glucose levels obtained a p-value of 0.438 in the treatment group and a p-value of 0.533 in the control group. Therefore, it can be concluded that there is no difference in fasting blood glucose levels between the treatment and control groups before and after treatments. It is in line with research conducted by Widyasulistya (2018) where there was no significant difference between blood glucose levels before and after treatments of giving guava juice for 3 days to football athletes[5].

Table 3. Repeated Measure ANOVA Multivariate Test

Variable	P-value
Kec_Energy	0.299
Kec_Carbohydrate	0.472
Kec_Fiber	0.370
Kec_VitC	0.293
Groups	0.521

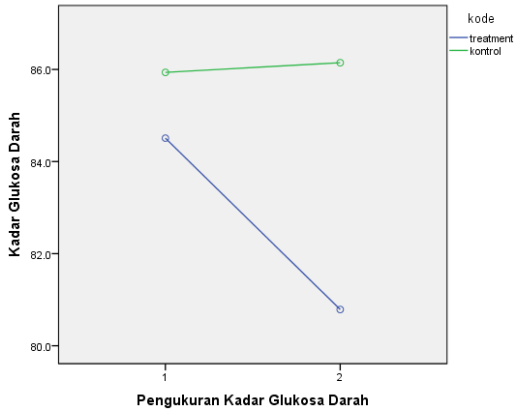

Figure 1. The difference in Fasting Blood Glucose Levels between Treatment and Control Groups

This test's results showed that there was no significant difference (p = 0.521) in blood glucose levels between the treatment group and the control group, although clinically there was a decrease in blood glucose levels in the treatment group by 3.24 mg/dl. Meanwhile, the level of adequate energy, carbohydrate, fiber, and vitamin C did not affect changes in blood glucose levels. As shown in Figure 1, there was a decrease in the treatment group after being given tomato and guava juice.

DISCUSSION

Tomato and red guava juices were given 2 times a day, in the morning and evening, as much as 300 ml per administration. This mixed juice contains soluble fiber such as pectin, lycopene, and vitamin C, with each contribution of 96% of lycopene, 78% of fiber, and 121% of vitamin C. The analysis showed that there was no effect of giving tomato and red guava juices on blood glucose levels in overweight adult females aged 45-55 years (p = 0.521). The level of energy, carbohydrate, fiber, and vitamin C intakes did not

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statistically have a significant effect on changes in blood glucose levels. The results of this research are in line with that of Ayuhapsari et al (2018) where they found that energy, carbohydrate, and fiber intakes did not affect changes in fasting blood glucose[6].

A factor that resulted in the non-significant effect of treatments in this research was the age of the subjects over 45 years. At that age, there will be an increase in blood glucose due to changes in pancreatic beta cells that produce insulin, resulting in a decreased insulin production. The decrease in insulin production results in a reduced amount of glucose that enters cells, so glucose remains in the blood vessels and causes increased blood glucose level[2].

The nutritional status of research subjects could cause less effective effects of treatments, with the research subjects' average BMI of 28.26 kg/m^2 included in the category of obesity level I. Obesity is related to conditions of impaired blood glucose, disrupted balance in glucose and insulin, and decreased insulin metabolism[3].

As seen in Table 1, the average fasting blood glucose levels before treatments in the treatment group were 84.77 mg/dl, and after treatments were 81.53 mg/dl. It showed a decrease of 3.24 mg/dl, but the decrease was still in the category of normal blood glucose level. Whereas in the control group the average fasting blood glucose levels were 85.66 mg/dl and went down to 85.40 mg/dl. There was only a decrease of 0.26 mg/dl. This showed that consumption of 600 ml of tomato and red guava juice for 21 days could reduce blood glucose levels by 3.24 mg/dl and/or maintain blood glucose levels in the normal category.

Changes in fasting blood glucose levels between the treatment and the control groups could be caused by the consumption of tomato and red guava juice. Tomato contains lycopene whose function is anti-diabetic by increasing the concentration of insulin[2]. Lycopene can lower blood glucose levels by inhibiting glucose absorption in the intestine, increasing glucose transport in the blood, stimulating glycogen synthesis, and inhibiting glucose synthesis. Lycopene prevents degenerative diseases such as diabetes through an oxidative mechanism. Lycopene binds to reactive oxygen and increases antioxidant potency, thereby reducing oxidative damage to lipids. Meanwhile, the non-oxidative mechanism of lycopene is done through gene function which improves gap-junction communication, hormone modulation, and immune response, all of which can reduce degenerative diseases such as DM[7].

Research conducted by Astuti and R (2018) revealed that provision of tomato juice to blood glucose levels in pre-diabetes as much as 200 ml for 21 days showed a decrease in blood glucose by 9.00 mg/dl[2]. Another research conducted by Yusni (2015) showed that the administration of tomato extract and mangosteen extract to diabetic mice each 50 mg/kg bb/day for seven days showed a decrease in blood glucose levels by 56.67%[7]. Research by Chairunnisa (2012) was conducted by giving tomato paste to diabetic mice as much as 62 mg for 7 days reduced blood glucose levels by 262 mg/dl with a decreasing percentage reaching 75.60%[8]. Another research conducted by Wuryaningrum (2016) on giving 250 ml of rainbow smoothies to type 2 DM patients for 10 days showed statistically significant results and decreased blood glucose levels by 43.75 mg/dl[9].

Changes in fasting blood glucose levels could be caused because of guava consumption. Guava contains water-soluble fiber, such as pectin, and vitamin C. Pectin can reduce blood glucose levels because it is hypoglycemic and its physiological function is to increase glucose tolerance in people suffering from DM[10]. Pectin plays a role in gel formation because there is a reaction of fiber with water in the gastrointestinal tract. This gel will slow down gastric emptying and the movement of food through the upper gastrointestinal tract and inhibit the mixing of the gastrointestinal contents with digestive enzymes so that there is a reduction in the absorption of nutrients in the proximal part. This inhibition affects the slow absorption of glucose, causing a decrease in blood glucose[11,12]. Pectin can envelop carbohydrate molecules so that they inhibit carbohydrate absorption and will be released slowly so that the amount of sugar that enters the blood is reduced and an increase in excess blood glucose levels can be avoided[13].

Vitamin C contained in red guava is higher than in orange, which is at 49 mg per 100 grams[14]. Vitamin C can increase insulin sensitivity and lower blood glucose levels by reducing glucose toxicity, preventing decreased beta-cell mass, and increasing the amount of insulin. These three mechanisms of vitamin C protect against organ damage in diabetes: vitamin C as an antioxidant, vitamin C inhibits intracellular sorbitol

accumulation, vitamin C reduces protein glycosylation[15].

Vitamin C plays a role in modulating insulin action in people suffering from diabetes and is associated with lowering blood glucose levels. Vitamin C can inhibit the accumulation of sorbitol caused by hyperglycemia through the polyol-sorbitol pathway. Vitamin C acts as an inhibitor of the aldolase reductase enzyme which converts glucose in cells into sorbitol, so it can prevent sorbitol buildup and reduce oxidative stress and improve endothelial function. The buildup of sorbitol in cells or tissues can cause damage or change in functions[16,17]. Research conducted by Santi (2013) revealed that the provision of red guava juice as much as 2 gr/head/day for 10 days to hyperglycemic mice resulted in decreased blood glucose levels by 164.5 gr/dl[13].

CONCLUSIONS

Consumption of tomato and red guava juice did not significantly reduce blood glucose levels, with a p-value > 0.05. Consumption of tomato and guava juice can be used as part of a daily diet to control blood sugar within the normal range.

REFERENCES

1. Betteng R, Pangemanan D, Mayulu N. Analisis Faktor Resiko Penyebab Terjadinya Diabetes Melitus Tipe 2 pada Wanita Usia Produktif Dipuskesmas Wawonasa. J e-Biomedik. 2014;Volume 2,(2):9.
2. Astuti YD, Murwani H. Pengaruh Pemberian Jus Tomat Terhadap Kadar Glukosa Darah Pada Prediabetes. J Nutr Coll. 2013;2:111–7.
3. Nabila R, Widyastuti A, Murbawani EA. Hubungan Lingkar Pergelangan Tangan Dengan Kadar Glukosa Darah Wanita Obesitas Usia 40-55 Tahun. J Nutr Coll [Internet]. 2018;7(2):92–9. Available from: http://ejournal3.undip.ac.id/index.php/jnc/
4. Eliana F. Penatalaksanaan DM Sesuai Konsensus Perkeni 2015. Satelit Simp. 2015;1–7.
5. Widyasulistya R, Rahmawati AY, Aristiati K. Pengaruh Pemberian Jus Buah Jambu Biji (Psidium Guajava L) Terhadap Kelelahan Otot Anaero. J Ris GIZI. 2018;40–7.
6. Ayuhapsari N, Wijaningsih W, Jaelani M. Efektifitas Pemberian Buah Apel Manalagi dengan Dosis Bervariasi terhadap Penurunan Kadar Glukosa Darah pada Penderita Diabetes Mellitus tipe II di Kelompok PROLANIS Puskesmas Genuk Kota Semarang. J Ris GIZI. 2018;13–8.
7. Yusni, Akbar IB, Rezania, Fahlevi R. Penurunan Kadar Gula Darah Akibat Pemberian Ekstrak Manggis (Garcinia mangostana) dan Tomat (Lycopersicum esculentum Mill) pada Tikus Diabetes Blood Sugar Levels Reduction by Mangosteen (Garcinia mangostana) and Tomato (Lycopersicum esculentum Mill. J Glob Med Heal Commun. 2017;5(1):57–63.
8. Chairunnisa R. Pengaruh Jumlah Pasta Tomat Terhadap Penurunan Kadar Gula Darah Pada Mencit Diabetes. J Teknol Ind Pertan. 2012;1–12.
9. Wuryaningrum E, Jaelani M, Sunarto. Efektifitas Rainbow Smoothies Terhadap Penurunan Kadar Gula Darah Pasien DM Tipe 2 Di Klub Senam Prolanis Bina Sehat Mandiri Cepu. J Ris GIZI. 2016;1–6.
10. Sunarti. Serat Pangan Dalam Penanganan Sindrom Metabolik. Yogyakarta: Gajah Mada University Press; 2018.
11. Lianah, Tyas DA, Armanda DT, Setyawati SM. Aplikasi Umbi Suweg (Amorphophallus campanulatus) sebagai Alternatif Penurun Gula Darah pada Penderita Diabetes Mellitus. J Biol Appl Biol. 2018;1(1):1–12.
12. Saputro PS, Estiasih T. Pengaruh Polisakarida Larut Air (PLA) Dan Serat Pangan Umbi-Umbian Terhadap Glukosa Darah. J Pangan dan Agroindustri. 2015;3(2):756–62.
13. Santi DA. Efek Jus Buah Jambu Biji Terhadap Gangguan Toleransi Glukosa Pada Tikus Putih Jantan Akibat Efek Samping Deksametason. J Ilm Mhs Univ Surabaya [Internet]. 2013;2(1):1–19. Available from:

http://webhosting.ubaya.ac.id/~journalubayaac/index.php/jimus/article/view/169

14. Astuti EP, Ismawati, Suwarlim T. Uji Efek Penurunan Glukosa Darah Air Perasan Buah Jambu Biji (Psidium guajava L) pada Mencit (Mus musculus) yang diberikan Glukosa per Oral. J Ilm Kesehat. 2009;3(2):107–13.

15. Santosh HN, David CM. Role of ascorbic acid in diabetes mellitus: A comprehensive review. J Med Radiol Pathol Surg [Internet]. 2017;4(1):2–3. Available from: http://www.jmrps.net/eJournals/ShowText.aspx?ID=79&Type=FREE&TYP=TOP&IN=_eJournals/images/JPLOGO.gif&IID=13&Value=1&isPDF=YES

16. Dini CY, Sabila M, Habibie IY, Nugroho FA. Asupan Vitamin C Dan E Tidak Mempengaruhi Kadar Gula Darah Puasa Pasien DM Tipe 2. Indones J Hum Nutr. 2017;4(2):65–78.

17. Ermawati T. Hubungan kadar glukosa darah dengan Pertumbuhan A. actinomycetemcomitans Pada Tikus Diabetes Melitus. J Kedokt GIGI. 2012;9(1):19–22.

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The implementation of nutrition improvement programs for underweight children, wasting and stunting in the Department of Health, Central Buton district, Southeast Sulawesi

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ABSTRACT

Background: Health status can affect the Human Development Index (HDI) of a country. To improve the quality of human resources, the Indonesian Government has applied various policies, such as the nutrition improvement program, since there are still lots of toddlers suffering from underweight, wasting dan stunting. Even one out of three toddlers in Indonesia were detected stunting.
Objectives: This study described the implementation of a nutrition improvement program for toddlers in the Central Buton District Health Office.
Materials and Method: This was a qualitative study that involved ten informants. Three of those informants are nutritionists in the District Health Office, Head of Public Health Department, Head of NutritionDepartement. Also, two nutrition workers in primary healthcare centers and two mothers of malnourished toddlers.
Results: These findings showed that in terms of input, trained human resources on nutrition were lacking, and the budget for the nutrition improvement program was inadequate. While, in the process, all implementors had done very well, although they still had no collaboration across sectors. In the output aspect, the health status of underweight, wasting, and stunting toddlers improved. Also, monitoring and evaluation were conducted on toddlers registered at the integrated service posts in 2018.
Conclusions: There were still constraints on the input, process, and output aspects, even though there was an increase in the nutritional status of children under five, but nutrition problems for children under five in Central Buton Regency were still high.

Keywords: Nutritional improvement program; Toddlers; Implementation

BACKGROUND

Nutritional problem is a problem in the life cycle, starting from pregnancy, infants, toddlers, teens, to the elderly. The nutritional problem can occur in all age groups, even nutritional problems in a certain age group will affect nutritional status in the next life cycle (intergenerational impact).[1]

Indonesia nowadays is still faces nutritional problems such as underweight, wasting, and stunting, this can affect the quality of human resources, because growth constraints during toddlers have the potential to experience non-communicable diseases in adulthood.[2] According to the President of the Republic of Indonesia, the fulfillment of nutrition is one of the best long-term investments that can be made, apart from requiring skilled human resources, of course, Indonesia also needs healthy human resources. The health status of a nation can affect the human development index (HDI), therefore the government is committed to making efforts to overcome various nutritional problems by issuing various policies on nutrition improvement.[3]

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Toddler age is a period where the process of growth and development occurs very rapidly, with high activity and learning changes. At this time the child's brain develops rapidly up to 80%, so it requires sufficient nutrient intake in both quantity and quality, if the nutritional intake is not fulfilled, the physical and intellectual growth of toddlers will experience disruption, as a result, toddlers become lost generation, and can adversely affect the country due to the lack of quality human resources.[4]

In 2017, the number of children under five in Indonesia is around 22.4 million every year, there are at least 5.2 million women in Indonesia who are pregnant, so the average number of baby-born every year is 4.9 million, and baby with short birth conditions have the potential to the increase in the number of children under five with stunting, whereas in 2018 there were 17.8% of children under five suffering from malnutrition, 12.7% among under-five children with malnutrition, 12.7% of whom were short, for that intervention to reduce stunting would reduce under-nutrition Malnutrition).[5]

Stunting is a chronic nutritional problem caused by a lack of nutritional intake for a long time, characterized by a lack of height from the standard children in general, this occurs since the child is still in the womb until the age of 2 years or the first 1000 days of life and the child is stunted after 2 years of age cannot be changed anymore so what can be done is to maximize the potential for brain development. In contrast to stunting, malnutrition takes place in a shorter time, that is, when the child experiences normal growth up to a certain age, then there is a change in diet which causes the toddler to no longer getting enough intake, as a result, the child loses weight in a short time but the recovery also fast. Apart from stunting and malnutrition, There is also malnutrition, namely children under five who are underweight children of their age. Another nutritional problem is obesity or excess nutrition. Nutritional status is an important indicator for the health of children under five whose physical impact is measured anthropometry and categorized based on WHO standard with index weight/age, height/age, and body weight/height.[6]

The problem of child nutrition in Indonesia is very worrying because 1 in 3 Indonesian children is stunted, nationally the proportion of stunting among toddlers is 37.2% in 2013 to 30.8%, in 2018, the prevalence of under-nutrition is 19.6% years 2013 to 13.8% in 2018, the prevalence of malnourished toddlers is 5.7% in 2013 to 3.9% in 2018, the prevalence of underweight children is 12.1% in 2013 to 10.2% in 2018 while obese toddlers are 11, 9% in 2013 to 8.0% in 2018, this is what then causes Indonesia to face a double burden. Although nationally from 2013 to 2018 there has been a decrease in the prevalence of nutritional problems in children under five, there are still gaps between provinces, especially the problem of stunting.[7]

The prevalence number of stunting at the provincial level is still very high, where 2 provinces have a stunting prevalence >40%, 18 provinces have a stunting prevalence of 30-40% and 23 provinces including Southeast Sulawesi have a stunting prevalence of 20-30%, and only DKI Jakarta has a stunting prevalence < 20%. Based on the results of monitoring the national nutritional status of Southeast Sulawesi in 2017, the highest stunting problem was in Central Buton Regency, namely 48.8% under five, 25.9% underweight, and 13.3% underweight. Also, the frequency of visits to children under five to the posyandu is decreasing along with the increasing age of the children, from 12,060 the number of children under five, with the coverage of under-fives weighed as much as 8,786 (73%) and 3,247 (27.14%) under-fives whose body weight is not monitored.[8]

Toddlers as the assets of our future (nation) must receive optimal nutritional care and attention as a series of individual and community nutritional needs through prevention, improvement, healing, and recovery efforts carried out in the community and health service facilities, including involving related sectors.[9] The central government gives authority to the provincial government which then the provincial government delegates to the city/regency government to be responsible for implementing efforts to improve nutrition. Based on the results of the field study, Central Buton is one of the districts in Southeast Sulawesi which is responsible for implementing the nutrition improvement program. The programs that have been implemented include monitoring the growth of toddlers at the health center, providing additional food, and tracking cases of toddler nutrition, this is done based on the regulation by the Indonesian Minister of Health no. 23 of 2014, to improve food consumption patterns following balanced nutrition improving behavior awareness of nutrition, physical activity, and health.[10,11] Based on this situation, the authors are interested in describing the implementation of the toddler nutrition improvement program which consists of early detection of the status of children under five years old in the

Central Buton District through system elements consisting of Input, Process, and Output.

MATERIALS AND METHODS

This research is an observational study with a qualitative descriptive design, which was conducted in September 2018 - August 2019. The subjects of this study were nutrition staff and mothers of malnutrition children under five years old in the work area of the Health Office of Central Buton District, Southeast Sulawesi Province. The selection of informants was carried out by using the purposive sampling technique by observing. Data collection methods were obtained through in-depth interviews, observation, and documentation of 10 informants consisting of 3 main nutrition staff informants (IU1, IU2, IU3) of the Health Office, and data validity was carried out on 7 triangulation informants consisting of the Head of the Department Health (IT1), Head of Public Health (IT2), Head of the Family Health and Nutrition Section (IT3), 2 Nutrition Implementers of Puskesmas (IT4, and IT5), and 2 mothers under five years old with malnutrition (IT6, and IT7). The data collected includes system elements in a program policy consisting of input elements which include human resources, infrastructure, and funds used in implementing the nutrition improvement program for children under five years old, especially monitoring the nutritional status of toddlers, next is the process element which includes planning, organizing, implementing, and monitoring evaluation. In addition to the two elements of input and process, there is also a third element, namely output as a reference to determine the success of the nutrition improvement program for children under five at the Health Office of Central Buton District, Southeast Sulawesi Province, where the nutritional status of children is assessed based on the Body Weight (BW) / Age (A) index which is categorized as undernutrition (Underweight), good nutrition and more nutrition. Body Height (BH) / Age (A) children were categorized as very short, short, and normal. Furthermore, the weight/height of children is categorized as very thin, thin, normal, and obese. This is based on the z-score according to the standard deviation value of growth according to the World Health Organization (WHO). Data analysis techniques in this study were carried out with data collection, data reduction, data display, and conclusion.

RESULTS

Central Buton Regency is one of the regencies in Southeast Sulawesi which consists of 7 districts. The Health Office is one of the elements implementing government affairs that is tasked with helping the regent carry out government affairs, especially in the health sector, this is stated in the Central Buton Regent Regulation No.13 of 2018 concerning the Position of the Organizational Structure, Duties and Functions and Work Procedure of the Health Service of Central Buton Regency. The Health Office consists of several fields, one of which is the public health sector, including a section of nutrition and family health which is responsible for running community nutrition improvement programs and carrying out coaching and cooperation with health centers in implementing community nutrition programs. maternal and child health services as well as other tasks related to family health and community nutrition. In this study, the authors focus on discussion with several aspects by a systems approach to be able to get an overview of the implementation of early detection programs and monitoring of the nutritional status of toddlers because a program will run well if it meets the target input, process and output indicators.

Input

1. Human Resources

The results of the interview on the implementation of the nutrition improvement program for children under five years old at the Health Office of Central Buton District based on the input element seen from the perspective of human resources (HR), that those involved in the nutrition improvement program were the Head of the Public Health Sector as the person in charge of the program, the head of the Family Health and Community Nutrition section. A program manager, and nutrition staff serving as report compilers and field technical advisors, and health center nutrition implementing staff (TPG) as executors of program activities. Human resources are people who are responsible for and coordinate the implementation of a program. Following are the results of interviews about human resources:

Based on the results, it was found that the nutrition staff of the Health Office was 3 people, 2 were civil servants (PNS), but one of them had never attended training on nutrition because only 6 months had worked as a nutrition staff at the health office and 1 person was still as an intern, the head of the family health and nutrition section that has an educational background (Diploma/DIII) in Midwifery with a long term of 4 years in the health department. The head of the public health sector has an educational background (Diploma/DI) Nutrition department, has served for 2

years, and the Head of the Health Office with a 2 years term. Nutrition staff at the Health Office can be said to be still lacking because at one time the nutrition staff who were still as an intern to become the person in charge of the nutrition improvement program, and the nutrition staff could get over-work. Besides, there is nutrition staff who have never attended training, especially training on nutrition during their time as nutrition staff at the Health Office.

IU1: the staff is still lacking, because we are only three of us, 1 is still an apprentice, but has worked 4 years and has attended training on nutrition, and 2 people are civil servants (PNS), but one of them has only worked for 6 months and has never attended any training while working at the health office, especially regarding nutrition, so sometimes we feel overwhelmed if suddenly there are reports of cases of malnutrition at the same time from various sub-districts, and the three of us and have to do tracking, some are doing intervention, field technical guidance and making nutrition reports to be accountable to during the evaluation meeting.

IT3: actually still lacking, because usually the person holding and responsible for one of the programs is an apprentice staff, as well as providing field technical guidance.

So it can be concluded that the nutrition staff at the Central Buton District Health Office in terms of quality and quantity is still not under the Minister of Health Regulation No. 26/2013 concerning the Implementation of Work and Practices for Nutritionists in Service Centers in section 17 states that nutrition workers in implementing nutrition services in health service facilities have the authority to participate in education, training, research and development of nutrition services.[12]

2. Facilities and infrastructure

Based on the results of interviews and observations of facilities and infrastructure, there were no obstacles in the provision of infrastructure because, in terms of the procurement of all supporting facilities, they were well maintained and available at the Health Office and the Puskesmas. The tools available consist of anthropometric tools, toddler weight scales, writing instruments, digital weighing devices, and height measurement tools, KMS (public health) books, recording and reporting forms, technical instructions for program implementation, additional food and medicines (vitamin A), which was available at the posyandu. Also, the District Health Office validates health service facilities based on a predetermined operational permit. Validation is done by comparing the suitability of the condition of the medical equipment facilities and infrastructure needed in the field and those available in health service facilities. Following are the results of interviews with key informants and triangulation.

IU2: for the too-provision by the government (Dinas) then distributed to each puskesmas, however, there must be a report if the puskesmas needs/changes equipment, and if food for toddlers is malnutrition or lacking, usually the Health Office provides instant food such as biscuits, while the puskesmas works together Posyandu cadres provide local food, porridge, eggs and so on, and there are no obstacles in the provision of facilities and infrastructure at the Health Office.

IT2: anthropometric equipment is usually the agency that provides according to the needs of the puskesmas, and so far there have been no obstacles in the procurement of sarpras because there is always coordination and validation of medical devices in health services between the office and the puskesmas.

3. Fund

One of the components of the resources needed in organizing a health program is health financing or funds. Based on the results of interviews from the three nutrition staff at the Health Office, it was stated that the nutrition improvement program had been budgeted for in the Central Buton Regency (APBD), but because of the reduction or rationalization of the APBD, not all activities from the nutrition improvement program received a budget, such as socialization on nutrition problems for children under five years old such as stunting. the stakeholders who have an important role in the success of the program cannot be done because the funds have not been programmed and are not sufficient. Following are the results of interviews with key informants and triangulations.

Process

1. Planning

Planning is the basis of the process of implementing a program, so it must be formulated and conveyed so that the results are following the desired objectives.

The planning process for the nutrition improvement program for children under five at the Health Office of

Central Buton District consists of human resource planning, implementation plans, budget plans, and monitoring and evaluation plans. Following are the results of interviews with informants.

Based on the results of the interview and data from budget planning book-keeping that there is a harmony between the statements of the main informant and the triangulation informant, it can be said that planning on the implementation of the nutrition improvement program for children under five years old has been carried out by the Health Office of Central Buton District so that program implementation can run well and achieve maximum results.

2. Organizing

Organizing is the process of preparing an organizational structure under the goals of the organization, its resources, and the environment. Organizing is very necessary to facilitate the implementation of programs that have been previously formulated in planning. The organization of the nutrition improvement program for children under five years old at the Health Office of Central Buton District is described in several stages, namely the division of tasks, the appointment of a coordinator, and the activities to be carried out. Following are the results of interviews with key informants and triangulation

IU3: for the organizational structure, the implementation of the nutrition improvement program at the health office is in accordance with the organizational structure of the head of the service, in charge of the community health sector, coordinating the family health and nutrition section and nutrition staff as executors and monitoring whether or not the program is running at the puskesmas level because it is the party fully implementing it puskesmas, assisted by posyandu cadres because the target is toddlers with nutritional problems. The activities are monitoring the growth of children under five, providing additional food, and nutrition counseling. Most of the activities are carried out at the posyandu, besides that, the puskesmas collaborates with villages/sub-districts to provide additional food so that nutritional problems such as malnutrition, malnutrition and stunting are reduced.

IT4: because the service as a monitor is not the implementer, so we always involve the puskesmas in its implementation, starting with the posyandu cadres, who report a problem to the puskesmas, then the puskesmas reports the problem to the health office, the office makes an activity plan citing the problem, then then reported to the government, in order to get support from both material and non-material.

3. Implementation

An implementation is an act of striving for all members involved in the program to achieve predetermined goals by their respective main tasks in planning. In the program for the implementation of nutrition improvement activities in the working area of the Health Office of Central Buton District, monitoring the growth of children under five at the posyandu, providing additional feeding for toddlers, and tracking cases. Following are the results of interviews with key informants and triangulation

IU1: It is still lacking because the nutrition improvement program has a variety of activities, so we are smart about using it, if the priority is usually we still try to do it with these funds, but we cannot do socialization so that we cannot work together with other sectors. outside of health to solve nutritional problems under five years old.

IT2: It is enough but there is still more supervision for funding program implementation, because there is a reduction or rationalization of the APBD Budget.

IU1: this nutrition improvement activity is carried out to reduce nutritional problems and improve nutritional status, so it must involve many people such as the head of the public health, head of health and nutrition, nutrition staff, health center nutrition officers, medical personnel, cross-program medics and posyandu cadres, all sources the power to work together to improve the nutritional status of toddlers, such as pmt in posyandu, toddler nutrition screening or toddler nutrition tracking, For our own budget plans, for example for next year's budget, we must input this year's report, even then the funds we include in the activity report, are usually not suitable with the funds we get, then again we determine priority activities then the budget is put in dpa, while monitoring is carried out every quarter.

IT1: us and the puskesmas, and the community are trying to coordinate so that the implementation of nutrition improvement programs and other programs runs well according to the desired results

IT3: nutrition staff as the person in charge of the program in the department, nutrition implementing staff at the health center in charge of the puskesmas level, cadres, medical personnel, environmental health, for budget planning, the range I forget to look at later in the bookkeeping, because it is the nutrition staff who compile the budget. We plan to evaluate the evaluation once a year, because we also do monitoring every three months, so from monitoring we can correct any mistakes in implementation.

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From the interview result with the nutrition staff as the main informant (IU1) and health center nutrition executives as triangulation informants (IT4), it was stated that the implementation of monitoring of the nutritional status of children under five was carried out guided by posyandu cadres under the auspices of the puskesmas. In addition, the next activity is the selection of cases of malnutrition, along with the results of interviews with nutrition staff at the health office (IU2), health center nutrition staff (IT4), and mothers of malnourished children under five years old (IT7). In addition to monitoring at the posyandu, there was also a screening to find out malnourished toddlers at the posyandu, puskesmas, and even home visits intending to find cases of malnutrition under five years old by measuring body weight according to age (BW/A) and weight according to height (BW/BH), as well as the presence of clinical signs such as abdominal disease. Also, the program for the improvement of nutrition for children under five is providing additional food for children under five. Following are the results of interviews with nutrition staff from the health office (IU3), TPG nutrition at the puskesmas (IT5), and mothers of malnourished children under five (IT7).

IU1: For nutritional status monitoring, it is done at the posyandu, if the office is just waiting for a report from the puskesmas if cases of malnutrition are found, so we only monitor what the puskesmas do.

IT4: We do monitoring the nutritional status of toddlers with cadres, like weighing them, because from weighing we can know whether there is a nutritional problem or not, but for malnutrition, we monitor it once a week by making home visits for 3 months.

4. Evaluation Monitoring

Monitoring and evaluation are carried out to determine performance in program implementation and to find out how the achievements of activities that have been implemented. The evaluation referred to in this paper is the evaluation and monitoring by the health office of the puskesmas as the field implementer in the nutrition improvement program for children under five at the Health Office of Central Buton District. Following are the results of interviews with key informants and triangulation informants.

From the statement of the nutrition staff as the main informant (IU1) and the head of the health department as the triangulation informant (IT1), that monitoring is carried out to determine the course of an activity, to prevent and minimize problems in the field, apart from monitoring the Health Office also evaluates the implementation of nutrition improvement programs. Following are the results of interviews with key informants and triangulation informants.

IU2: usually also done during the inspection at the posyandu, at the puskesmas, and even at home visits to find cases of malnutrition under five years old, usually due to lack of nutritious food intake and congenital diseases.

IT4: We conduct home visits if there are reports of cases of malnutrition, from posyandu cadres, for the counseling to be conducted every two months, even nutrition cases can also be obtained from the puskesmas, when a child being examined for health at the puskesmas is suspected of suffering from malnutrition.

IT7: So every time you weigh it at the clinic, your body weight does not increase, sometimes it goes down from the previous scale, you were also sick and then you were taken to the health center.

IU3-DKK: pmt is given on the guidance by cadres who work closely with the health center nutrition officers, but the materials are from the office such as biscuits, then the service distributes them to each puskesmas, later the puskesmas will be distributed at the posyandu, but usually for the posyandu we always recommend pmt in the form of local food.

IT5-PKM: after giving the pmt, we usually conduct counseling first to mothers of toddlers with nutritional status problems, to always provide food that contains energy and protein, as well as for the pmt that we provide if possible are sufficient for children who suffer from nutritional problems every day, and do not eat it by his brother or other people. Because pmt is only given to children who have nutritional status problems such as malnutrition, malnutrition, and stunting. But for toddlers with malnutrition cases once a week we check and monitor the child's weight at home for 90 days, besides that we are very helpful because the village is also involved in holding pmt.

IT6-IB: Every week the health worker visits the house, the child's weight is continuously measured, and given food, biscuits, packaged baby porridge, milk.

From the interview's result with nutrition staff at the Health Office as the main informant (IU3) and the head of the family health and nutrition section as triangulation informants (IT3), it was stated that the evaluation at the Health Office of Central Buton District was carried out to determine the performance achievements of activities.

DISCUSSION

1. Human Resources

Human resources (HR) is an important aspect of implementing a program. Based on the results of the research, in carrying out the nutrition improvement program for children under five, it is carried out by the public health sector, especially the family health and nutrition section of the Health Office of Central Buton District, but in its implementation, it involves the puskesmas and cadres, who are tasked with carrying outweighing activities for children under five at posyandu, providing additional food, tracking the nutrition of children under five, as well as other activities related to nutrition problems. Also, the results of the research show that the Health Office has limited human resources in the nutrition improvement program, both in terms of numbers, where there are only 3 nutrition staff, namely 2 civil servants (PNS), and 1 apprentice staff. If this is not done yet, it can become an obstacle in implementing the nutrition improvement program for children under five at the Health Office of Central Buton District.[12] Based on previous research, there was a lack of health personnel resources at the Health Office of Central Buton District, especially nutrition staff, and there were still health centers that did not have nutrition workers, as well as inadequate access to roads so that they encountered obstacles in both the networking and tracking malnutrition activities.[13]

IU1: for field monitoring we usually do it by monitoring the results of reports from the puskesmas, from here we can see the progress of activities in the field, and if there are obstacles, while for monitoring at the service level we usually hold a meeting every quarter to find out about program progress and solve problems. which is a barrier to implementation

IT1: supervision is carried out in stages, from the head of the department, head of the field, head of sections and programmers, so that each of them reports the course of activities and the results obtained, and is reported at the periodic meeting every quarter.

IU-3: evaluation is carried out once a year, namely at the end of the year, if there are things that have not been achieved then we will go to the field to see, what are the obstacles, such as yesterday's coverage of under-fives weighing is still below the national target, so we are working with the health center to carry out a sweeping of under-five weighing each village / kelurahan in their respective puskesmas area, by mobilizing cadres to take notes and toddlers who do not participate in posyandu, so that at the next posyandu they can attend the posyandu

IT3: usually at the end of each year we hold an evaluation meeting to find out the results of the implementation of the program that has been running for almost a year, and compare whether the results obtained are on target, and take corrective action if the results are not on target, so that the following year can reach the target .

2. Facilities and infrastructure

Apart from competent human resources, supporting facilities and infrastructure are also needed in the implementation of nutrition improvement program activities for children under five. Based on the results obtained, the facilities and infrastructure for the nutrition improvement program for children under five in the working area of the Health Office of Central Buton District are complete, both at the puskesmas and posyandu, such as anthropometric tools, tables for posyandu implementation activities, besides that the posyandu also involves the role of the village in providing materials local food for supplementary feeding (PMT) in the form of eggs and green bean porridge, this is very influential in the success of the implementation of the nutrition improvement program for children under five. Although the number of active posyandu in Central Buton District decreased from 91 posyandu in 2017 to 85 (62%) active posyandu in 2018, However, the level of awareness of mothers to invite their children to posyandu increased from 73% in 2017 to 85.7% in 2018, this can be a picture of the maximum implementation of several priority posyandu programs, especially nutrition improvement programs. In line with previous research that posyandu is expected to be able to organize five priority programs, namely maternal and child health, family planning, nutrition improvement, immunization, and diarrhea prevention. Availability of adequate facilities and infrastructure, optimal management, and utilization can help achieve the success of a program. In line with previous research that posyandu is expected to be able to organize five priority programs, namely maternal and child health, family planning, nutrition improvement, immunization,

and diarrhea prevention. Availability of adequate facilities and infrastructure, optimal management, and utilization can help achieve the success of a program. In line with previous research that posyandu is expected to be able to organize five priority programs, namely maternal and child health, family planning, nutrition improvement, immunization, and diarrhea prevention. Availability of adequate facilities and infrastructure, optimal management, and utilization can help achieve the success of a program.[14]

3. Fund

After competent human resources and complete infrastructure are needed, funds are also needed to support the implementation of a policy or program, because it will become complicated when realizing policy objectives when the funds needed are not sufficient to finance the entire series of activities of a program. In the implementation of the program to improve malnutrition, malnutrition, and stunting at the Health Office of Central Buton District, it is known that some program implementers have stated that the funds are not fully sufficient to carry out innovative activities such as cross-sector socialization, making posters and campaigns on improving nutrition, especially stunting toddlers. Based on secondary data, funds for the nutrition improvement program for children under five at the central Buton District Health office are around Rp. 10.100,000.00 in 2018, the lack of funds could be one of the causes for not maximizing the achievement of health development targets in the Central Buton District. Following previous research, although the availability of health service Facilities continues to increase, if this availability is not supported by the quality of health service in terms of human resources and financial support, it can result in the implementation of policies/programs being hampered in achieving goals because insufficient funds will affect the quality implementation of programs in the community.[15]

4. Planning

Based on the previous research, planning must be clear, both in terms of budget, human resources, and implementation targets according to the target, such as the activity plan for the empowerment team for family welfare is always synergized with the work plan and budget for regional work units.[16] The Health Office of Central Buton District has established a concept to minimize the problem of nutritional status in toddlers, especially stunting, by determining the structure of the program implementation where the person in charge of the program is the nutrition staff at the health office, and the person in charge at the puskesmas level is the puskesmas nutrition officer. In this case, the health office nutrition staff is tasked with providing technical field guidance and making reports to be accountable to the section head, while the puskesmas is in charge of executing activities and goes directly to the community, together with health or community cadres who have been empowered and become partners for the puskesmas as a community mobilizer around them to care about the health and take part in the utilization of the health services provided, Also, other cross-programs are also involved, such as medical personnel, paramedics, environmental health and community health centers, but this has not been involved in other sectors. The target in implementing the program is toddlers with nutritional problems, as for the target plan, which is to reduce underweight toddlers to 5.3%, wasting toddlers 2.7%, and stunting toddlers 14% in 2019.

For budget planning is prepared a year before the implementation of the program carried out by the nutrition staff of the health office then the budget activity plan (RKA) is submitted to the government, usually the budget that comes out is less than the planned budget, after which the budget is reported in the form of a budget utilization document (DPA).) by the nutrition staff of the Health Office of Central Buton District. The Monitoring Plan will be carried out every quarter and evaluation is carried out once a year, namely at the end of the activity or towards the end of the year. To get maximum activity results, planning must be formulated clearly so that the process runs systematically, in other words, managerial planning will provide a comprehensive perspective on the activities to be carried out, both resources, time, or implamentation.[17]

5. Organizing

Based on the results of the research on the organization of the nutrition improvement program for children under five at the Health Office of Central Buton District, it is described in several stages, namely the division of tasks, the determination of the coordinator, and the activities to be carried out. The nutrition improvement program for children under five at the Health Office of Central Buton District is carried out following the existing organizational structure at

the Health Office and in its implementation, it involves various parties, both from the scope of the health office and from the scope of the village / kelurahan. The nutrition improvement program that is implemented as the policyholder is the Head of the Health Office of Central Buton District, then the authority for its implementation is handed over to the public health sector which houses 3 sections, namely, the family health and nutrition section, the health promotion section, and the environmental health section.

Then regarding the division of tasks and responsibilities in implementation, it is entirely up to the puskesmas because there are already nutrition officers or regional supervisors whose job is to monitor the development of nutrition in their area. Based on the policy of the Health Office of Central Buton District government that all health workers at the health center, whether midwives, nutrition workers, or other health workers must cooperate in efforts to treat cases of nutrition in children under five, both in the form of screening and tracking, but the Health Office has not been able to cooperate with other people. other sectors/stakeholders, due to the lack of funds for socialization and advocacy.[18]

6. Implementation

The implementation of the program is one indicator of the success of a program because the success of the program will be obtained if the planned program can be implemented properly. The implementation of the nutrition improvement intervention program for malnourished children under five, malnutrition and stunting at the Central Buton district health office is by monitoring the nutritional status of children under five at the posyandu, screening for cases of malnourished children under five, and providing additional food for malnourished toddlers. From the results of the study, it was found that growth monitoring at the posyandu consisted of weighing children under five, measuring the height of toddlers which was carried out every month by posyandu cadres together with puskesmas officers, consisting of midwives, nutrition implementing officers, and nurses, giving vitamins and medicines, supplementary feeding for toddlers, as well as monitoring the growth of children under five with stunting, wasting, underweight to prevent and improve the nutritional status of children under five. Growth monitoring is a follow-up to policies and programs at the community level so that people have the power and efforts to address health problems so that nutritional problems can be resolved properly. Apart from that, the selection is also carried out to trace cases of malnourished toddlers, so that all mothers of toddlers routinely take their children to the posyandu. The implementation of screening is carried out in two stages, the first is that the screening is carried out every two months at the posyandu, secondly, the screening is carried out at the health center when the toddler visits the health center to have his disease checked and the health worker knows that the toddler is suffering from malnutrition

To fulfill and improve the nutritional status of children under five, nutrition officers together with posyandu cadres provide additional food for toddlers. The categories for under-nutrition and stunting children were given additional food in the form of biscuits which were given by cadres at the posyandu, while toddlers with cases of malnutrition were given factory food, biscuits, milk, eggs, and local food if there was assistance from the village / kelurahan. as well as educating mothers of toddlers on providing nutritious foods both at the community center at their home. Apart from that, nutrition workers at the health center, as well as from the local government office, also conduct home visits to malnourished toddlers every week for 3 months to find out the progress and monitor the growth of toddlers by measuring BH, BW, Lila, and head circumference of toddlers, as well as monitoring the environment where toddlers live. [19]

7. Evaluation monitoring

Monitoring evaluation is very necessary so that the implementation stage can run well and on target according to the program plan. Based on the results of the study, it was found that in the nutrition improvement program for children under five, the health department also carried out a monitoring stage which was carried out every quarter, monitoring was carried out by the health office based on the activity report of the puskesmas because from these reports the health department nutrition staff could find out and supervise the program activities and prevent any deviations in the field. If irregularities are found during supervision, the service office can make corrections as soon as possible so that activities can return to the predetermined path. The monitoring stages are also leveled from the posyandu level, the puskesmas level, and the health department level. Monitoring by the Health Office can be carried out in 2 stages, namely direct visits or by observing the results of the activity report. Monitoring is the process of observing activities on an ongoing basis to minimize any deviations because

if there are deviations, modifications or changes are needed so that implementation remains on the planned path.[20,21]

In addition to monitoring, the Health Office also conducts an evaluation every once a year, the evaluation is carried out at the end of the year, but if there is an extraordinary event (KLB) in an area, we usually do an evaluation as well. Evaluation is carried out in meetings and meetings at the Health Office, where at the meeting each program holder describes the results of the achievement of the implementation of activities as well as obstacles during program implementation. The evaluation of the results obtained from the implementation of the nutrition improvement program for children under five at the Health Office of Central Buton District consisted of the results of monitoring the status of toddlers at the center, the results of the screening of cases of malnutrition, the results of the provision of additional food for under-nutrition, malnutrition, and stunting in the district. Central Buton, If there are indicators that have not met the target, an evaluation meeting is held at the health department level, to find out the obstacles and how to overcome them so that the target year can be achieved in the next implementation. The health office as the holder of the authority for the nutrition improvement program plays a full role in observing all existing activities in the field by continuing to coordinate with the puskesmas and cadres so that problems that occur can be minimized properly.[22] Evaluation is an orderly and systematic process of comparing the results achieved with the benchmarks set criteria, then drawing conclusions based on suggestions and input from each program implementer, because it will be difficult to know the extent to which the objectives of the planning are achieved if no evaluation is carried out.[23]

Output

Nutrition status monitoring activities are actively carried out by nutrition officers at the health center through the weighing month for under-fives which is carried out twice a year. The state of poor nutrition will reduce the body's resistance which will cause children to get sick easily, resulting in death. Prevention of cases of malnutrition, deficiency, and stunting is carried out by providing additional food (PMT) which is funded through the Central Buton Regency APBD and Southeast Sulawesi Provincial APBD, PMT which is given in the form of milk, MP-ASI biscuits, and milk porridge. Almost all of the cases assisted had gained weight. Prevention of malnutrition under five who require treatment is carried out at the health center, but if there is a congenital disease, the toddler is referred to the Central Buton Hospital.

Nutritional status	2017	2018
Underweight	25.9%	17%
Stunting	48.8%	28%
Wasting	13.3%	9.5%

Based on the results of research and primary data, it is revealed that all toddlers with nutritional problems have received 100% treatment, so there is an increase in the nutritional status of toddlers in 2018, which is marked by a very significant decrease in the problem of nutritional status of children from 2017-1018. In addition, there was an increase in the coverage of under-fives weighing, where the frequency of under-five visits to posyandu in 2017, from 12,060 the number of toddlers, with the coverage of under-five weighed as much as 8,786 (73%) and 3,247 (27.14%) under-fives whose body weight was not monitored, while in 2018 The number of toddlers weighed was 10,870 children with a community participation level of 85.7% of the results obtained, exceeding the national target that has been set, namely 80%, and exceeding the scope of achievement in 2017.[10]

CONCLUSIONS

The nutrition improvement program for children under five in Central Buton District still has several unfulfilled aspects, such as a lack of human resources in quantity and quality, a lack of budget for nutrition improvement programs, which is only around Rp. 10,100,000.00 in 2018, lack of socialization, especially stunting across other sectors, as a result, the problem of stunting in Central Buton Regency is the highest in the Southeast Sulawesi province in 2018, for that the local government needs to make even better efforts.

There is still a need for training for nutrition staff at the Health Office and Nutrition Implementers at health centers, need to increase health funds from the APBD for health development programs, especially community nutrition, need cross-sector and cross-program cooperation in reducing nutrition problems in children under five in Central Buton district, and increasing utilization and socialization of cadres to mothers of children under five so that local communities have higher motivation in efforts to

improve health status both within the family and in the community.

REFERENCES

1. Mitra. Permasalahan Anak Pendek (Stunting) dan Intervensi untuk Mencegah Terjadinya Stunting (Suatu Kajian Kepustakaan). J Kesehat Komunitas. 2015;2(5):254–61.
2. Renyoet BS, Nai HME. Estimasi potensi kerugian ekonomi akibat wasting pada balita di indonesia. J Gizi Indones. 2019;7(2):127.
3. Kementerian Kesehatan Republik Indonesia. Bersama Cegah Stunting. 2018;(Warta Kesmas Tahun 2018).
4. Khoeroh H, Indriyanti D. Evaluasi Penatalaksanaan Gizi Balita Stunting. Unnes J Public Heal. 2017;6(3).
5. Kementerian Kesehatan Republik Indonesia. Gizi, Investasi Masa Depan Bangsa. 2017;(Warta Kesmas Tahun 2017).
6. Zilda O, Sudiarti T. Faktor Risiko Stunting pada Balita (24-59 Bulan) di Sumatera. J Gizi dan Pangan. 2013;8(3):175–80.
7. Badan Penelitian dan Pengembangan Kesehatan. Kementerian Kesehatan. Has Utama Riskesdas 2018. 2018;
8. Gizi Masyarakat D, Jenderal KM D, Kesehatan RI K. Hasil Pemantauan Status Gizi (PSG) Tahun 2017. Buku saku pemantauan Status Gizi. 2017;
9. Rosha CH B, Sari K, Yunita SP I, Amaliah N, Utami N. Peran Intervensi Gizi Spesifik dan Sensitif dalam Perbaikan Masalah Gizi Balita di Kota Bogor. Bul Penelit Kesehat. 2016;44:127–38.
10. Dinas Kesehatan Kabupaten Buton Tengah. Profil Kesehatan Kabupaten Buton Tengah Tahun 2018. 2019;
11. Menteri Kesehatan Republik Indonesia. Peraturan Menteri Kesehatan Republik Indonesia Nomor 23 Tahun 2014. 2014;
12. Peraturan Menteri Kesehatan RI No 26 tahun 2003. Penyelenggaraan Pekerjaan dan Praktik Tenaga Gizi. 2013.
13. Sugiarto A, Retnaningsih C, Indrayati Y. Pendayagunaan Bidan Sebagai Tenaga Kesehatan Di Puskesmas dalam pelaksanaan kebijakan upaya perbaikan gizi pada balita di Kabupaten Buton Tengah Provinsi Sulawesi Tenggara. Soepra J Huk Kesehat. 2018;4(1):1–17.
14. Boli EB, Baliwati YF, Sukandar D. Komitmen Politik dan Peluang Pengembangan Kebijakan Gizi Pemerintah Daerah Provinsi Nusa Tenggara Timur Political Commitment and Opportunity to Advance Nutrition Policy in East Nusa Tenggara Province. J MKMI. 2018;14(4):351–9.
15. Subarsono. Analisis Kebijakan Publik. Pelajar P, editor. Yogyakarta; 2012.
16. Tomi S, Dian H, Soemarno. Evaluasi Program Perbaikan Gizi Masyarakat (Penanggulangan Gangguan Akibat Kekurangan Iodium) di Kota Malang. J Pembang Dan Alam Lestari. 2016;7(1):44–51.
17. Azwar A. Pengantar Administrasi Kesehatan. Bina Rupa Aksara, editor. jakarta; 2012.
18. Sutisna. Manajemen Kesehatan. UNS, editor. surakarta; 2009.
19. Febry F. Jurnal Ilmu Kesehatan Masyarakat. 2014;3(November 2012).
20. Sulaeman S E. Manajemen Kesehatan Teori dan Praktik di Puskesmas Yogyakarta. Yogyakarta: Gadja Mada University Press; 2011.
21. Wijayanti T, Niken P. Studi Kualitatif Proses Asuhan Gizi Terstandar Di Ruang Rawat Inap Rs St. Elisabeth Semarang. J Nutr Coll. 2013;2(1):170–83.
22. Sawitri N. Pelaksanaan Program Perbaikan Gizi Masyarakat Oleh Dinas Kesehatan Kota Pekanbaru. J JOM Fisip. 2018;Vol. 5 No.(1):Hal : 1-15.
23. Sarwono J. Metode Penelitian Kuantitatif dan Kualitatif. In: Graha Ilmu. Yogyakarta; 2006.

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Profile of nutritional status, energy availability, haemoglobin levels and bone density in *santriwati* (Islamic female student) with chronic energy deficiency risk

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ABSTRACT

Background: Santriwati (Islamic female student), women of reproductive age, were susceptible to experienced Chronic Energi Deficiency (CED). CED reflects the low energy availability of someone who can risk reducing bone density. **Objectives**: This study aimed to analyze the differences in body mass index, body fat percentage, hemoglobin levels, energy availability, and bone mineral density of female students who experienced CED risk and not experienced CED risk.
Materials and Methods: The research design was a cross-sectional study, with 101 female students as subjects who were selected by random sampling. The research was conducted from February to March 2019 at the Kyai Galang Sewu Islamic Boarding School, Semarang. CED risk data was taken using the upper arm circumference measurement. Percent body fat and BMI data were taken using BIA. Energy availability data is obtained from the difference between energy intake (energy intake) and energy output (energy expenditure through physical activity) divided by Fat-Free Mass (FFM). Energy intake data was taken using the SQ-FFQ questionnaire, and energy expenditure was calculated using the 24-hour activity record form. Anemia data were collected using strip hemoglobin measurements. Bone density data were taken using the Osteosys Sonost 3000 densitometer. Bivariate analysis used the Independent T-Test.
Results: A total of 57.2% of subjects experienced anemia. Subjects who had underweight nutritional status were 20.8%. Santriwati experienced osteopenia as much as 13.9%. There was no difference in bone density and hemoglobin levels between female students who were at risk of CED and not CED risk ($p > 0.05$), but there were differences in energy availability, body fat percentage, BMI between those at risk of CED and not CED risk ($p < 0.05$)
Conclusion: subjects at risk of CED (Lila <23.5 cm) had lower energy availability, body fat, and BMI than subjects who were not at risk of CED.

Keywords: Anemia; Body fat; BMI; Bone Density; CED risk; Santriwati

BACKGROUND

The number of women of reproductive age (WRA) in Indonesia has the largest number in Southeast Asia with 65 million.[1] Nutritional problem in WRA in Indonesia is still high. One of the nutritional problems that often occur in WRA is Chronic Energy Deficiency (CED). CED is a condition when a person suffers an imbalanced nutrient intake (energy and protein) that lasts for a long time.[2] The prevalence of CED risk on non-pregnant Indonesian women in 2018 based on the Mid-Upper Arm Circumference (MUAC) indicator is 14.5%.[3] The prevalence of CED risk on WRA, 15-19 years old, has increased from 30.9% to 36.3%, and the risk on WRA 20-24 years old also increased from 28.2% to 23.3% in 2007-2018.[3]

One of the WRA groups in Indonesia that tends to have nutritional problems is *santriwati*, female students in Islamic schools. A study conducted at the Al-Hidayah Islamic Boarding School in Grobogan Regency showed that 51.1% of the students were malnourished caused by insufficient nutritional intake.[4] Other research at Slafiyah Kauman Pemalang Islamic Boarding School also showed that 38% of santriwati were malnourished.[5] Body Mass Index (BMI) is used as the indicator to assess the nutritional status, where the body weight (kg) is divided by the square of height (m^2). BMI is

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a valid predictor and is widely used around the world.[6]

Another nutrition issue that always occurs in santriwati is anemia. Studies conducted in Islamic Boarding Schools in Indonesia showed that some Santriwati have low hemoglobin. For example, is in Husada Islamic Boarding School, 83.90% of santriwati experience anemia.[7] In Darul Ulum Peterongan Jombang Islamic Boarding School showed that 57.5% of santriwati had anaemia.[8] The malnutrition and low hemoglobin status of santriwati were usually caused by low food intake both in quantity and quality, for example, is the research that was conducted in Kepung Kediri observed a difference between teenage girls who lived in boarding schools than at home. The study showed that more teenage girls who lived in boarding schools experienced anemia due to the lack of knowledge, parental control and the ability to provide healthy food.[9]

The low energy reserves and nutrients can cause CED in WRA during adolescence may continue to pregnancy and breastfeeding.[10] Energy availability represents the remaining energy in the body or energy reserves for metabolic processes so that it can carry out the physiological function properly.

Energy availability is the difference between energy intake and energy expended during activity (relative to fat-free mass).[11] The energy availability value of 30 kcal/kg FFM /day is the threshold of energy availability required for physiological body functions.[12] Low energy availability usually occurs in athletes. They have a high energy expenditure and are often not compensated with adequate food intake to maintain their body weight.[13]

Aside from the abovementioned problems, a health problem that often arises in women of reproductive age is low bone density. Women were more likely to have a four-time risk of developing osteoporosis than men.[14] Research conducted by Ria Stella on bone density in young adult women showed that nutritional status and body weight had a significant association with bone density. Low bone density was associated with a lower percentage of body fat in subjects, where the body fat mass produces estrogen for the formation of bone mass.[15]

Based on these backgrounds, the researchers were interested in examining the differences between body mass index, percentage of body fat, hemoglobin levels, energy availability, and bone density in female students who are at risk for CED and those who are not at risk for CED.

MATERIALS AND METHODS

This study was conducted at Kyai Galang Sewu Islamic Boarding School, Semarang, in April 2019. This is an observational study with a cross-sectional design. The random sampling technique was used to select a total of 101 participants who fulfill the inclusion criteria. The inclusion criteria include, santriwati who were active at Kyai Galang Sewu Islamic Boarding School, did not suffer from infectious diseases (typhoid, chronic diarrhea, upper respiratory tract infections (URTI), TB, hepatitis, malaria, and dengue fever), did not consume any supplement or drug which may affect body composition, and willing to participate in this study.

For this study, the dependent variables are bone density, hemoglobin level, energy availability (EA), BMI, and body fat percentage. In comparison, the independent variable is chronic energy deficiency (CED) status, which is classified into at risk (CED group) and not at risk (normal group) of CED. The CED status was determined by measuring the participants' mid-upper arm circumference (MUAC) using MUAC tape. The measurement was performed on the left arm. For left-handed women, their right arm was measured. Wearing clothes as thin as possible, and if possible, rolling up the sleeves until the acromion is visible. Mark the acromion and the olecranon process, then mark the mid-point between these marks. Wrap a MUAC tape around the mid-point mark. The participants were categorized as at risk of CED if the mid-upper arm circumference is less than 23.5 cm.[3]

The bone density was measured using a densitometer from Osteosys Sonost 3000 with a quantitative ultrasound method. The bone density was normal if the T-score $-1 \leq SD < 2.5$, the participants were categorized as osteopenia if the T-score is $-2.5 \leq SD < -1$ and osteoporosis if the T-score was < -2.5.[16]

The hemoglobin level was determined by the hemoglobin test strip. The assessment was carried out by drawing blood from the participants to stain the strip before putting the stained strip into a hemoglobin meter from the Hemocue brand. The normal hemoglobin for women is 12 g/dL; anemia is defined by the hemoglobin of less than 12 g/dL.[7]

The BMI is calculated as weight divided by the square of the height in meters. The participants' body height was measured using a microtoise or stature meter to an accuracy of 0.01 cm. the participants stood barefooted and bare of any accessory on their heads.[17] The body weight was measured using a BIA Tanita DC 360 scale. The participants are categorized as underweight if the BMI was <18.5 kg/m^2, normal if the BMI was 18.5-24.9 kg/m^2, and overweight if the BMI was 25-29.9 kg/m^2; and obese if the BMI was ≥30 kg/m^2.

To calculate the body fat percentage, a body composition measurement using BIA Tanita DC 360 was conducted. Previously, the result of the height measurement from the stature meter was put into BIA Tanita DC 360. Twenty-four hours before the data collection, participants were recommended to drink at least eight glasses of water in a day and avoid caffeine and alcohol to achieve good hydration status.[18] The body fat percentage was classified into athletic (10-15%), good (16-19%), normal or acceptable (20-25%), overfat (26-29%), and obese (>30%).[19]

The energy availability was calculated from the difference between energy intake and energy expenditure divided by fat-free mass (FFM). The energy availability status was classified into low EA (<30 kcal/kg FFM/day), good (30-45 kcal/kg FFM/day), and high (>45 kcal/kg FFM/day).[20] The energy intake was obtained by analyzing 24 hour-food recall 6x24 hours using Nutrisurvey 2007 application. During physical activities within 24 hours, the energy expenditure was calculated using the following formula: 3.5 x bodyweight x Basal Metabolic Rate x time.[21] The fat-free mass was obtained from body composition measurement using BIA Tanita DC 360.

The univariate analysis was performed to describe each variable. The bivariate analysis using an independent t-test was performed to analyze the differences in bone density, hemoglobin level, BMI, body fat percentage, and energy availability among santriwati in the CED and normal groups. This study has been approved by the Health Research Ethics Committee (KEPK) Faculty of Medicine, Universitas Diponegoro and Central Public Hospital dr. Kariadi number 162/EC/KEPK/FK-UNDIP/V/2019.

RESULTS

Participants Characteristics

Table 1. Characteristics of participants

Variable	Median	Minimum	Maximum
Age (year)	20	18	24
Weight (kg)	46.7	32.9	67.5
Height (cm)	152.5	139	169.5
Body Mass Index (kg/m^2)	20	14	32
Mid-upper arm circumference (cm)	23.5	19	30.5
Body Fat Percentage (%)	27.3	16	44.9
Haemoglobin (g/dL)	11.6	6.3	15.1
Bone Density (T-Score)	-0.5	-1.6	1.8
Energy intake (kcal)	1310028.5	815 815	1601 0596
Energy availability (kcal/kgFFM/day)	1.9	1	9,6

Table 1 shows the distribution of the participants' characteristics, namely BMI, body fat percentage, mid-upper arm circumference (MUAC), bone density, and hemoglobin level. Some participants recorded a BMI of 14, while others recorded 32 kg/m^2. By the same token, the median hemoglobin level was 11.6 g/dL, but one of the participants recorded only 6.3 g/dL. The bone density among santriwati was relatively normal, although some recorded negative T-score, it was still within the normal range.

Table 2. Characteristics of participants based on nutritional status, anemia status, Chronic Energy Deficiency, and bone density

Category	n	%
Anemia Status		
Anemia	58	57.4
Normal	43	42.6
Chronic Energy Deficiency		
Risk	45	44.6
Normal	56	55.4
Body Fat Percentage		
Normal	68	67.3
Overfat	16	15.8
Obese	17	16.8
Body Mass Index for Age (BMI/U)		
Underweight	21	20.8
Normal	60	59.4
Overweight	11	10.9
Obese	9	8.9
Bone Density		
Normal	87	86.2
Osteopenia	14	13.9

Table 2 further displays the participants' characteristics. Based on hemoglobin assessment, 57.4% of the participants (n=58) were anemic. The nutritional status assessment using MUAC measurement reported 44.6% (n=45) participants were at risk of CED; meanwhile, 68.3% (n=68) had normal body fat percentage. Nutritional status based on BMI showed that 20.8% (n=21) of the participants were underweight. Most of the participants had a normal bone density, but 13.9% (n=14) had osteopenia.

Table 3. Difference of bone density, haemoglobin levels, Chronic Energy availability, body mass index and body fat percentage between CED risk and normal groups

Variable	CED risk (n=45) Mean ±DS	Normal (n=56) Mean ±DS	*P value*
Bone Density (T-Score)	-0.48±0.5	-0.4±0.56	0.458 [a]
Haemoglobin Levels (g/dL)	11.29±1.75	11.48±1.95	0.61 [a]
Energy Availibility (kcal/kgFFM/day)	1.77±0.78	3.98±2.74	<0.001[a*]
Body Mass Index (kg/m^2)	18.68±1.47	22.16±2.83	<0.001[a*]
Body Fat Percentage (%)	24.89±3.76	31.56±5.08	<0.001[a*]

[a]=Independent t-test *significant (p<0.05)

Based on table 3, there was a difference in energy availability between CED risk and normal groups (p<0.001). The mean of energy availability in the CED risk group was lower (1.77 kcal/kgFFM/day ±0.78) than the normal group (3.98 kcal/kgFFM/day ±2.74). Additionally, there was a difference in BMI between the normal and CED groups (p<0.001). The mean BMI in the CED group was lower (18.68 kg/m^2±1.47) than the normal group (22.16 kg/m^2±2.83). There was a difference in body fat percentage between the normal and CED groups (p<0.001). The mean body fat percentage in the CED group was lower (24.89 (%±3.76) than the normal group (31.56 (%±5.08). However, there were no differences in bone density and hemoglobin

levels among the CED and normal groups (p=0.458; p=0.61)

DISCUSSION

The santriwati at Kyai Galang Sewu Islamic Boarding School also receive normal education where most of the santriwati were college students. Based on the analysis, 44.6% of the participants were at risk of CED, determined by the MUAC measurement. MUAC is considered practical to use in the field to assess the risk of CED.[22] The study conducted at AL-Munawwir Islamic Boarding School Yogyakarta reported 26.7% of santriwati were at risk of CED.[23] Furthermore, most of the santriwati were anemic. Anemia among women of reproductive age may increase the risk of maternal death, low birth weight, prone to infection, miscarriage, and increase the risk of premature labor.[24] A report conducted at Salafiyah Syafi'iyah Islamic Boarding School Sukorejo, Situbondo, stated that 79% of the participants were anemic based on their hemoglobin level.[25] Nutritional status measurements based on BMI also indicated that 20.8% of participants in this study were underweight. Another study conducted at Al-Hidayah Islamic Boarding School, Grobogan District, reported that 51.1% of santri had low nutritional status.[4]

The Difference of BMI between Santriwati With and Without Risk of CED

There was a difference in BMI between the normal group and the CED group. It was indicated by the mean BMI in the CED group, which was lower compared to the normal group. Another study by Brito et al. supported this result with their report, which showed a significant association between MUAC and BMI. The lower the MUAC is, the lower BMI would be.[26] Another study in India registered a positive relation between MUAC and BMI among pregnant women.[6] BMI measurement is the most common measurement of nutritional status. Yet, due to the needs of several tools and skills, other means of measurement, such as MUAC, may be used to determine women's nutritional status. This measurement is considered practical and gives an overview of women's nutritional status well.[6] More articles showed an association between MUAC and BMI, which also explains the difference in BMI between santriwati with and without CED risk. The BMI of the CED group was lower than the normal group.

The Difference of Body Fat Percentage between Santriwati With and Without Risk of CED

Based on the analysis, there was a difference in body fat percentage between the CED and normal groups. This finding is supported by another study in India that reported a significant association between BMI and body fat percentage. The lower the BMI score is, the lower the body fat percentage would be.[27] BMI also gives an overview of CED in women.[28] It may explain the difference in the mean body fat percentage between the CED and normal groups. The body fat percentage of the CED group was lower than the normal group.

The Difference of Energy Availability between Santriwati With and Without Risk of CED

Based on the analysis, there was a difference in energy availability between the CED and normal groups. The mean score of energy availability in the CED group was lower compared to the normal group. The energy status assessment, such as energy availability measurement, is an accurate and objective measurement that can also reflect CED's compensation compared to using only nutritional status assessment.[29] The energy availability refers to the amount of energy leftover and available for body function after the energy expended for physical activities is subtracted from the energy consumed.[20] Low energy availability among santriwati was mostly caused by low energy consumption. Low energy reserves and nutrients may increase the risk of CED.[11] Loucks stated that low energy availability might cause loss of muscle mass or fat mass in the body. If it continues for a prolonged time, then physiological function may be compromised.[30] CED's risk increases if the depletion of muscle mass develops continually without adequate energy intake.[31]

The Difference of Haemoglobin Level between Santriwati With and Without Risk of CED

Based on the statistical analysis, there was no significant difference in CED and normal groups' hemoglobin levels. It contrasts with Pramodya et al. at Kediri District's findings, which reported a significant difference in hemoglobin levels between students with and without CED.[32] However, a report

by Dea Intartanti showed an insignificant association between nutritional status and anemia among female adolescents.[33] This may be since macronutrients (carbohydrate, protein, fat) intake is not the only affecting factor hemoglobin level. Micronutrient intake also affects the hemoglobin factor, it was reported that the micronutrient intake is relatively good in the said study.[33] Energy intake deficiency may cause anemia as the protein breakdown is no longer aimed at erythropoiesis (red blood cell production) but instead to produces energy or glucose.[32] Another study by Wiraprasidi et al. at Lolak Public Health Centre also reported similar findings; there was an insignificant association between hemoglobin level and MUAC.[34]

The Difference of Bone Density between Santriwati With and Without Risk of CED

A significant difference in bone density between CED and normal groups was not found based on the analysis. This finding supports a previous study by Ana Yuliah Rahmawati, which stated no significant association between BMI and bone density among participants.[35] Likewise, Shera Mutiara reported no significant association between nutritional status and bone density among female adolescents.[19] Many factors influence bone density; one of them being protein intake. Adequate protein intake is necessary to maintain estrogen production, which regulates bone synthesis.[15] However, the protein intake in both the CED and normal groups was relatively similar; this may justify bone density similarity in both groups. Another influencing factor is body fat percentage. A significant association between bone density and body fat percentage was reported in a previous study conducted in Bandung. The higher the body fat percentage, the lower risk of osteoporosis is. However, maintaining a body fat percentage in the normal range is recommended to prevent degenerative diseases.[36]

CONCLUSIONS

Based on the hemoglobin level, 57.4% of santriwati had anemia. Nutritional status assessment based on MUAC showed that 44.6% of participants were at risk of CED. On the other hand, 67.3% of the participants had a normal body fat percentage. Santriwati with osteopenia accounted for 13.9% of the total participants. There were significant differences in energy availability, BMI, body fat percentage between CED and normal groups. The mean scores in the CED group were lower than the normal group. Yet, there were no significant differences in bone density and hemoglobin levels between CED and normal groups.

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REFERENCES

1. Mujiati I, Budijanto D, Khairani. Situasi Keluarga Berencana di Indonesia. Indonesia: Kementerian Kesehatan Republik Indonesia; 2013.
2. Muliawati S. Faktor Penyebab Ibu Hamil Kurang Energi Kronis di Puskesmas Sambi Kecamatan Sambi Kabupaten Boyolali tahun 2012. 1. 2013;3(3):40–62.
3. Kementerian Kesehatan RI. Riset Kesehatan Dasar 2018 (RISKESDAS 2018). Indonesia: Kementerian Kesehatan Republik Indonesia; 2018.
4. Peni Catur Inayati. Hubungan antara Status Gizi dan menstruasi dengan Kejadian Anemia pada Santri Putri Pondok Pesantren Al-Hidayah Kecamatan Karangrayung Kabupaten Grobogan tahun 2009 [Internet] [other]. Universitas Negeri Semarang; 2009 [cited 2020 Sep 21]. Available from: https://lib.unnes.ac.id/1384/
5. Hidayah N, Rahfiludin MZ, Aruben R. Hubungan Status Gizi, Asupan Zat Gizi, dan Aktivitas Fisik dengan Siklus Menstruasi Remaja Putri Pondok Pesantren Salafiyah Kauman Kabupaten Pemalang Tahun 2016. Jurnal Kesehatan Masyarakat (e-Journal). 2016 Nov 2;4(4):537–444.
6. Kumar P, Sinha R, Patil N, Kumar V. Relationship between mid-upper arm circumference and BMI for identifying maternal wasting and severe wasting: a cross-sectional assessment. Public Health Nutr. 2019;22(14):2548–52.
7. Priyanto LD. The Relationship of Age, Educational Background, and Physical Activity

on Female Students with Anemia. JBE. 2018 Aug 30;6(2):139–46.
8. Sya`Bani IRN, Sumarmi S. Hubungan Status Gizi dengan Kejadian Anemia pada Santriwati di Pondok Pesantren Darul Ulum Peterongan Jombang. Jurnal Keperawatan Muhammadiyah. 2017 Mar 28;1(2):7–15.
9. Purwandari ES. Comparison Anemia In Adolescent Girls Who Stay At Pondok Pesantren And At Home In Madrasah Aliyah Pondok Pesantren Darussalam Kepung Kediri. J I K E B. 2018 Sep 25;4(2):114–9.
10. Ariyani DE, Achadi EL, Irawati A. Validity Mid-Upper Arm Circumference to Detect Chronic Energy Malnutrition Risk of Indonesia Women. Kesmas: National Public Health Journal. 2012 Sep 1;7(2):83.
11. Ong JL, Brownlee IA. Energy Expenditure, Availability, and Dietary Intake Assessment in Competitive Female Dragon Boat Athletes. Sports (Basel). 2017 Jun 21;5(2):45.
12. Papageorgiou M, Dolan E, Elliott-Sale KJ, Sale C. Reduced energy availability: implications for bone health in physically active populations. Eur J Nutr. 2018;57(3):847–59.
13. Sygo J, Coates AM, Sesbreno E, Mountjoy ML, Burr JF. Prevalence of Indicators of Low Energy Availability in Elite Female Sprinters. Int J Sport Nutr Exerc Metab. 2018 Sep 1;28(5):490–6.
14. Noprisanti N, Masrul M, Defrin D. Hubungan Asupan Protein, Kalsium, Phosfor, Dan Magnesium Dengan Kepadatan Tulang Pada Remaja Putri di SMP Negeri 5 Padang. Jurnal Kesehatan Andalas. 2018 Oct 17;7:29–36.
15. Septriani RS, Fitranti DY. Hubungan Asupan Protein dan Kafein dengan Kepadatan Tulang pada Wanita Dewasa Muda. J NutriColl. 2013 Oct 3;2(4):483–90.
16. Schtscherbyna A, Ribeiro BG, Farias MLF de. Bone Health, Bone Mineral Density, and Sports Performance. In: Nutrition and Enhanced Sports Performances. San Diego: Elsevier; 2013. p. 75–80.
17. Zhang Y, Wang Z, Chu Z, Zhao J. Profiles of body mass index and the nutritional status among children and adolescents categorized by waist-to-height ratio cut-offs. International Journal of Cardiology. 2016 Nov;223:529–33.
18. Lee RD, Nierman DC. Anthropometry. In: Nutritional Assessment. 3rd ed. New York: McGraw-Hill Education; 2003. p. 184–203.
19. Rahmani SM, Probosari E. Hubungan Indikator Obesitas dengan Kepadatan Tulang pada Wanita Dewasa Awal. Journal of Nutrition College. 2016;5(4):444–51.
20. Márquez S, Molinero O. Energy availability, menstrual dysfunction and bone health in sports; an overview of the female athlete triad. Nutr Hosp. 2013 Aug;28(4):1010–7.
21. Setyarsih L, Ardiaria M, Fitranti DY. Hubungan Densitas Energi dan Asupan Cairan dengan Berat Jenis Urin pada Remaja. Journal of Nutrition College. 2017;6(4):326–32.
22. Sandjaja. Risiko Kurang Energi Kronis (KEK) pada Ibu Hamil di Indonesia. Gizindo. 2014 Sep 1;32(2):128–38.
23. Alifah HN, Anita DC. Hubungan Status Gizi dengan Kadar Hemoglobin pada Santriwati di Pondok Pesantren Al-Munawwir Krapyak Bantul Yogyakarta [Internet] [s1_sarjana]. [Yogyakarta]: Universitas 'Aisyiyah Yogyakarta; 2017 [cited 2020 Sep 21]. Available from: http://lib.unisayogya.ac.id/
24. Sudikno S, Sandjaja S. Prevalensi Dan Faktor Risiko Anemia Pada Wanita Usia Subur Di Rumah Tangga Miskin Di Kabupaten Tasikmalaya Dan Ciamis, Provinsi Jawa Barat. Kespro. 2016;7(2):71–82.
25. Hamidiyah A, Rohmani L, Zahro NA. Faktor Determinan Anemia Santri Putri. Oksitosin : Jurnal Ilmiah Kebidanan. 2019 Feb 1;6(1):64–72.
26. Benítez Brito N, Suárez Llanos JP, Fuentes Ferrer M, Oliva García JG, Delgado Brito I, Pereyra-García Castro F, et al. Relationship between Mid-Upper Arm Circumference and Body Mass Index in Inpatients. PLoS ONE. 2016;11(8):e0160480.
27. Misra P, Singh AK, Archana S, Lohiya A, Kant S. Relationship between body mass index and percentage of body fat, estimated by bio-electrical impedance among adult females in a rural community of North India: A cross-sectional study. J Postgrad Med. 2019 Sep;65(3):134–40.
28. Shetty PS, James WP. Body mass index. A measure of chronic energy deficiency in adults. FAO Food Nutr Pap. 1994;56:1–57.

29. Williams NI, Statuta SM, Austin A. Female Athlete Triad: Future Directions for Energy Availability and Eating Disorder Research and Practice. Clin Sports Med. 2017 Oct;36(4):671–86.
30. Holtzman B, Ackerman KE. Measurement, Determinants, and Implications of Energy Intake in Athletes. Nutrients. 2019 Mar;11(3):665.
31. Petrika Y, Hadi H, Nurdiati DS. Tingkat asupan energi dan ketersediaan pangan berhubungan dengan risiko kekurangan energi kronik (KEK) pada ibu hamil. Jurnal Gizi dan Dietetik Indonesia (Indonesian Journal of Nutrition and Dietetics). 2016 Aug 30;2(3):140–9.
32. Wardhani JP, Rahfiludin MZ, Pradigdo SF. Perbedaan Aktivitas Fisik, Kadar Hb, dan Kesegaran Jasmani (Studi pada Siswi KEK dan Tidak KEK di SMAN 1 Grogol Kabupaten Kediri). Jurnal Kesehatan Masyarakat (e-Journal). 2017 Dec 13;3(3):205–12.
33. Indartanti D, Kartini A. Hubungan Status Gizi dengan Kejadian Anemia pada Remaja Putri. Journal of Nutrition College. 2014;3(2):310–6.
34. Wiraprasidi IPA, Kawengian SE, Mayulu N. Faktor-Faktor yang Berhubungan dengan Kadar Hemoglobin pada Ibu Hamil di Puskesmas Lolak. eBiomedik [Internet]. 2017 [cited 2020 Sep 21];5(2). Available from: https://ejournal.unsrat.ac.id/index.php/ebiomedik/article/view/18602
35. Rahmawati AY. Hubungan antara Indeks Massa Tubuh (IMT), Asupan Zat Gizi dan Riwayat Reproduksi dengan Kepadatan Mineral Tulang pada Wanita Pre-Menopause. Jurnal Riset Kesehatan. 2016;5(2):83–91.
36. Widyanti LRE, Kusumastuty I, Arfiani EP. Hubungan Komposisi Tubuh dengan Kepadatan Tulang Wanita Usia Subur di Kota Bandung. Indonesian Journal of Human Nutrition. 2017 Jun 25;4(1):23–33.

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Ganyong-kelor snack bar's glycemic index as a diet for diabetics

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ABSTRACT

Background: Lack of insulin or the inability of cells to respond to insulin causes high blood glucose levels or hyperglycemia, a hallmark of diabetes. Consumption of foods with a low glycemic index and high fiber has been shown to provide the same benefits as pharmacological therapy in the control of postprandial hyperglycemia and can prevent the incidence of hypoglycemia in people with diabetes. Ganyong (Canna edulis) is a food source of carbohydrates and fiber. Kelor (Moringa oliefera) contains protein and some phytochemical compounds which have a hypoglycemic effect.
Objectives: The objective of the study was to analyze the glycemic index of ganyong-kelor snack bars as a diet for diabetics.
Materials and Methods: Ten respondents fasted for 10 hours and checked their fasting blood glucose levels, then consumed 105 grams of bread as the reference food. Every 30 minutes after eating, the blood glucose levels were checked. In the following week, after fasted, all respondents consumed 157 grams of a ganyong-kelor snack bar and checked their blood glucose levels every 30 minutes.
Results: Every 100 grams of ganyong-kelor snack bar contains 230.13 kcal, 31.97 grams of carbohydrates, 9.25 grams of fat, and 4.75 grams of protein. In this study, bread was used as a reference food. If bread was corrected with glucose as a reference food, the glycemic index of the ganyong-kelor snack bar was 38.08. The calculation of the glycemic load used the converted-glycemic index and the total carbohydrates contained in 100 grams of the food. Ganyong-kelor snack bar had a glycemic load value of 12.10.
Conclusions: Ganyong-kelor snack bar had good nutritional content and was categorized as food with a low glycemic index. The hypoglycemic effect of the ganyong-kelor snack bar came from its high fiber content. Ganyong-kelor snack bar can be consumed as a healthy snack for diabetic people.

Keywords: Ganyong; Canna; Kelor; Moringa; Diabetes

BACKGROUND

Diabetes mellitus is a chronic condition that occurs as a result of increased levels of glucose in the blood caused by the body's unable to produce sufficient amounts of insulin or the body is unable to use the insulin produced effectively.[1] Lack of insulin or the inability of cells to respond to insulin causes high blood glucose levels, or hyperglycemia, which is a hallmark of diabetes.[2] In 2017 the prevalence of diabetes mellitus globally is 8.8% or around 425 million adults with 85-95% of these cases are type 2 diabetes mellitus and is expected to increase to 9.9% in 2045.[2-4] The prevalence of diabetes mellitus in Indonesia increases rapidly from 5.7% in 2007 to 6.9 % in 2013 to 8.5% in 2018.[5] Diabetes mellitus has a significant economic impact on countries, health systems, and diabetics or families through direct medical costs and job losses and reduced wages.[5]

Modifiable risk factors for type 2 diabetes mellitus include poor diet and nutrition, excess adiposity, low physical activity, prediabetes or impaired glucose tolerance (IGT), smoking habits, and a history of fetal exposure to high blood glucose during pregnancy.[4] Recent evidence suggests an association between regular exposure to a high glycemic load diet and causing postprandial hyperglycemia.[6-11] Consumption of foods with low glycemic index and high fiber has been shown to provide the same benefits as pharmacological therapy in the control of postprandial hyperglycemia in the medium term and can prevent the incidence of hypoglycemia in people with diabetes.[11-13] This type of diet can improve insulin sensitivity and fat metabolism.[14] This can provide a good choice for diabetics and can reduce health care costs.

Ganyong (*Canna edulis*) is a food source of carbohydrates with a low glycemic index (20.8) and high fiber (8.59%). The starch content of ganyong consists of 25% amylose.[15.16] The protein content of kelor (*Moringa oliefera*) leaves is 35% of the dry weight. Kelor leaves contain some phytochemical compounds such as alkaloids, flavonoids, glycosides, tannins, and steroids, which have a hypoglycemic effect.[17,18] Diabetics must maintain their diet, type, and calorie content in food, especially in those who use insulin secretion-enhancing drug therapy or

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insulin injection therapy.[19] G*anyong-kelor* snack bar is expected to have low index glycemic and can be consumed as a snack for diabetics without causing hyperglycemia.

MATERIALS AND METHODS

This research was an experimental study. *Ganyong-kelor* snack bar is made using ingredients such as ganyong/ canna obtained from Giwangan Market, canna flour obtained from the Center for Food and Nutrition Studies Universitas Gadjah Mada, kelor/ moringa leaves obtained from Purworejo, non-calorie sweetener, skim milk, margarine, eggs, and essence chocolate. This research has been approved and received ethical permission from *Komite Etik Penelitian Kesehatan Politeknik Kesehatan* (KEPK) Yogyakarta (LB.01.01/KE-01/XXXIV/731/2018). Testing of nutritional content includes analysis of protein (the Kjeldahl method), fat (the Soxhlet method), carbohydrates (the difference method), crude fiber (the gravimetric method), and amylose content (the IRRI method) carried out at the Food Technology Laboratory, Universitas Gadjah Mada.

The glycemic index test was carried out on 10 subjects who had fasted for 10 hours then ate the reference food in the form of white bread with 50 grams of carbohydrates. Every 30 minutes (for 2 hours) a 20 μL blood sample was taken to measure the postprandial blood glucose level. One week later, the same procedure is carried out by providing a snack bar containing 50 grams of carbohydrates. The glycemic index was obtained by the incremental area under the blood glucose response curve (IAUC) method by comparing the area under the blood glucose response curve to a snack bar with a reference food.

RESULTS

Ten selected respondents who had signed an informed consent have normal nutritional status measured by body mass index (BMI) and fasting blood sugar 70-100 mg/dL. Respondents fasted for 10 hours and checked their blood sugar twice with an interval of 7 days. The average result of the first measurement of fasting blood sugar for all respondents was 84.90 mg/dL while the second measurement was 84.30 mg/dL. The mean value of the two measurements is within the normal fasting blood glucose range (70-100 mg/dL).

After the respondents fasted and checked their fasting blood glucose levels, all respondents consumed 105 grams (50 gr carbohydrates) of bread as the reference food. Every 30 minutes after eating, the blood glucose levels were checked for each respondent. In the following week, after the respondents fasted, all respondents consumed 157 grams (50 gr carbohydrates) of the *ganyong-kelor* snack bar and checked their blood glucose levels every 30 minutes.

Table 1. Nutrient Content per 100g G*anyong-Kelor* Snackbar

Nutrient	100 g G*anyong-Kelor* Snackbar
Energy	230.13 kkal
Water	49.79 %
Carbohydrate	31.97 %
Amilosa	10.04 %
Fat	9.25 %
Protein	4.75 %
Ash	4.23 %
Crude fiber	2.64 %

The blood glucose response of each respondent after consuming the reference food and *ganyong-kelor* snack bar is seen in Figure 1. There was an increase in blood glucose levels (51.11%) from 84.9 mg/dL to 128.3 mg/dL within 30 minutes after consuming bread. This blood glucose level becomes the peak of the increase in blood glucose levels. In the next 30 minutes, there was a decrease in blood glucose levels to 121.7 mg/dL or 5.14% of the peak value and continued to decrease to 24.70% at 2 hours after consuming bread.

The peak increase in blood glucose levels after snack bar consumption reached 114.7 mg/dL or 36.06% from fasting blood glucose (84.3 mg/dL). Measurement of blood glucose levels in the next 30 minutes to 2 hours showed a decrease in blood glucose levels of 14.55%, 16.73%, and 20.48% respectively from the highest levels. Compared to bread, consumption of the *ganyong-kelor* snack bar caused a slower increase in blood glucose levels and closer to normal blood glucose levels after 2 hours of snack bar consumption. The glycemic index is calculated from the increase in blood glucose with fasting glucose levels as the initial value. The glycemic index is obtained by calculating the broad curve of the increase in blood glucose levels after

consuming bread (A) and consuming ganyong-kelor snack bar (B) in Figure 2. The B/A ratio multiplied by 100 will reveal the glycemic index of the ganyong-kelor snack bar.The ganyong-kelor snack bar has a glycemic index value of 55.19. In this study, bread was used as a reference food. If this study uses glucose as a reference (glycemic index 100 and glycemic index of bread 69) and corrected bread as a reference food, it will make the glycemic index value of the ganyong-kelor snack bar to 38.08. The calculation of the glycemic load uses the converted-glycemic index value and the total carbohydrates contained in 100 grams of the food. Ganyong-kelor snack bar has a glycemic load value of 12.10.

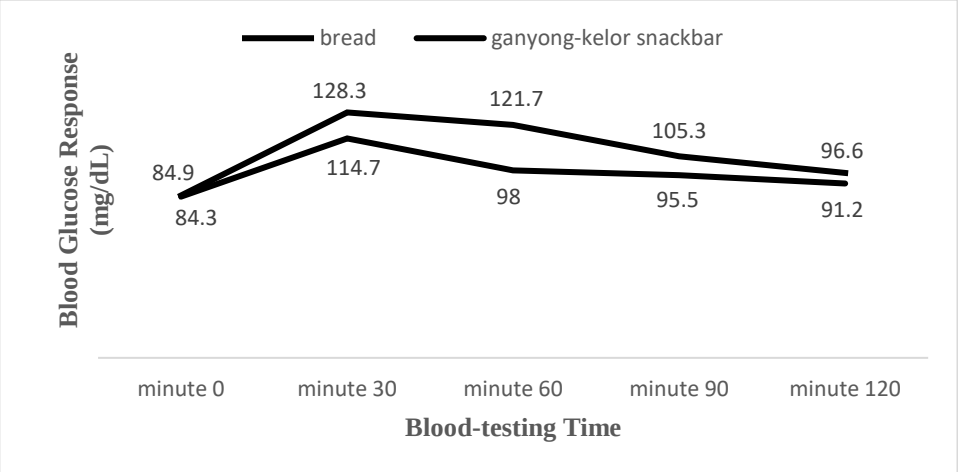

Figure 1. Blood Glucose Response After Consuming Bread and *Ganyong-Kelor* Snackbar

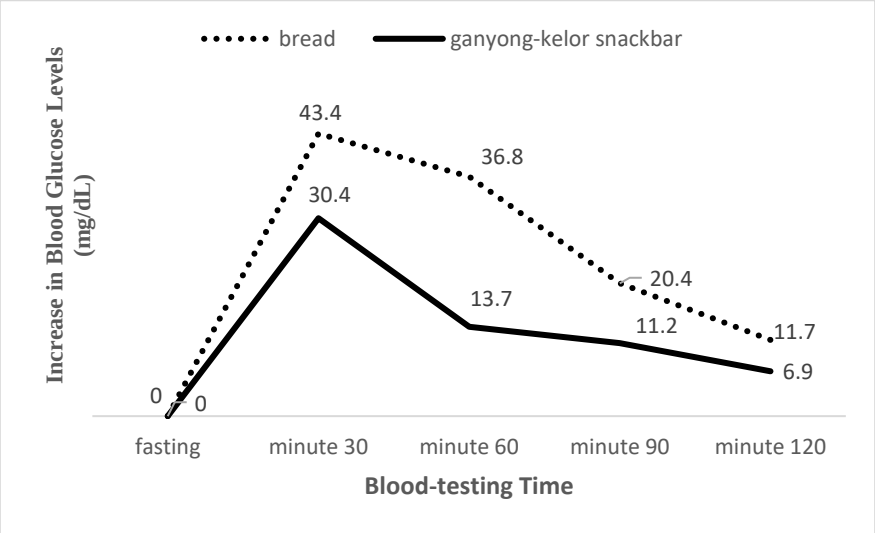

Figure 2. Increase in Blood Glucose Levels After Consuming Bread and G*anyong-Kelor* Snackbar

Table 2. Glycemic Index of G*anyong-Kelor Snackbar* and Bread

Food	Glycemic Index (with bread as reference)	Glycemic Index (with glucose as reference)	Glycemic Load
Bread	100	69	32.85
ganyong-kelor snackbar	55.19	38.08	12.10

DISCUSSION

Diabetics must maintain their diet. type, and calorie content in food, especially in those who use insulin secretion-enhancing drug therapy or insulin

injection therapy.[19] A snack is needed to accompany the main meal in meeting nutritional needs and to help control diabetics' blood glucose levels. Snack bars are a food product that has begun to be developed as a snack for diabetics that can help prevent hyperglycemia by using foods high in fiber and low on the glycemic index. It is recommended that diabetics have a snack that contains 10-15% of their daily energy needs and can be consumed 2-3 times a day.[19]

Adequate energy to do daily activities and maintain ideal body weight is met by consuming a *ganyong-kelor* snack bar with the appropriate portion and calorie content. Consumption of foods with excess calories can lead to obesity which is a risk factor for type 2 diabetes.[21] In every 100 grams of *ganyong-kelor* snack bar contains 230.13 kcal. Diabetics can consume 43.45 grams of the *ganyong-kelor* snack bar to get 100 kcal of calorie intake. This portion contains 13.89 grams of carbohydrates, 4.01 grams of fat, and 2.06 grams of protein. *Ganyong-kelor* snack bar has a glycemic index of 55.19. This study using bread as a reference food. If using glucose as a reference, makes the glycemic index of the g*anyong-kelor* snack bar to 38.08. Based on the classification of the glycemic index, after correcting the *ganyong-kelor snack bar* is included in a low glycemic index food (> 55).

The ingredients of the *ganyong-kelor snack bar* that are thought to have an impact on the low glycemic index are ganyong/ canna and kelor/ moringa leaves. In these two food ingredients, fiber content is the part that greatly affects the glycemic index. The total dietary fiber in 100 grams of canna starch ranges from 81.9 to 85.9% dry weight. The high amylose and fiber content in canna affects the low glycemic index of the *ganyong-kelor snack bar*. As a high-carbohydrate food, ganyong has a lower glycemic index (20.8) when compared to rice (72.8).[15] Consumption of foods with a low glycemic index can improve insulin sensitivity and decrease the rate of glucose absorption which improves glycemic control in diabetics.[11-13, 22]

Food with a low glycemic index will be digested and converted into glucose gradually and slowly resulting in a slower release of glucose into the bloodstream so that the increase in blood glucose levels will be lower and the fluctuation of the increase in blood glucose is relatively shorter. This will affect increasing insulin secretion and glucose consumption by liver cells which will result in reduced blood glucose levels. Consumption of foods with a low glycemic index can improve insulin sensitivity and reduce the rate of glucose absorption. This can improve glycemic control in people with diabetes mellitus.[11-13,22] Some of the factors that influence the glycemic index are carbohydrate composition, tertiary structure, and enzymatic digestion.[22] Clinical studies show that consumption of foods high in water-soluble fiber such as guar gum, pectin, β-glucans found in nuts, vegetables, fruit, and oat cereals can control insulin levels and lower postprandial blood glucose levels.[22] The hypoglycemic effect of a diet high in soluble fiber can be achieved through slowing gastric emptying and enzymatic absorption, shorter intestinal transit times, and the formation of a physical barrier on carbohydrates that slows down glucose uptake.[22,23]

The glycemic load is defined as the glycemic index of a food multiplied by the carbohydrate content in the product. The calculation of the glycemic load is intended to provide information about the effect of food consumption on increasing blood glucose levels associated with the glycemic index. The lower the carbohydrate content, the lower the glycemic load. Then the food consumed will trigger a lower increase in blood glucose levels compared to the reference food. *Ganyong-kelor snack bar* has a glycemic load value of 12.10. This food is included in the category of moderate glycemic load foods (range of 11 to 19).

This study has many limitations. The material used as the standard for measuring the glycemic index is bread. The author suggests further research using glucose as a reference. However, the analysis of the results of this study can provide important evidence regarding the use of canna and moringa leaves as materials for snack bars, introducing *ganyong-kelor snack bar* as an alternative snack for diabetics, and can be the starting point for further research.

CONCLUSIONS

Ganyong-kelor snack bar has good nutritional content and is categorized as food with a low glycemic index. Ganyong-kelor snack bar can be consumed as a healthy snack for diabetes without causing hyperglycemia. Every 100 grams of *ganyong-kelor snack bar* contains calories of 230.13 kcal, 31.97 grams of carbohydrates, 9.25 grams of fat, and 4.75 grams of protein. The glycemic index of the *ganyong-kelor snack bar was* 38.08 (low category) with a glycemic load of 12.10 (moderate category). The hypoglycemic effect of snack bars comes from their high fiber content. The hypoglycemic effect of a diet high in fiber can be achieved through slowing gastric emptying and enzymatic absorption, shorter intestinal transit times, and the formation of a physical barrier on carbohydrates that slows down glucose uptake. Knowing the glycemic load on food will help us to choose food and determine food

portions wisely so that it will limit carbohydrate intake too high which can cause an increase in blood glucose levels. This is especially important for diabetics or people who want to limit their calorie intake to control their weight.

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REFERENCES

1. World Health Organization. Definition, Diagnosis, and Classification of Diabetes Mellitus and its Complications. Part 1: Diagnosis and Classification of Diabetes Mellitus (WHO/NCD/NCS/99.2). Geneva: World Health Organization; 1999. *http://apps.who.int/iris/handle/10665/66040*

2. The International Diabetes Federation. Diabetes Atlas 8th Edition. Brussels: The International Diabetes Federation; 2017. *https://www.idf.org/e-library/epidemiology-research/diabetes-atlas.html*

3. World Health Organization. Global report on Diabetes. Geneva: World Health Organization; 2016. *http://apps.who.int/iris/bitstream/10665/204871/1/9789241565257_eng.pdf*

4. NCD Risk Factor Collaboration. Worldwide trends in diabetes since 1980: a pooled analysis of 751 population-based studies with 4.4 million participants. Lancet. April 2016; 387: 1513-1530. DOI: *http://dx.doi.org/10.1016/S0140-6736(16)00618-8*

5. Badan Penelitian dan Pengembangan Kesehatan. Laporan Nasional Riskesdas 2018. Jakarta: Lembaga Penerbit Badan Penelitian dan Pengembangan Kesehatan; 2019.

6. Castro-Acosta ML, Stone SG, Mok JE, Mhajan RK, Fu CI, Lenihan-Geels GN, et al. Apple and blackcurrant polyphenol-rich drinks decrease postprandial glucose, insulin, and incretin response to a high-carbohydrate meal in healthy men and women. Journal of Nutritional Biochemistry. 2017; 49:53–62. DOI: *http://dx.doi.org/10.1016/j.jnutbio.2017.07.013*

7. Schulze MB, Manson JE, Ludwig DS, Colditz GA, Stampfer MJ, Willett WC, et al. Sugar-sweetened beverages, weight gain, and incidence of type 2 diabetes in young and middle-aged women. JAMA. 2004; 292(8): 927–34. DOI: *http://dx.doi.org/10.1001/jama.292.8.927*

8. Sluijs I, van der Schouw YT, van der AD, Spijkerman AM, Hu FB, Grobbee DE, et al. Carbohydrate quantity and quality and risk of type 2 diabetes in the European prospective investigation into cancer and nutrition-Netherlands (EPIC-NL) study. Am J Clin Nutr. 2010; 92: 905–11. DOI: *https://doi.org/10.3945/ajcn.2010.29620*

9. de Koning L, Malik VS, Rimm EB, Willett WC, Hu FB. Sugar-sweetened and artificially sweetened beverage consumption and risk of type 2 diabetes in men. Am J Clin Nutr. 2011; 93(6): 1321–27. DOI: *https://doi.org/10.3945/ajcn.110.007922*

10. Blaak EE, Antoine JM, Benton D, Bjorck I, Bozzetto L, Brouns F, et al. Impact of postprandial glycemia on health and prevention of disease. Obes Rev. 2012; 13(10): 923–84. DOI: *https://doi.org/10.1111/j.1467-789X.2012.01011.x*

11. Riccardi G, Rivellese AA, Giacco R. Role of glycemic index and glycemic load in the healthy state, in prediabetes, and diabetes. Am J Clin Nutr. 2008; 87(1): 269S–74S. DOI: https://doi.org/10.1093/ajcn/87.1.269S

12. Brand-Miller J, Hayne S, Petocz P, Colagiuri S. Low glycemic index diets in the management of diabetes: a meta-analysis of randomized controlled trials. Diabetes Care. 2003; 26(8): 2261–67. DOI: https://doi.org/10.2337/diacare.26.8.2261

13. American Dietetic Assosiation. Nutrition recommendation and interventions for diabetes (position statement). Diabetes Care. January 2008; 31(1): S61-S78. DOI: *https://doi.org/10.2337/dc08-S061*

14. Rizkalla SW, Taghrid L, Laromiguiere M, Huet D, Boillot J, Rigoir A, et al. Improved plasma glucose control, whole-body glucose utilization, and lipid profile on a low glycemic index diet in type 2 diabetic men. Diabetes Care. 2004; 27(8): 1866–72. DOI: https://doi.org/10.2337/diacare.27.8.1866

15. Lukitaningsih E, Rumiyati, Ika Puspitasari. Kajian glisemik indeks dan makronutrien dari umbi-umbian dalam upaya pencarian sumber pangan fungsional. Pharmacon. 2012; 13(1): 18-

23. https://publikasiilmiah.ums.ac.id/bitstream/handle/11617/3377/2012-13-1-18.pdf?sequence=1&isAllowed=y
16. Carolina A, Ilmi FN. Production of Indonesian *canna edulis* type iv resistant starch through acetylation modification. International Food Research Journal. 2016; 23(2): 491-97. http://www.ifrj.upm.edu.my/23%20(02)%202016/(6).pdf
17. Tshingani K, Donnen P, Mukumbi H, Duez P, Dramaix-Wilmet M. Impact of *Moringa oleifera lam.* leaf powder supplementation versus nutritional counseling on the body mass index and immune response of HIV patients on antiretroviral therapy: a single-blind randomized control trial. BMC Complementary and Alternative Medicine. 2017; 17:420-33. DOI: *https://doi.org/10.1186/s12906-017-1920-z*
18. Jaiswal D, Rai PK, Mehta S, Chatterji S, Shukla S, Rai DK, et al. Role of *Moringa oleifera* in regulation of diabetes-induced oxidative stress. Asian Pacific Journal of Tropical Medicine. 2013; 6(6): 426-32. DOI: *https://doi.org/10.1016/S1995-7645(13)60068-1*
19. Perkumpulan Endokrinologi Indonesia. Konsensus pengelolaan dan pencegahan diabetes melitus tipe 2 di Indonesia 2015. Jakarta: PB. Perkeni; 2015. https://pbperkeni.or.id/wp-content/uploads/2019/01/4.-Konsensus-Pengelolaan-dan-Pencegahan-Diabetes-melitus-tipe-2-di-Indonesia-PERKENI-2015.pdf
20. Mortensen A. Sweeteners permitted in the European Union: safety aspects. Scandinavian Journal of Food and Nutrition. 2006; 50(3): 104-16. DOI: https://doi.org/10.1080/17482970600982719
21. Franz MJ. Medical nutrition therapy for diabetes mellitus and hypoglycemia of nondiabetic origin. In: Mahan LK, Escottt-Stump S. Krause's food, and the nutrition care process. 13th edition. Philadelphia: Elsevier; 2012. 675-710.
22. Frost G. Carbohydrate: Glucose tolerance and the glycemic (glycaemic) index. In: Caballero B. Encyclopedia of food sciences and nutrition. New York: Academic Press; 2003. 2917-22.
23. Gallagher ML. Intake: The nutrient and their metabolism. In: Mahan LK, Escottt-Stump S. Krause's food, and the nutrition care process. 13th edition. Philadelphia: Elsevier; 2012. 32-41.

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Household food security and diet quality with chronic energy deficiency among preconception women

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ABSTRACT

Background: Chronic energy deficiency (CED) that occurs at risk preconception women during pregnancy increased low birth weight (LBW) in infants. Household food security and diet quality are factors that cause CED. Therefore, this study aims to determine the relationship between household food security and diet quality with CED preconception women.
Materials and Methods: We used a cross-sectional study design. The subject of 70 preconception women aged 16-35 years registered in the religious affairs office in Sumowono and Pringapus Subdistrict were selected by consecutive sampling method. Weight and height were measured to assess body mass index to determine CED. Household food security was measured using the Household Food Security Scale Module (HFSSM). Food intake data were obtained using the Semi-Quantitative Food Frequency Questionnaire (SQ-FFQ) and DQI-I (Diet Quality Index-International) to measure diet quality. Bivariate analyses were tested using Rank Spearman and Pearson Product Moment.
Results: The prevalence of subjects with CED risk was 48.6% and subjects with CED was 15.7%. 38.6% of subjects married at the age of 16-20 years, 75.1% of subjects had low household food security and 80% of subjects had low diet quality. There was no significant relationship between household food security and diet quality with CED, namely (p = 0.537) and (p = 0.711). The components of diet quality, namely variation, adequacy, moderation and balance also did not show a significant relationship with CED, respectively with p-value (p = 0.711), (p = 0.523), (p = 0.412), (p = 0.604)
Conclusions: There was no correlation between household food security and CED, also no correlation between diet quality and CED.

Key Words: CED; Diet quality; food security; Preconception woman

BACKGROUND

The preconception period is the period before pregnancy. Preconception women are women of reproductive age, who will have to be ready to become mothers. The preconception period needs different nutritional support compared to the previous period. The nutritional status of pregnant women is determined by the period before pregnancy, thus the poor nutritional status of preconception women will have an impact on the pregnancy. Preconception women are also defined as women of reproductive age who are also prone to CED.[1]

Chronic energy deficiency (CED) in women is defined as a condition when a person experiences a prolonged or chronic lack of calories and protein. CED is characterized by a mid-upper arm circumference (MUAC) of <23.5 cm.[2] Another indicator in defining CED is body mass index (BMI) of <18.5 kg/m^2.[3] Based on the results of Basic Health Research (Riskesdas) 2013, the prevalence of pregnant women aged 15-49 years who suffer from CED was 24.3%, while the prevalence of women of reproductive age who were not pregnant was 20.8%.[4] A preceding research stated that the prevalence of CED in pregnant women in Semarang District was 10.28%.[5] Another study conducted by Sumarmi also stated that the prevalence of CED among future brides in 4 sub-districts of Probolinggo District was 27.3%.[6]

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The impact of the incidence of CED among pregnant women in Indonesia includes the high rate of infant mortality (IMR) due to low birth weight (LBW) where the prevalence of LBW reached 10.2% in 2013.[7] Other impacts that can arise due to CED are bleeding, abnormal weight gain in pregnant women, and infectious diseases. Pregnant women with CED have a high risk of miscarriage, abortion, stillbirth, congenital defects, anemia in the baby, and die in the womb (Asphyxia intrapartum).[8]

The quality and quantity of diet are some of the factors that cause CED in preconception women. The quality of the diet is an important index to determine the intake of macro and micro-nutrients as well as dietary patterns that may affect the risk of diet-related diseases. According to researches that have been conducted in developing countries such as Indonesia and India, it is known that the quality of diet will affect the nutritional status of women of reproductive age (WRA) including brides.[9] Other studies have shown that a poor quality diet before pregnancy will affect the nutritional status of pregnant women which may lead to low birth weight (LBW) in children.[10]

The quality of one's diet is determined by the food security of the household, the better household food security, the better its diet quality. Food insecurity in WRA often results in the consumption of food that is not following their needs. This can cause a lack of energy, protein, and micronutrients which will affect the nutritional status of women of childbearing age, namely chronic energy deficiency (CED).[11,12]

Sumowono and Pringapus subdistricts are areas with a fairly high prevalence of infant mortality rate (IMR) and maternal mortality rate (MMR). Based on the Health Profile of Semarang District in 2016, there were 5 cases of MMR within 2011-2016. Meanwhile, during the same period, 70 and 49 cases of IMR were recorded in Sumowono and Pringapus districts respectively.[13] One of the risk factors of low birth weight (LBW) and also a major risk factor in infant mortality is CED during pregnancy.[14] The number of CED cases in Sumowono and Pringapus districts occurred due to the intake of protein and energy of their people which was relatively low compared to people in other areas in Central Java.[15] Therefore, this study aims to examine the relationship between household food security and diet quality with chronic energy deficiency (CED) in preconception women.

MATERIALS AND METHODS

This study was conducted between March-April 2018 in the Office of Religious Affairs (*Kantor Urusan Agama*/KUA) of Sumowono and Pringapus sub-districts, Semarang District. This is a cross-sectional study within the scope of Public Health Science. This study has been granted a permit by the Health Research Ethics Commission of the Faculty of Medicine, Universitas Diponegoro - Central Public Hospital Dr. Kariadi Semarang with No. 480 / EC / FK-RSDK / VII / 2018.

The minimum sample size was calculated using a correlative analytic formula and the 70 study participants were selected through the consecutive sampling method. The sample inclusion criteria include preconception women registered in the Office of Religious Affairs Sumowono and Pringapus sub-districts, aged 16-35 years, never and not currently pregnant (at the time of the data collection), and willing to fill in their information and sign a statement of willingness as research study participants.

The independent variables in this study were household food security and diet quality, while the dependent variable was chronic energy deficiency (CED). The cut-off point of CED is Mid-Upper Arm Circumference (MUAC) of <23.5 cm and has a BMI of <18.5 kg / m^2. The collected data in this study were study participants' general data, anthropometric data, household food security, and diet quality.

Household food security is a condition when people have the right to have physical and economic access at any time to obtain sufficient food to meet their needs for a productive and healthy life. The household food security was measured using the Household Food Security Scale Module (HFSSM) on study participants who live with no children aged under 18 years. Each of the questions from the HFSSM was assigned a 1 score if the study participants answered frequently or occasionally and a 0 score for never. Food security is categorized into 4 categories, namely high food security with 0 scores, moderate food security with a score of 1-2, low food security with a score of 3-5, and very low (food insecurity) with a score of 6-10.[16,17]

Diet quality is an assessment of food consumption that consists of 4 categories, namely variation, sufficiency, moderation, and overall balance based on dietary guidelines using the Diet Quality Index International (DQI-I) form. The total score of the DQI-I varies between 0 as the lowest score and 100 as the highest score. Diet quality is rated low if the score is ≤ 60 and is rated high if the score is > 60.[18] Data on the diet quality was collected through food intake interviews using the Semi-Quantitative Food Frequency (SQ-FFQ) form during the last month. Food photo books are used to minimize bias in intake data collection.

Data processing and analysis were carried by a computer program. Univariate analysis was performed to describe the characteristics of the subject. The bivariate analysis started with a normality test with the Kolmogorov-Smirnov test. Furthermore, bivariate analysis was carried out to determine the correlation between the independent and dependent variables with the Rank-Spearman and Pearson Product Moment tests.

RESULTS

Subject Characteristics

The participants' characteristics consist of data on age, education, occupation, nutritional status, household food security, and diet quality.

Table 1. Subjects characteristics

Characteristics	n	%
Age		
Adolescents (16-20 years old)	27	38.6
Adult (21-35 years old)	43	61.4
Educational Level		
Elementary School	8	11.4
Junior High School	23	32.9
Senior High School	29	41.4
Bachelor	10	14.3
Occupation		
Employment	45	64.3
Unemployment	25	35,7
Chronic Energy Deficiency (CED)		
CED (BMI <18.5kg/$m^{2,}$ MUAC< 23.5 cm)	11	15.7
Normal (BMI ≥18.5kg/$m^{2,}$ MUAC< 23.5 cm)	59	84.3
Risk of Chronic Energy Deficiency		
Risk of CED (MUAC< 23.5 cm)	34	48.6
Normal (MUAC ≥ 23.5 cm)	36	51.4
Household Food Security		
High Food Security	14	17.1
Medium Food Security	2	2.4
Low Food Security	53	75.1
Very low (Food Insecurity)	1	1.4
Diet Quality		
Low (score ≤60)	56	80
High (score >60)	14	20

A total of 38.6% of the study participants aged 16-20 years old, within the youth category. Most of the participants were junior and senior high school level, only 14.3% of the participants had an education up to diploma or bachelor's degree. The nutritional status based on BMI indicated that 11

participants (15.7%) suffered from CED and the other 84.3% of participants were normal The MUAC measurement showed that 48.6% of participants were at risk of CED and another 51.4% of the participants were normal. A total of 53 participants (75.1%) were in the low food security category. Most of the participants (80%) also had poor diet quality, and only 14 participants (20%) had good diet quality.

Table 2 describes the minimum, maximum, average, and median values of age, BMI, MUAC, household food security score, and diet quality score. The youngest study participant was 16 years old. The average BMI was 21.46 kg/m^2, which was normal. However, there was a participant who had a BMI score of 15.6 kg/m^2 which was classified as CED. On the other hand, there was a study participant who had a BMI score of 32.8 kg/m^2 which was classified as over-nutrition. The average MUAC score was 23.9 cm which was normal, but some study participants' MUAC were less than 23.5 cm. They were classified as at risk of CED. The median score of household food security was 3, which showed that most of the participants had low household food security. The average score of diet quality was 51.77, which was classified as low dietary quality.

Table 2. Minimum, maximum, average, and median values of age, BMI, MUAC, household food security score, and diet quality score

Variable	Minimum	Maximum	Mean±DS/Median
Age (years)	16	29	21±3.4[b]
BMI (kg/m^2)	15.6	32.8	21.45±3.23[b]
MUAC (cm)	17	30.7	23.8±2.83[b]
Household Food Security (score)	0	8	3[a]
Diet Quality (score)	28	75	52±11[b]

[a] Median [b] Mean (Deviation Standard)

Household Food Security

Table 3 showed that 72.9% of the participants were worried about not being able to buy food before they get another income and were only able to buy food to survive. A total of 57.1% of participants were unable to consume a balanced diet because they had no money. There were 21.4% of the participants who consciously reduced their eating portions due to financial constraints. Furthermore, there were 14.3% of the participants ate less than usual. Meanwhile, 5.7% of the participants were unable to buy food when they got hungry. A total of 17.1% of the participants lost their weight due to financial constraints to buy food, and 7.1% of the participants experienced not eating any food for the whole day because they did not have enough money to buy food.

Diet Quality

The diet quality consists of four aspects, i.e., variety, adequacy, moderation, and overall balance. Table 4 shows that the median score of the variation of the participants was 17 out of a maximum score of 20, which means that the participants were already consuming a variety of foods including overall variance and variety of protein. The minimum score (0) was given if a participant did not consume any fruit and vegetable. Moderation scores showed that the average consumption of total fat was 36%, which was relatively high compared to the recommendation of DQI-I, which is less than 30% energy needs. Additionally, the average score of saturated fat consumption was 16.9%, which was relatively high. It was also higher than the recommendation (less than 10%). The adequacy aspect, shown in table 5, consists of some components, i.e., vegetable, fruit, staple foods, fiber intake, protein intake, iron intake, calcium intake, and vitamin C intake. The results of this study showed that the consumption of vegetable, iron, and calcium groups was low, which lead to the low quality of the overall diet. On the other hand, the intake of staple food, protein, and vitamin C was adequate.

Table 3. Components of the HFSSM Questionnaire

Statements	Responses	n	%
Household Level			
Worried about not being able to buy food before getting next income	S, K	51	72.9
	T	19	27.1
Only buying food to survive and unable to stock on food	S, K	51	72.9
	T	19	27.1
Unable to consume a balanced diet	S, K	40	57.1
	T	30	42.9
Individual Level			
Having the experience to reduce an eating portion or skip meal time due to having not enough money to buy food	S, K	15	21.4
	T	55	78.6
Having the experience to eat less than you usually consumed due to having not enough money to buy food	S, K	10	14.3
	T	60	85.7
Feeling hungry, but do not have enough money to buy food	S, K	4	5.7
	T	66	94.3
Having an experience of weight loss due to having not enough money to buy food	S, K	12	17.1
	T	58	82.9
Having an experience to not eat all day long due to having not enough money to buy food	S, K	5	7.1
	T	65	92.9

S= happens often, K=sometimes happens, T= never happen

Table 4. Value of Minimum, Maximum, and Mean Diet Quality Subjects

Variable	Minimum	Maximum	Mean[a]±DS/Median[b]
Variety (score)	4	20	17[a]
Overall group food variety	1	5	4[a]
Within-group variety for Protein sources	1	6	6[a]
Adequacy (score)	11	38	22±6,3[b]
Vegetable group (servings/day)	0	4	0,5[a]
Fruit Group (servings/day)	0	12	1,2[a]
Grain Group (servings/day)	0.1	7.3	3.65±1.5[b]
Fiber intake (gr/day)	2	60.1	12.35[a]
Protein Intake (gr/day)	7.5	20	13.25±2.9[b]
Iron Intake (mg/day)	1.7	32.8	9.5[a]
Calcium Intake (mg/day)	41.1	1505.6	415.4[a]
vitamin C Intake (mg/day)	5.8	139.3	112.05[a]
Moderation (score)	3	24	12[a]
Total Fat (% total energy/day)	16.2	59	36±8.59[b]
Saturated Fat (% total energy/day)	3.6	37	16.9[a]
Cholesterol (mg/day)	26	921.9	240[a]
Natrium (mg/day)	94.5	4565.6	559.3[a]
Empty Calory Food (gr/day)	1	41	14±9.3[b]
Overall Balance (skor)	0	6	0.0[a]
Macronutrient Ratio (KH:P:L)	0	6	0.8[a]
Fatty Acid Ratio (PUFA:MUFA:SFA)	0	2	0.0[a]

An assessment by moderation category was used to evaluate food intake that indicated a link to chronic diseases and might need to be restricted, i.e. fats, saturated fats, cholesterol, sodium, and low-nutrient foods. This study showed that the moderation score was low, which means that the food intake of the participants was still not appropriate.

Table 6 shows the total consumption of fats, saturated fats, and junk food which then contributing to the score of the diet quality. According to the balanced nutritional guidelines, the intake of cholesterol and sodium of the participants was relatively good.

The last category was the balance category consisting of macronutrient balance and fatty acid ratio. The balance category evaluated the overall diet in terms of the proportion of energy sources and fatty acid composition. Tables 5 and 6 show that most participants consumed foods high in fat and enough for protein which led to the low intake of carbohydrates. Moreover, the intake of saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) were high (>10%). The recommendation stated that the intake of MUFA should be higher compared to the intake of PUFA.

Table 5. Description of Adequacy in Diet Quality

Variable	components	n	%
Adequacy			
Vegetable Group	Good (≥3-5 servings/day)	2	2.9%
	Adequate (<3-1.5 servings/day)	9	12.9%
	Less (<1.5 servings/day)	59	84.3%
Fruit Group	Good (≥2-3 servings/day)	22	31.4%
	adequate (<2-1 servings/day)	26	37.1%
	Less (<1 servings/day)	22	31.4%
Grain Group	Good (≥3-5 servings/day)	50	71.4%
	Adequate (<3-1.5 servings/day)	17	24.3%
	Less (<1.5 servings/day)	3	4.3%
Fiber Intake	Good (≥20-30 gram/day)	14	20%
	Adequate (<20-10 gram/day)	31	44.3%
	Less (<10 gram/day)	25	35.7%
Protein intake	Good (≥15% energi/day)	21	30%
	Adequate (<15-7.5 energy/day)	49	70%
Iron intake	Good (≥ 100 % RDA mg/day)	3	4.3%
	Adequate (<100%-50%RDA/day)	18	25.7%
	Less (<50 %RDA/day)	49	70%
Calcium Intake	Good (≥ 100 % RDA mg/day)	2	2.9%
	Adequate (<100%-50%RDA/day)	22	31.4%
	Less (<50 %RDA/day)	46	65.7%
Vitamin C intake	Good (≥ 100 % RDA mg/day)	45	64.3%
	Adequate (<100%-50%RDA/day)	11	15.7%
	Less (<50 %RDA/day)	14	20%
Carbohydrate Intake	Excessive (>100% carbohydrate need/day)	27	38.6%
	Adequate (80-100% (>100% carbohydrate need/day)	8	11.4%
	Less (<80% (>100% carbohydrate need/day)	35	50%
Energy intake	Excessive (>100% energy need/day)	27	38.5%
	Adequate (80-100% energy need/day)	18	25.7%
	Less (<80% energy need/day)	25	35.7%

Table 6. Description Moderation in Diet Quality

Variable	Components	n	%
Moderation			
Total Fat	Good (≤30% total energy/day)	17	24.3%
	excessive (>30% total energy/day)	53	75.7%
Saturated Fat	Good (≤10% total energy/day)	10	14.3%
	excessive (>10% total energy/day)	60	85.7%
Cholesterol	Good (≤300 mg/day)	44	62.9%
	excessive (>300 mg/dayi)	26	37.1%
Natrium	Good (≤2400 mg/day)	66	94.3%
	excessive (>2400 mg/day)	4	5.7%
Empty Calory Foods	Good (≤10% total energy/day)	22	31.4%
	excessive (>10% total energy/day)	48	68.6%

Relations between Household Food Security and Diet Quality with CED

Bivariate analysis was conducted to investigate the relationship between household food security and the quality of diet with CED in the study participants. Table 7 shows no significant link (p=0.537) between household food security and chronic energy deficiency.

Table 7. Relations between Household Food Security and Diet Quality with CED

Variable	CED	
	p	**r**
Household Food Security	0.537[b]	0.075
Diet Quality	0.711[a]	0.045

[a] Pearson [b]Rank Spearman

DISCUSSION

Study Participant Characteristics

This study consisted of 38.6% of adolescents (16-20) years. Marriage under the age of 20 years also occurred in Palestine where the prevalence of married women under the age of 18 was 41.4% while in India it was 44.5%.[19,20] Marriage under the age of 20 years is an important risk factor of adolescent pregnancy, where a pregnant young mother and suffers from CED would have a higher risk of perinatal death and give birth to low birth weight (LBW) babies.[21] Education was one of the underlying factors of early marriage in Indonesia.[22] Most of the participants were only graduated from junior and high school. Also, some of the participants only graduated from elementary school.

The prevalence of participants who were at risk of CED based on MUAC measurement was still high (48.6%). However, this prevalence was lower (15.7%) when assessed based on BMI measurement. The higher prevalence of CED based on MUAC measurement in this study was also reported by a preceding study conducted among brides in Probolinggo, which was only 27.3%, and a study conducted in Gorontalo, which was only 28.3%.[6,23] Meanwhile, the prevalence of CED in this study was lower compared to a study conducted in India which reported a prevalence of CED among preconception women of 32.1%.[21] We also found that 14.3% of the participants were over nutrition, which means that there was a double burden of malnutrition in this study.

Household Food Security

75.1% of the study participants in this study had low food security and 1.4% had food insecurity. The prevalence of low food security observed in this study is higher compared to the prevalence of food security among adult women in Malaysia which was only 43.5%.[24] The high prevalence of low food security may be affected by culture and habits among villagers who prioritize the food sufficiency of children. Besides children, working adults with an important role in the household, like fathers, become the main prioritize to get first and more food compared to other family members.[25]

The statement in the HFSSM questionnaire that contributed the highest score is the statement at the household level. A total of 72.9% of the participants were worried they could not buy food before they get another income and could only buy enough food to survive and not being able to stock up on food. Furthermore, 57.1% of the participants could not buy balanced food. This is due to 64.3% of the participants were working but only get low income as factory workers (regional minimum wage) in Semarang District. The other 35.7% of participants were unemployed thus dependent on their parents. The economy is the underlying factor for household food security. Low – middle-income households tend to have low food security. While high-income households often spend more on healthy food.[26]

Low income in a household and the number of family members that live in it will affect the household food security.[27] Low-income households with many dependent members lead to insufficient intake of each family member due to limited access to food. This condition was shown in the statement at the individual level where 21.4% of participants have experienced reducing of skipping a meal due to financial constraint. A total of 14.3% of participants have experienced eating less than usual and 5.7% of participants have experienced hunger due to financial constraints which led them to not be able to purchase food.

Diet Quality

Results show that the average score of diet quality among study participants was low. A total of 80% of the participants had a low-quality diet and the other 20% had high diet quality. The low-quality diet among participants was affected by the discrepancy between the nutrient intake and the recommendation.

The quality of diet consists of four categories which are variation, sufficiency, moderation, and overall balance. The variation evaluates the variation of the overall food and overall protein of the participants. Results show that in general, the participants consume varied food. Insufficiency, moderation, and overall balance categories, the results show a low mean which led to a low score of diet quality. Another study in Spain reported a similar finding, where female adolescents in the said study were found to consume varied food, but the other categories (sufficiency, moderation, and overall balance) contributed a low score to the total diet quality score.[28]

The sufficiency of vegetables, iron, and calcium in participants recorded a low mean. The average vegetable intake was only 0.5 portions/day compared to the recommended 3-5 portions/day. Similarly, a study in Spain also showed that the average vegetable and fruit intake among female adolescents was low.[28] Iron and calcium are important minerals needed during pregnancy. Most of the participants consumed iron and calcium-rich food which are not following the recommendation. A preceding study on pre-pregnancy women in Bogor supports this finding. The said study found that the iron and calcium intake pre-pregnancy among women was low.[29] Low intake of vegetable, fruit, and other micronutrients like iron and calcium can cause low diet quality among preconception women.[30]

The DQI-I score shows an imbalance of total fat, total saturated fat, and low-nutrients food compare to the requirement. Based on the DQI-I, the recommended consumption of total fat per day is <30% total energy/day, saturated fat is <10% total energy/day and low-nutrients food is <10% total energy/day. However, most of the participants still consumed high-fat and low-nutrient food like sugar, which contributes a low score to their diet quality score. A high intake of total and saturated fat resulted from the high consumption of fried food.

A low score was also found in the overall balance category. The high intake of fat among participants hinders the balance of other macronutrient intakes. Very few of the participants consumed total fat less than 30% of the total energy/day. This has an impact on the low intake of carbohydrates and protein. Results show that the consumption of carbohydrates and protein among the participants was relatively low compared to the recommendation even though many of the participants consumed sufficient protein. The high intake of fat increases the total energy intake which then affects the nutritional status.[31]

This study also reported an imbalance of fatty acids. The average intake of SFA was higher than PUFA and MUFA. This finding is in line with the findings in Iran which showed that the high intake of SFA was higher than PUFA and MUFA in female adolescents. This may be caused by the shift in the diet pattern where adolescents tend to consume

more high-energy foods like fast food and snack.[32] Fast food has become an easy and cheap option even though it contributes to excessive energy intake.

Relations between Household Food Security and Diet Quality with CED

Based on the test, household food security is not significantly correlated to CED among preconception women (p=0.536). Very limited studies evaluated household food security of preconception women using the HFSSM questionnaire. This study is supported by studies conducted in Bangladesh among pregnant women and in Kenya among women of reproductive age between 15-49 years which found no significant relations between household food security with nutritional status by BMI measurement.[33,34]Yet, the results of these studies contradicted the findings of a study conducted in Ethiopia which found a significant status between household food security and nutritional status by BMI measurement.[35]

In this study, household food security did not indicate low energy and protein intake. This can be seen from the high (and sometimes excessive) intake of energy and protein among the participants which was 64.2%. The high energy intake was acquired from sugar and fat intake and low fiber which tend to be low in micronutrients. Thus, the CED rate was not high as the participants were close to being over nutrient.

On the other hand, diet patterns in society also went through a shift where most households with low food security only paid attention to quantity and not the quality of the food. For example, by accessing fried food and low intake of vegetables and fruit. A study in Kenya backed this finding. It reported a shift in diet patterns in the household with low food security. Fried food was chosen because it is cheap and easy to consume compared to vegetables and fruit even though fried food is high in energy and is unhealthy.[34,36] Statements from most of the participants portray this by stating that they cannot consume a balanced diet due to financial constraints. Also, access to fried food is easier, as most of the participants work as factory workers where access to buy food was limited. A limited break time is a reason that the participants consumed an unhealthy diet that in prolonged time will cause overnutrition.[37]

Very limited studies evaluated the diet quality of preconception women using the DQI-I questionnaire. The overall diet quality of the participants in this study did not show a significant correlation with CED (p=0.711). This finding is supported by findings of other studies conducted in the USA among female adolescents which explains the quality diet of female adolescents did not have a significant relationship with their nutritional status based on BMI.[38] However, this finding contradicts a preceding study that reported a significant relationship between diet quality and BMI before pregnancy among pregnant women.[1] Another direct factor that may cause CED in preconception women is infection. During infection or another sickness, we tend to lose appetite thus lead to malnutrition. If it continues for a prolonged time, it can cause CED.[39,40]

This disagreement may happen because CED in preconception women is not only affected by household food security and diet quality. Energy and protein intake plays an important role in the incidence of CED among preconception women. A low protein and energy intake for a prolonged time can cause CED.[23] In this study, most of the participants had sufficient energy intake, moreover, 38.5% of the participants had excessive energy intake. Looking at the sufficiency of staple food, the average consumption of staple food of a participant was 3.65 portions/day and is categorized as sufficient. Also, the protein intake of the participants was categorized as sufficient and good, thus it can be concluded that by quantity the intake of the participants was adequate.

This study differs from the Central Java Individual Food Consumption Survey in 2014 which stated that women who live in the village tend to have low consumption of energy and protein thus causing malnutrition. However, in this study, the participants had high energy high-fat diets thus tend to be over nutrition. This is supported by the result that shows 14.35% of the participants had over nutrition. Diets that tend to be over nutritious is a low vegetable and fruit yet high fat intake.[15,28]

Diet quality is influenced by household food security, where a family with high food security will have a good diet quality.[12,41] A low household food security caused most of the participants in this study to consume cheap and easily accessed food like fried food. It was shown by the high intake of fat among

the participants in the moderation category which was more than 30% of total energy. A low moderation score will affect the overall balance score where most of the participants had a high intake of fat and impair the balance of other macronutrients. A high intake of fat will contribute to excessive energy thus cause a build-up of fat on the adipose tissue and plays an important role in the incidence of overnutrition.[42]

CONCLUSIONS

A total of 48.1% of participants were at risk of CED, but only 15% of preconception women were categorized as CED. The household food security of the participants was mostly low (75.1%). Most of the participants (80%) had low-quality diets where the mean diet quality was 51.77. there was no significant correlation between household food security and diet quality with CED.

REFERENCES

1. Shin D, Lee KW, Song WO. Pre-Pregnancy Weight Status Is Associated with Diet Quality and Nutritional Biomarkers during Pregnancy. Nutrients. 2016 Mar 11;8(3):1–12.
2. Stephanie P, Kartika SKA. Gambaran Kejadian Kurang Energi Kronik dan Pola Makan Wanita Usia Subur di Desa Pesinggahan Kecamatan Klungkung Bali 2014. E-Jurnal Medika Udayana. 2016 May 31;5(6):1–6.
3. Das S, Bose K. Body Mass Index, and Chronic Energy Deficiency among Adult Santals of Purulia District, West Bengal, India. Journal of Human Sciences. 2010 Jul 25;7(2):488–503.
4. Kementerian Kesehatan RI. Riset Kesehatan Dasar 2013 (RISKESDAS 2013). Indonesia: Kementerian Kesehatan Republik Indonesia; 2013.
5. Budiono I. Pengembangan Model Indeks Pembangunan Gizi. KEMAS: Jurnal Kesehatan Masyarakat. 2013 Jan 3;8(2):113–20.
6. Putri SI, Sumarmi S. Perbandingan Konsumsi Zat Gizi, Status Gizi, dan Kadar Hemoglobin Pengantin Wanita di Wilayah Pantai dan Pertanian Kabupaten Probolinggo. Media Gizi Indonesia. 2012 Jan;9(1):1–6.
7. Kementerian Kesehatan Republik Indonesia. Pusat Data dan Informasi Kementrian Kesehatan RI. Kementerian Kesehatan Republik Indonesia; 2014.
8. Rahmi L. Faktor-Faktor yang Berhubungan dengan Kekurangan Energi Kronik (KEK) pada Ibu Hamil di Puskesmas Belimbing Padang Tahun 2016. Jurnal Kesehatan Medika Saintika. 2017 Jun 12;8(1):35–46.
9. Arimond M, Elin L, Wiesmann D, Joseph M, Carriquiry A. Dietary Diversity as a Measure of Women's Diet Quality in Resource-Poor Areas: Results from Rural Bangladesh Site. Food and Nutrition Technical Assistance. 2008;58.
10. Potdar RD, Sahariah SA, Gandhi M, Kehoe SH, Brown N, Sane H, et al. Improving women's diet quality preconceptionally and during gestation: effects on birth weight and prevalence of low birth weight--a randomized controlled efficacy trial in India (Mumbai Maternal Nutrition Project). Am J Clin Nutr. 2014 Nov;100(5):1257–68.
11. Na M, Mehra S, Christian P, Ali H, Shaikh S, Shamim AA, et al. Maternal Dietary Diversity Decreases with Household Food Insecurity in Rural Bangladesh: A Longitudinal Analysis. J Nutr. 2016 Oct;146(10):2109–16.
12. Abraham S, Miruts G, Shumye A. Magnitude of chronic energy deficiency and its associated factors among women of reproductive age in the Kunama population, Tigray, Ethiopia, in 2014. BMC Nutrition. 2015 Jun 22;1(1):12.
13. Kementerian Kesehatan Republik Indonesia. Profil Kesehatan Provinsi Jawa Tengah Tahun 2016. Kementerian Kesehatan Republik Indonesia; 2016.
14. Kementerian Kesehatan Republik Indonesia. Profil Kesehatan Kabupaten Semarang 2014. Kementerian Kesehatan Republik Indonesia; 2014.
15. Santoso B, Sulistiowati E, Fajarwati T, Pambudi J. Survei Konsumsi Makanan Individu Provinsi Jawa Tengah 2014. Semarang: Badan Penelitian dan Pengembangan; 2014.
16. Usfar AA, Fahmida U, Februhartanty J. Household food security status measured by the US-Household Food Security/Hunger Survey Module (US-FSSM) is in line with coping strategy indicators found in urban and rural Indonesia. Asia Pac J Clin Nutr. 2007;16(2):368–74.
17. Adereti DT, Fasina OO. Gender analysis of food security status of rural households in Ondo

State, Nigeria. Russ Agricult Sci. 2017 Jul 1;43(4):353–60.
18. Guerrero MLP, Pérez-Rodríguez F. Diet Quality Indices for Nutrition Assessment: Types and Applications. In: Functional Food - Improve Health through Adequate Food [Internet]. IntechOpen; 2017 [cited 2020 Nov 16]. Available from: https://www.intechopen.com/books/functional-food-improve-health-through-adequate-food/diet-quality-indices-for-nutrition-assessment-types-and-applications
19. Ghrayeb FA, Rusli A M, Ismail I M, Ghrayeb NF, Al Rifai A. Prevalence of Early Marriage among Women in Rural Palestinian Community: A Cross-Sectional Study. International Medical Journal. 2015 Oct 3;22(4):291–4.
20. Raj A, Saggurti N, Balaiah D, Silverman JG. Prevalence of Child Marriage and its Impact on the Fertility and Fertility Control Behaviors of Young Women in India. Lancet. 2009 May 30;373(9678):1883–9.
21. Islam A, Islam N, Bharati P, Aik S, Hossain G. Socio-economic and demographic factors influencing nutritional status among early childbearing young mothers in Bangladesh. BMC Women's Health. 2016 26;16(1):58.
22. Rumble L, Peterman A, Irdiana N, Triyana M, Minnick E. An empirical exploration of female child marriage determinants in Indonesia. BMC Public Health. 2018 Mar 27;18(1):407.
23. Hubu N, Nuryani N, Hano YH. Pengetahuan, Asupan Energy dan Zat Gizi Berhubungan dengan Kekurangan Energy Kronis pada Wanita Prakonsepsi. GJPH. 2018 Apr 19;1(1):015.
24. Atiqah AN, Norazmir M, Anuar MK, Fahmi MM, Norazlanshah H. Food security status: it's association with inflammatory marker and lipid profile among young adult. [Internet]. undefined. 2015 [cited 2020 Nov 16]. Available from: /paper/Food-security-status%3A-it%27s-association-with-marker-Atiqah-Norazmir/557339a0f893c6e1e813a700a3ad236a335e5b50
25. Cordeiro LS, Wilde PE, Semu H, Levinson FJ. Household food security is inversely associated with undernutrition among adolescents from Kilosa, Tanzania. J Nutr. 2012 Sep;142(9):1741–7.
26. Hackett M, Melgar-Quiñonez H, Taylor CA, Alvarez Uribe MC. Factors associated with the household food security of participants of the MANA food supplement program in Colombia. Arch Latinoam Nutr. 2010 Mar;60(1):42–7.
27. Ahmed UI, Ying L, Bashir MK, Abid M, Zulfiqar F. Status and determinants of small farming households' food security and role of market access in enhancing food security in rural Pakistan. PLoS One. 2017;12(10):e0185466.
28. Mariscal-Arcas M, Romaguera D, Rivas A, Feriche B, Pons A, Tur JA, et al. Diet quality of young people in southern Spain evaluated by a Mediterranean adaptation of the Diet Quality Index-International (DQI-I). Br J Nutr. 2007 Dec;98(6):1267–73.
29. Madanijah S, Briawan D, Rimbawan R, Zulaikhah Z, Andarwulan N, Nuraida L, et al. Nutritional status of pre-pregnant and pregnant women residing in Bogor district, Indonesia: a cross-sectional dietary and nutrient intake study. Br J Nutr. 2016;116 Suppl 1:S57-66.
30. Shamim AA, Mashreky SR, Ferdous T, Tegenfeldt K, Roy S, Rahman AKMF, et al. Pregnant Women Diet Quality and Its Sociodemographic Determinants in Southwestern Bangladesh. Food Nutr Bull. 2016 Mar;37(1):14–26.
31. Austin GL, Ogden LG, Hill JO. Trends in carbohydrate, fat, and protein intakes and association with energy intake in normal-weight, overweight, and obese individuals: 1971-2006. Am J Clin Nutr. 2011 Apr;93(4):836–43.
32. Mohseni-Takalloo S, Mirmiran P, Hosseini-Esfahani F, Azizi F. Dietary Fat Intake and Its Relationship with Serum Lipid Profiles in Tehranian Adolescents. JFNR. 2014 Jun 16;2(6):330–4.
33. Mridha MK, Matias SL, Arnold CD, Dewey KG. Factors associated with nutritional status and dietary practices of Bangladeshi adolescents in early pregnancy. Ann N Y Acad Sci. 2018 Feb 18;1416:66–76.
34. Keino S, Plasqui G, van den Borne B. Household food insecurity access: a predictor of overweight and underweight among Kenyan

women. Agriculture & Food Security. 2014 Jan 28;3(1):2.
35. Abdu J, Kahssay M, Gebremedhin M. Household Food Insecurity, Underweight Status, and Associated Characteristics among Women of Reproductive Age Group in Assayita District, Afar Regional State, Ethiopia [Internet]. Vol. 2018, Journal of Environmental and Public Health. Hindawi; 2018 [cited 2020 Nov 16]. p. e7659204. Available from: https://www.hindawi.com/journals/jeph/2018/7659204/
36. Gulliford MC, Mahabir D, Rocke B. Food insecurity, food choices, and body mass index in adults: nutrition transition in Trinidad and Tobago. Int J Epidemiol. 2003 Aug;32(4):508–16.
37. Nobrega S, Champagne N, Abreu M, Goldstein-Gelb M, Montano M, Lopez I, et al. Obesity/Overweight and the Role of Working Conditions: A Qualitative, Participatory Investigation. Health Promot Pract. 2016 Jan;17(1):127–36.
38. Feskanich D, Rockett HRH, Colditz GA. Modifying the Healthy Eating Index to assess diet quality in children and adolescents. J Am Diet Assoc. 2004 Sep;104(9):1375–83.
39. Dobner J, Kaser S. Body mass index and the risk of infection - from underweight to obesity. Clin Microbiol Infect. 2018 Jan;24(1):24–8.
40. Katona P, Katona-Apte J. The Interaction between Nutrition and Infection. CLIN INFECT DIS. 2008 May 15;46(10):1582–8.
41. Leung CW, Epel ES, Ritchie LD, Crawford PB, Laraia BA. Food Insecurity Is Inversely Associated with Diet Quality of Lower-Income Adults. Journal of the Academy of Nutrition and Dietetics. 2014 Dec;114(12):1943-1953.e2.
42. Coelho DF, Pereira-Lancha LO, Chaves DS, Diwan D, Ferraz R, Campos-Ferraz PL, et al. Effect of high-fat diets on body composition, lipid metabolism and insulin sensitivity, and the role of exercise on these parameters. Brazilian Journal of Medical and Biological Research. 2011 Oct;44(10):966–72.

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Correlation between wrist circumference with blood pressure and creatinine level among elderly

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ABSTRACT

Background: Hypertension is a highly prevalent health problem which incidence is greatest among the elderly. Hypertension may increase creatinine level and leads to other health problems like diabetes mellitus, kidney damage, and cardiovascular disease. Wrist circumference is a simple anthropometric measurement that can be used to identify hypertension and increasing level of serum creatinine.
Objectives: To analyze the correlation of wrist circumference with blood pressure and creatinine level among the elderly.
Materials and Methods: This was a cross-sectional study with a purposive sampling method. Subjects of this study were 84 women aged 60 years old or above at Unit Rehabilitasi Sosial Pucang Gading Semarang. The independent variable of this study was wrist circumference, and the dependent variables were systolic blood pressure, diastolic blood pressure, and creatinine level. The result was analyzed using the Spearman-rho test.
Results: The participants of this research were 49% women aged 60-65 years old, with an average age was 65.5 years old. The prevalence of hypertension was 61.9%. Most hypertension incidence in this research was caused by high systolic blood pressure (50%), and the rest was caused by high diastolic blood pressure (3.9%) and both (46.1%). The level of creatinine was normal with an average level was 0.75 mg/dL. There was no correlation of wrist circumference with systolic blood pressure systolic (r=0.15; p=0.19), diastolic blood pressure (r=0.1; p=0.38), and creatinine serum (r=0.18; p=0.09) among elderly.
Conclusions: There was no correlation of wrist circumference with blood pressure and creatinine level among the elderly.

Keywords: Wrist circumference; Blood pressure; Creatinine level; Elderly

BACKGROUND

According to World Health Organization (WHO), the elderly are individuals with the chronological age of 60 years or more. The Elderly is the last phase of the human life cycle and is initiated by the aging process. This aging process is contributing to frailty which posing the elderly to diseases, both infectious and non-infectious ones.[1] Elderly population in Indonesia is 9.7% based on Riskesdas 2019 and was predicted to be increased more than 2 folds in 2050. Based on Riskesdas 2018 data, the most prevalent disease for the elderly in Indonesia is hypertension.[2]

Hypertension is a condition in which a person has systolic blood pressure ≥140 mmHg and/or diastolic blood pressure ≥of 90 mmHg.[3] Hypertension increase the risk of diabetes and coronary heart disease. Moreover, hypertension could lead to chronic kidney disease through the increased blood creatinine level.[4] Prevalence of hypertension in Indonesia is 34.1% where 59.4% of citizens with the age of >54 years are having hypertension.[2] Hypertension prevalence in Central Java is 37.6%, even higher than national prevalence.[5] Health Department of Central Java stated hypertension prevalence in Semarang City in 2018 is 6.3%, whereas much as 14.9% of the citizen with the age of >45 years are having hypertension.[6]

Hypertension is preventable and treatable by early detection. Hypertension is usually detected by performing a blood pressure examination using a sphygmomanometer. However, a simple anthropometry measurement that is quick and easy could be considered as a parameter to detect hypertension, one of them is wrist circumference.[7-9] Measurement of waist circumference is easier to be

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done since it does not require ratio calculations and trained individuals.[10] Inflammation state in hypertension indirectly decreases the bone mineral density, including on wrist circumference.[9]

Besides as a predictor of hypertension, many studies show the correlation between waist circumference and creatinine levels. A large-scale study conducted on 2.400 respondents in Iran shows that simple anthropometric measurement as wrist and neck circumference has a significant correlation with creatinine level and able to predict the risk of chronic kidney disease in adolescents and adults.[11]

The population-based study associated with the correlation between waist circumference with blood pressure and creatinine levels in Indonesia is still limited, especially among the elderly in Semarang City. The purpose of this study is to analyze whether waist circumference is associated with blood pressure and creatinine levels among the elderly.

MATERIALS AND METHODS

This study is an observational study with a cross-sectional design among 84 elderly in Unit Rehabilitasi Sosial Pucang Gading Semarang from May to July 2019. Samples were collected via the purposive sampling method. Institutional Review Board of Ethical Clearance (*Komisi Etik Penelitian Kesehatan*/KEPK) from the Medical Faculty of Diponegoro University has approved this study and publish ethical clearance141/EC/FK-RSDK/1V/2018.

Inclusion criteria in this study are women with the age of 60 years or more, communicative, and willing to be involved in this study by signing informed consent. Exclusion criteria are the history of/currently taking long-term corticosteroid medication, routinely consuming calcium supplements, and having chronic diseases such as diabetes, kidney failure, liver disease, gastrointestinal disturbance, and thyroid dysfunction.

The dependent variable in this study is wrist circumference. Wrist circumference measurement was performed using measuring tape with 0,1 cm accuracy. The measuring tape was wrapped around the wrist from the Lister tubercle on the distal radius to the distal ulnae on the right hand in a sitting position. The independent variables are blood pressure and creatinine level. Blood pressure measurement was performed twice using a digital sphygmomanometer. Blood sampling was performed by laboratory personnel and creatinine levels were tested using a photometer automatic chemistry analyzer.

Data analysis was performed using the computer-based statistic program SPSS, consisting of univariate and bivariate analysis. Univariate analysis was performed to describe the subject's characteristics such as age, body mass index (BMI), blood pressure, and creatinine levels. Bivariate analysis was performed to analyze the correlation between waist circumference with blood pressure and creatinine levels by the Spearman correlation test. Coefficient correlation (r) approaching 0 shows a weak correlation among variables and r value approaching -1 or 1 shows a strong correlation. P-value <0.05 shows a significant correlation between variables.

RESULTS

Table 1 shows the anthropometry and biochemistry characteristics of the subjects in this study. Based on Table 1, most of the subjects are 60-65 years old (58.3%) with an average of 65.5 years old. Forty-three subjects (51.2%) are obese.

The prevalence of hypertension in this study is 61.9%. As much as 50% of hypertension in the subjects of this study is marked by high systolic pressure. Hypertension with high diastolic blood pressure is only 3.9% and the rest (46.1%) is caused by high systolic and diastolic blood pressure. The biochemical test shows creatinine levels on most subjects are still in the normal range with an average of 0.75 mg/dL.

Table 2 shows the correlation between waist circumference and systolic blood pressure, diastolic blood pressure, and creatinine level to analyze the strength and direction of the variables.

Statistical analysis shows there is no correlation between waist circumference and systolic blood pressure, diastolic blood pressure, and creatinine level with a p-value > 0.05.

DISCUSSION

The prevalence of hypertension in this study, whether based on systolic or diastolic blood pressure or both, is 61.9%. This result is in accordance and able to show highly prevalent hypertension among the elderly in Indonesia that reach 59.4% in 2018.[2] This number shows the importance of a quick and good indicator to detect hypertension.

Previous studies show that waist circumference correlated with hypertension and increased creatinine level.[11-13] One of the factors influencing bone growth is a hormone called insulin-like growth factor 1 (IGF-1). This hormone is

stimulated by growth hormone and synthesized in the liver. Insulin-like growth factor 1 stimulates nitric oxide (NO) formation by endothelial cells and cells in blood vessels smooth muscle and stimulates Na-K-ATPase pump, therefore causing blood vessels relaxation and decrease blood pressure.[14,15] Wrist circumference which is one of the indicators of bone surface dimension could be used to predict hypertension risk and increased creatinine level. [16]

Table 1. Baseline Characteristics

	Numbers (%)
Total subjects	84
Age (years)	
60 – 65	49 (58.3%)
>65	35 (41.7%)
Body Mass Index (kg/m^2)	
< 18.5	2 (2.4%)
18.5 – 22.9	30 (35.7%)
23 – 24.9	9 (10.7%)
≥ 25	43 (51.2%)
Systolic Blood Pressure (mmHg)	
Normal	34 (40.5%)
Hypertension	50 (59.5%)
Diastolic Blood Pressure (mmHg)	
Normal	58 (69.1%)
Hypertension	26 (30.9%)
Creatinine Level (mg/dL)	
Normal	80 (95.2%)
High	4 (4.8%)
Wrist Circumference (cm)	15.35 ± 0.14

Note: BMI: Body Mass Index

Table 2. Correlation between wrist circumference with blood pressure and creatinine level

Variable	**Coefficient correlation (r)[a]**	**Sig. (*p-value)***
Systolic blood pressure (mmHg)	0.15	0.19
Diastolic blood pressure (mmHg)	0.10	0.38
Creatinine level (mg/dL)	0.18	0.09

Note: [a]pearman test

Physiologically, aging will cause the thickening and hardening of blood vessels. This condition is associated with chronic inflammation in blood vessels and increased oxidative stress thus decreasing the elasticity of blood vessels and inhibits blood flow. The blockaded blood vessels will increase heart workload in pumping blood, therefore increase blood pressure.[17] Blocked blood vessels in the kidney will cause a decreased glomerular filtration rate. This will result in decreased creatinine excretion and the risk of increased creatinine in blood among the elderly.[18]

There was not found a significant correlation between waist circumference with blood pressure and creatinine level in this study. This was presumably caused by the influence of several factors such as physical activity, food intake, previous history of the disease, and other factors that were not included in this study.

Increased physical activity will decrease the catecholamine level in the blood. This condition will indirectly decrease the sympathetic nervous system which responds to the decreased blood flow rate and vasodilatation.[19] Moreover, blood pressure and creatinine levels are easily influenced by food intake. Elderly with poor food intakes such as consuming high calories food, high sodium, protein, fat and low in potassium and fiber will increase the risk of increased blood pressure and creatinine level.[20-25] High sodium and low potassium food will influence fluid volume and blood vessel elasticity which contribute to increased blood pressure.[20, 21]

High protein intake, especially animal-based protein is associated with hypertension and kidney failure.[22] Fiber is known for its potency in increasing insulin sensitivity and endothelial cells which will contribute to blood vessel elasticity.[23]

High fat and dense calorie food will increase excess caloric intake and fat deposition in blood vessels that will result in blocked blood flow from and to organs, and disturbing its physiological condition.[24, 25] Obesity and hyperglycemia will increase the risk of increased blood pressure and blood creatinine level. Inflammation caused by obesity and hyperglycemia will increase free radicals and decrease adiponectin, and result in the damage and elasticity impairment of the blood vessels.[24,25] This could also increase blood pressure and blood creatinine levels if kidney blood vessels are affected.[17, 18]

CONCLUSIONS

Wrist circumference is not correlated with blood pressure and creatinine levels in this study. This is presumably caused by several factors such as physical activity, food intake, and previous history of the disease which could influence the correlation between waist circumference with blood pressure and creatinine level.

Further studies should be carried out to validate the correlation between waist circumference with blood pressure and creatinine level among the elderly by considering the food intake and physical activity of the elderly.

REFERENCES

1. World Health Organization. World Report on Ageing and Health. Geneva, Switzerland: WHO; 2015.
2. Kementerian Kesehatan RI. Laporan Nasional Riset Kesehatan Dasar 2018. Kementerian Kesehatan RI. Jakarta:2018.
3. Chobanian A V, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL, et al. The Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. Journal of the American Medical Association. 2003;289:2560-2571.
4. Ghaffari S, Pourafkari L, Tajlil A, Sahebihagh MH, Mohammadpoorasl A, Tabrizi JS, et al. The Prevalence, Awareness, and Control Rate of Hypertension among Elderly in Northwest of Iran. Journal of Cardiovascular and Thoracic Research. 2016;8(4):176–82.
5. Dinas Kesehatan Jawa Tengah. Profil Kesehatan Provinsi Jawa Tengah Tahun 2018. Semarang, Jawa Tengah: Dinkes Jateng; 2018.
6. Dinas Kesehatan Kota Semarang. Profil Kesehatan Kota Semarang Tahun 2018. Semarang, Jawa Tengah: DKK Semarang; 2018.
7. Kalantari S, Khalili D, Asgari S, Fahimfar N, Hadaegh F, Tohidi M, et al. Predictors of Early Adulthood Hypertension during Adolescence: A Population-based Cohort Study. BMC Public Health. 2017;17(1):1–8.
8. Zampetti S, Campagna G, Lucantoni F, Marandola L, D'Onofrio L, Chiesa C, et al. Wrist Circumference is Associated with Increased Systolic Blood Pressure in Children with Overweight/Obesity Article. Hypertension Research. 2018;41(3):193–7.
9. Mohebi R, Mohebi A, Sheikholeslami F, Azizi F, Hadaegh F. Wrist Circumference as A Novel Predictor of Hypertension and Cardiovascular Disease: Results of A Decade Follow Up in A West Asian Cohort. Journal of the American Society of Hypertension. 2014;8(11):800–7.
10. Shafiee G, Qorbani M, Heshmat R, Djalalinia S. Wrist Circumference as a Novel Predictor of Obesity in Children and Adolescents: the CASPIAN-IV Study. Journal of pediatric endrocinology & metabolism. 2018;31(7):1-9.
11. Mehrkash M, Heshmat R, Qorbani M, Motlagh ME, Djalalinia S, Zamani S, et al. Association of Neck, Wrist and Hip Circumferences with Kidney Function in Children and Adolescents: The CASPIAN- V Study. Journal of Pediatric Research. 2019;6(3):234–41.
12. Mitrea A, Soare A, Popa SG, Tudor MN, Mota M, Pozzilli P. Wrist Circumference: an Independent Predictor of Both Insulin Resistance and Chronic Kidney Disease in an Elderly Population. Romanian Journal of Diabetes Nutrition and Metabolic Disease. 2013;20(3):323–9.
13. Li Y, Liu Y, He J, Ma P, Yu L, Sun G. The Association of Wrist Circumference with Hypertension in Northeastern Chinese Residents in Comparison with other Anthropometric Obesity Indices. PeerJ. 2019;2019(8):1–16.
14. Hunt KJ, Lukanova A, Rinaldi S, Lundin E, Norat T, Palmqvist R, et al. A Potential Inverse Association Between Insulin-Like Growth Factor I and Hypertension in a Cross-Sectional Study. Annals of Epidemiology. 2006;16(7):563–71.

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15. Zhang L, Curhan GC, Forman JP. Plasma Insulin-like Growth Factor-1 Level and Risk of Incident Hypertension in Nondiabetic Women. Journal of Hypertension. 2011;29(2):229–35.
16. Masoud MI, Masoud I, Kent RL, Gowharji N, Hassan AH, Cohen LE. Relationship between Blood-Spot Insulin-like Growth Factor 1 Levels and Hand-Wrist Assessment of Skeletal Maturity. American Journal of Orthodontics and Dentofacial Orthopedics. 2009;136(1):59–64.
17. Buford TW. Hypertension and Ageing. Ageing Research Reviews. 2016; 26: 96-111.
18. Weinstein JR, Anderson S. The Aging Kidney: Physiological Change. Advances in Chronic Kidney Disease. 2010; 17(4): P302-307.
19. Kokkinos PF, Giannelou A, Athanasiosmanolis, Pittaras A. Physical Activity in the Prevention and Management of High Blood Pressure. Hellenic Journal of Cardiology. 2009;50:52-9.
20. Appel LJ. The Effects of Dietary Factors on Blood Pressure. Cardiology Clinics. 2017;35(2):197–212.
21. Naismith DJ, Braschi A. The Effect of Low-Dose Potassium Supplementation on Blood Pressure in Apparently Healthy Volunteers. British Journal of Nutrition. 2003;90(1):53-60.
22. Kamper A-L, Strandgaard S. Long-Term Effects of High-Protein Diets on Renal Function. Annual Review of Nutrition. 2017;37(1):347–69.
23. Streppel MT, Arends LR, Veer Pvt, Grobbee DE, Geleijnse JM. Dietary Fiber and Blood Pressure: A Meta-analysis of Randomized Placebo-Controlled Trials. Arch Intern Med. 2005;165:150-6.
24. Kim DH, Kim C, Ding EL, Townsend MK, Lipsitz LA. Adiponectin Levels and the Risk of Hypertension: a Systematic Review And Meta-Analysis. Hypertension. 2013;62(1):27-32.
25. Donmezi S, Gokalp O, Dogan M, Vural H, Yigit B. Effects of Short-Term Hyperglycemia on the Vasoconstriction of the Aorta. Turkish Journal of Medical Sciences. 2014;44:941-5.

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Catfish (Clarias sp.) as an animal protein source to improve serum albumin levels of hemodialysis patients

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ABSTRACT

Background: Hemodialysis patients often experience hypoalbuminemia complications, which occur mainly due to decreased synthesis due to inflammation, lack of protein intake, the fluid status of patients, and losses from the dialysate. Another problem in hemodialysis is malnutrition, with a prevalence between 23–73% globally. Gastrointestinal disorders such as nausea, vomiting, and decreased appetite also often occur in hemodialysis. Therefore, hemodialysis patients need to get nutritional support, which can be given in the form of catfish abon, one of the local Indonesian food.
Objective: To determine the effectiveness of the use of catfish as a source of animal protein to improve the albumin levels of hemodialysis patients.
Materials and Methods: This was a quasi-experimental study with a pre-post test design. This study involved 34 hemodialysis patients as subjects, with inclusion criteria, were routinely two times a week, aged >18 years, willing to be the subject and follow the research procedures, have albumin levels ≥3.0 g/dL, and no catfish allergies. Patients with anasarca edema, experiencing complications of diabetes mellitus and malignancy were excluded. The dependent variable was albumin content, while the independent variable was catfish as an animal protein source. Data were analyzed univariate and bivariate by Fisher's Exact test.
Results: Fisher's Exact test results on the effectiveness of using catfish as an animal protein source to improve albumin levels of hemodialysis patients showed p-value=0.048.
Conclusion: The use of catfish as an effective animal protein source significantly affected on improving albumin levels in hemodialysis patients.

Keywords: Nutritional support; Catfish (Clarias sp.); Albumin levels; Hemodialysis patients.

BACKGROUND

Chronic kidney disease is a global widespread epidemic disease, which a prevalence rate is 5-15%. The incidence rate of end-stage renal disease patients requiring dialysis is also increasing (1). Basic Health Research in Indonesia shows that the prevalence of chronic kidney disease nationally increased from 0.2% in 2013 to 0.38% in 2018. Province Special Region of Yogyakarta, where the Panembahan Senopati Hospital is located, ranked 12th nationally for the prevalence of chronic kidney disease (2).

Panembahan Senopati Bantul Hospital is a large type B hospital, which obtained a hospital-level plenary accreditation certificate. Panembahan Senopati Bantul Hospital is one of the hospitals that has a Hemodialysis Unit in Bantul Regency. Based on data from the 2013 Panembahan Senopati Bantul General Hospital annual report, the number of chronic kidney disease patients undergoing hemodialysis is increasing every year. In 2011, the number of routine hemodialysis patients was 111 people, in 2012, it increased to 125 people, in 2013, it became 142 people, and in 2014 it increased again to 144 people. The hemodialysis patient is accommodated in a room (Hemodialysis Unit) with a capacity of 22 beds.

Hemodialysis patients often experience hypoalbuminemia complications, which occur mainly due to decreased synthesis due to inflammation, lack of protein intake, the fluid status of patients, and losses from dialysate (3,4). Another problem that often arises in hemodialysis is malnutrition, with a prevalence between 23–73%

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globally (5). Gastrointestinal disorders such as nausea, vomiting, and decreased appetite also often occur in hemodialysis patients (6).

Protein-energy malnutrition is a major risk factor for mortality and inflammation; the presence of co-morbid conditions like cardiovascular disease increases this risk further (7). Both low protein intake and a high state of inflammation are associated with low serum albumin in maintenance hemodialysis patients (8). Therefore, hemodialysis patients need to get nutritional support, one of them with catfish.

Catfish have many advantages in terms of nutritional content but are still often underestimated by the community because of conventional processing. Catfish can be processed into various types of food products, one of which is catfish *abon*, so it is expected to increase the acceptability and intake of hemodialysis patients. *Abon* is one of the local food in Indonesia. This study was conducted to determine the effectiveness of the use of catfish as a source of animal protein to improve the albumin levels of hemodialysis patients.

MATERIALS AND METHODS

This research was conducted at the Hemodialysis Unit of Panembahan Senopati Bantul Hospital in January–December 2017. This research type was a quasi-experimental design with a pre-post design. This study involved 34 subjects with inclusion criteria for routine hemodialysis patients twice a week, aged >18 years, willing to be the subject of research and follow the research procedures, have albumin levels ≥3.0 g/dL, and there was no catfish allergy. Whereas patients with anasarca edema, complications of diabetes mellitus, and malignancy were excluded from the study.

An experiment in the form of providing nutritional support for catfish *abon* for 21 days, based on the half-life of albumin. The nutritional content of catfish *abon* which is given as nutritional support is shown in Table 1. With a protein requirement of 1.2 g/kg body weight per day, nutritional support for hemodialysis patients can be given protein additions of ±0.2 g/kg body weight per day. The protein content of catfish *abon* based on proximate test results is 54.89 grams per 100 grams of catfish *abon*. With the nutritional support needs of hemodialysis patients of 0.2 g/kg ideal body weight/day, the need for *abon* is 0.36 g/kg ideal bodyweight/day.

Table 1. Nutrient content of catfish *abon* in the study

Nutrient	Unit	Nutrient content
Energy	kcal	421.294
Protein	g	54.894
Fat	g	25.159
Carbohydrate	g	326.091
Crude fiber	g	0.127
Sodium	ppm	1619.25
Potassium	ppm	421.294

Before being given an intervention (pre-intervention), subjects first measured levels of albumin. Then the subjects were given the intervention to provide nutritional support in the form of catfish *abon* for 21 days and were asked to record all the food and drinks they consume every day in the food record questionnaire. To ensure subjects consume catfish *abon* according to the nutritional support needs of protein and record food and drink intake every day, the researchers did the monitoring by sending a reminder in the form of a Short Message Service reminder. Messages sent to the subject only contained a reminder to consume *abon* given in the study but did not contain material related to dietary therapy on hemodialysis. Nutrition monitoring using SMS is proven to improve dietary adherence of hemodialysis patients (9). If the patient adheres to the given diet, the nutritional status of the patient will increase and can indirectly increase the albumin levels (10,11). This is related to changes in behavior which is the most influential factor on adherence to the diet of hemodialysis patients (12).

The contents of the SMS were sent to subjects only to remind and ensure subjects to eat catfish *abon* and record their food and drink intake from sources other than *abon* regularly every day. The SMS does not contain material related to the hemodialysis diet so as not to cause bias related to nutritional counseling provided by the hospital nutritionist. After 21 days post-intervention, the subject's albumin level was measured again.

The dependent variable of this study was albumin levels, while the independent variable was the use of catfish as an animal protein source. Albumin level was on a nominal scale with two categories, i.e. hypoalbuminemia when the albumin levels were <3.50 g/dL and normal when the albumin levels were ≥3.50 g/dL. In this study, there are external variables that are thought to affect albumin levels, namely patient intake. Patient's intake was defined as the overall energy, protein, fat, and carbohydrate intake of hemodialysis patients obtained by the food record method, then processed using the Nutrisurvey program, compared to the patient's needs calculated individually, and presented. The calculation of its needs using the formula according to Pernefri, namely energy needs of 35 kcal/kg BW/day, protein requirements of 1.2 g/kg BB/day, fat 20% of total energy needs, and carbohydrates are the remaining total energy requirements.

The data obtained were then analyzed univariately to determine the frequency distribution of each variable and bivariate analysis using the Fisher's Exact test. This research has obtained Ethical Clearance from the Health Research Ethics Commission, Faculty of Health Science, Universitas Respati Yogyakarta No: 330.4/FIKES/PL/II/2017 dated 15 February 2017.

RESULTS

Hemodialysis patients routinely come 2 times a week with a fixed schedule, namely Monday–Thursday, Tuesday–Friday, and Wednesday–Saturday. The hemodialysis patient schedule is divided into 3 shifts, namely the morning shift (07.00–11.00 WIB), afternoon (11.00–15.00 WIB), and evening (16.00–20.00 WIB).

Subjects in this study were patients with a diagnosis of chronic kidney disease (CKD) with hemodialysis routinely 2 times a week. The number of subjects there were 34 people taken by purposive sampling technique from 3 shifts (morning, afternoon, evening) on Monday–Thursday, Tuesday–Friday, Wednesday–Saturday. Subject characteristics in this study include age, gender, education, and complication of the disease, which are shown in Table 2.

Table 2. Frequency distribution of subject characteristics

Subject characteristics	Category	Frequency (n)	Percentage (%)
Age	17 – 25 years	1	2,9
	26 – 35 years	4	11,7
	36 – 45 years	11	32,3
	46 – 55 years	9	26,4
	56 – 65 years	7	20,5
	≥ 65 years	2	5,8
Total		34	100
Gender	Man	15	44,1
	Woman	19	55,8
Total		34	100
Education	Primary school	13	38,2
	Junior high school	5	14,7
	Senior high school	14	41,1
	University	2	5,8
Total		34	100
Complication of disease	With complications	19	55,8
	No complications	15	44,1
Total		34	100

Based on Table 2, the majority of subjects belonging to the adult age category (11 people or 32.3%). Based on gender, there were more female subjects (19 people; 55.8%) than male subjects (15 people; 44.1%). Based on the level of education, the majority of subjects had a high school / vocational education (14 people or 41.1%) and at least were university graduates, namely 2 people or 5.8%. Based on complications, there were more subjects with disease complications (19 people; 55.8%) than subjects without disease complications (15 people; 44.1%). Complications of the subject's diseases include hypertension, gastrointestinal disorders (gastritis), gout, and lupus.

In this study, subjects were given nutritional support for catfish *abon* for 21 days, under the need of subjects that was 0.36 g/kg BB/day. The body weight used to calculate the needs of catfish *abon* is

the post hemodialysis body weight of each subject. Subjects consume all catfish *abon* given and controlled by sending a Short Message Service to the subject to remind the subject to consume the catfish *abon*. Even based on the results of the interview with the subjects, they liked the catfish *abon* and wanted to be given the catfish *abon* for a longer time after the study was completed. It means that the catfish *abon* product has good acceptance. Variable utilization of catfish as an animal protein source can be divided into pre-intervention and post-intervention.

Table 3 shows that in the pre-intervention condition, subjects with normal albumin levels were higher (73.53%) than subjects who experienced hypoalbumin (26.47%). Similarly, in the post-intervention condition, subjects with normal albumin levels were more (88.24%) than subjects who experienced hypoalbumin (11.76%). Table 3 shows that the number of subjects with normal albumin levels increased in the post-intervention condition compared to the pre-intervention.

Table 3. Frequency distribution of subject based on albumin levels

Data retrieval time	Category	Frequency (n)	Percentage (%)
Pre-intervention	Hypoalbumin	9	26.47
	Normal	25	73.53
Total		34	100
Post-intervention	Hypoalbumin	4	11.76
	Normal	30	88.24
Total		34	100

Total nutrient intake and non-catfish *abon* nutrient intake during the intervention in hemodialysis patients

The results of this study consider external variables that are thought to affect the results of the study, namely the patient's nutritional intake. The patient's nutritional intake is divided into the total nutrient intake during the intervention and the patient's nutritional intake from sources other than catfish *abon*.

The total nutrient intake during the intervention in this study included intake from catfish *abon* plus intake from non-catfish *abon*. Nutrient intake was obtained from the average intake of subjects for 21 days obtained by the Food Record method, processed using the Nutrisurvey program, then compared with the patient's needs calculated per individual. The intake is categorized to be low if the intake <80% needs, good if the intake 80–110% needs, and high if >110% needs.

Table 4. Frequency distribution of subject based on total nutrient intake during the intervention

Variable	Category	Frequency (n)	Percentage (%)
Energy intake	Low	13	38.2
	Good	18	52.9
	High	3	8.8
Total		34	100
Protein intake	Low	18	52.9
	Good	14	41.1
	High	2	5.8
Total		34	100
Fat intake	Low	21	61.7
	Good	11	32.3
	High	2	5.8
Total		34	100
Carbohydrate intake	Low	12	35.2
	Good	18	52.9
	High	4	17.6
Total		34	100

Table 4 shows that the majority of subjects consumed energy in the good category, as many as 18 people (52.9%). Most subjects consume protein in the less category, as many as 18 people (52.9%). Most of the subjects consume fat with fewer categories, namely as many as 21 people (61.7%). Most subjects consume carbohydrates in the good category, namely as many as 18 people (52.9%).

The intake of non-*abon* nutrients during the intervention was the intake of subjects sourced from food and beverages other than *abon* catfish. The intake of non-*abon* nutrients was obtained from the average intake of non-*abon* subjects for 21 days obtained by the Food Record method, processed using the Nutrisurvey program, then compared with the patient's needs calculated per individual. The intake of non-*abon* nutrients is categorized to be less if the intake <80% needs, both if the intake 80–110% needs, and more if >110% needs.

Table 5. Frequency distribution of subject based on nutrition intake from non-catfish *abon* source during the intervention

Variable	Category	Frequency (n)	Percentage (%)
Energy intake	Low	17	50
	Good	15	44.1
	High	2	5.8
Total		34	100
Protein intake	Low	28	82.3
	Good	5	14.7
	High	1	2.9
Total		34	100
Fat intake	Low	25	73.5
	Good	7	20.5
	High	2	5.8
Total		34	100
Carbohydrate intake	Low	12	35.2
	Good	18	52.9
	High	4	11.7
Total		34	100

Table 5 based on the intake of energy nutrients shows that the majority of subjects consume energy from non-catfish *abon* sources with fewer categories, as many as 17 people (50%). Based on the intake of protein nutrients from non-catfish *abon* sources the majority of subjects consumed protein in the less category, as many as 28 people (82.3%). Based on the intake of non-catfish *abon* fat nutrients, the majority of subjects consumed less fat, namely 25 people (73.5%). Based on the intake of non-*abon* carbohydrate nutrients, the majority of subjects who consumed carbohydrates in the good category, namely as many as 18 people (52.9%).

Effectiveness of catfish *abon* as nutritional support to albumin levels of hemodialysis patients

Fisher's Exact test results in Table 6 show that the use of catfish as an effective animal protein source has a significant effect on improving albumin levels of hemodialysis patients with p-value=0.048 (p-value<0.05).

Table 6. Bivariate analysis result of the effectiveness of catfish *abon* as nutritional support to albumin levels of hemodialysis patients using Fisher's Exact test

Category of albumin levels		Post-intervention		Total	*p-value*
		Hypoalbumin	Normal		
Pre-intervention	Hypoalbumin	3 (33.30%)	6 (66.70%)	9 (100.00%)	0.048
	Normal	1 (4.00%)	24 (96.00%)	25 (100.00%)	
Total		4 (11.80%)	30 (88.20%)	34 (100.00%)	

DISCUSSIONS

This study was dominated by adult subjects. This study was consistent with the general description of CKD patients undergoing hemodialysis in Indonesia, as reported by the Indonesian Renal Registry, which in 2011 found 89% of CKD patients undergoing hemodialysis aged 35–70 years with the most age groups of 45–54 years which are 27%.

Age is one of the factors that can affect an individual's health status. At the age of 40–70 years, glomerular filtration rate will progressively decrease to 50% from normal, i a decrease in the ability of the kidney tubules to reabsorb and concentrate urine, decrease the ability to empty the bladder thereby increasing the risk of infection and obstruction, and decreasing fluid intake which is a risk factor for kidney damage (13). Gender and age affect the incidence of glomerulonephritis which is one of the risk factors for chronic kidney failure (14).

The results of this study, more female subjects than male subjects. These results are similar to previous studies that the sex of patients with chronic kidney failure who performed hemodialysis were more women, as many as 33 people (52.4%), compared to 30 men (47.6%) (15).

Most of the subjects in this study had high school/vocational education and at least a university education graduate. In patients who have higher education will have broader knowledge also allows patients to be able to control themselves in overcoming the problems faced, have high self-confidence, experienced, and have precise estimates of how to handle events and easily understand what is happening recommended by health workers, will be able to reduce anxiety so that it can help the individual in making decisions (16). Knowledge of cognitive is a very important domain for the formation of action, behavior based on knowledge will be more lasting than those not based on knowledge (17).

Based on complications, the number of subjects with complications is higher than subjects without complications. Complications of the subject's diseases include hypertension, gastrointestinal disorders (gastritis), gout, and lupus.

Albumin levels of Hemodialysis Patients

In this study, there was an increase in the number of normal albumin subjects who experienced an increase in the condition of post-intervention compared to pre-intervention. Albumin is a very important serum in the body which is the main determinant of blood plasma osmotic pressure. The effect caused by decreased albumin will cause a shift in fluid from the intravascular space (18).

Hypoalbumin in patients with chronic kidney failure who have undergone hemodialysis can be caused by severe malnutrition caused by the inflammatory process in patients with chronic kidney failure. The presence of inflammation is associated with anorexia in dialysis patients. Chronic inflammation can also result in a rapid decrease in skeletal muscle protein and other tissues, reducing muscle and fat resulting in hypoalbumin (19). In addition to the inflammatory process of hemodialysis which removes protein and vitamins along with the dialysate which during hemodialysis runs it will lose 10–12 grams of amino acids, glucose will also be released via dialysate (20).

Albumin deficiency occurs when production is reduced and loss of albumin. More protein is lost than is made by a healthy liver (21). Decreased albumin levels in patients undergoing hemodialysis are influenced by malnutrition due to the inflammatory process that still occurs due to lack of hemodialysis time (22).

Total nutrient intake and non-catfish *abon* nutrient intake during the intervention in hemodialysis patients

Nutrient intake includes energy, protein, fat, and carbohydrates. The total nutrient intake during the intervention in this study included intake from catfish *abon* plus intake from non-catfish *abon*. The intake of non-catfish *abon* nutrients during the intervention was the intake of subjects sourced from food and beverages other than catfish *abon*.

Patients with chronic kidney failure with hemodialysis are recommended high protein intake to maintain nitrogen balance and replace amino acids lost during the hemodialysis process, ie 1–1.2 g/kg BW/day with 50% protein should be of high biological value because protein intake is very necessary remembering its function in the body. The effect of protein intake plays an important role in the prevention of nutrition of patients with chronic kidney failure because the symptoms of the uremic syndrome are caused by the accumulation of body protein catabolism. Therefore, the better the protein intake, the better it is in maintaining its nutritional status (23).

Cultivated fat intake 30% of calorie intake. On the one hand, fat intake is sufficient to meet calorie needs, while on the other hand fat also worsens kidney function and increases morbidity due to atherosclerosis. Consumption of complex carbohydrates such as rice, bread, sweet potatoes,

and cassava can spur the removal of excess uric acid in the blood (24).

Food intake of hemodialysis patients refers to the level of deterioration in kidney function. Food intake that must be limited in consumption is, protein intake is limited to 1–1.2 g/kg/day, potassium intake is limited to 40–70 meq/day, given the decreased function of potassium excretion and excretion of urea nitrogen by the kidneys. Meanwhile, the number of calories given is 30–35 kcal/kg BB/day (25).

Effectiveness of catfish *abon* as nutritional support to albumin levels of hemodialysis patients

In this study after being tested using Fisher's Exact, it was shown that providing nutritional support for catfish *abon* was effective in influencing the albumin levels of hemodialysis patients. Catfish *abon* has never been given as a form of dietary therapy in hemodialysis patients that is needed to replace the protein lost during hemodialysis and prevent catabolism of protein so that it can maintain serum creatinine levels within normal limits, overcome hypoalbuminemia, and enhance immunity.

Nutrient-based dietary guidelines emphasize animal-based protein foods for preventing and managing protein-energy wasting in hemodialysis patients (26). Fish is an animal food that contains proteins of good quality due to the complete content of essential amino acids. Fish can be extracted to obtain plasma protein (sarcoplasmic) containing albumin and another nutrient that has the potency to improve hypoalbuminemia condition (27).

There is a similar study with the basic ingredients of processed fish which are nutritional support in hemodialysis patients, the fish used is cork fish filtrate. Giving an extra intradialytic diet can maintain and increase serum albumin levels, as a result of protein intake during the hemodialysis process so that it can replace the amino acids lost especially during hemodialysis (25). The high intensity of use of cork fish (snakehead fish) to produce various products of fish albumin has put the natural stock of this fish species under great pressure. Therefore, it is necessary to find alternative sources of fish albumin other than the cork fish (28). Catfish is one of the sources of animal protein that has a high biological value, has a high protein content, and can be accessed by the public at a low price. The protein from catfish contains albumin, which can improve the albumin levels of hemodialysis patients if consumed with an adequate amount.

CONCLUSIONS

The use of catfish as an effective animal protein source has a significant effect on improving albumin levels in hemodialysis patients. Based on these conclusions, it is recommended to the hospital conduct continuous monitoring of the intake of hemodialysis patients, especially animal protein sources, to maintain the nutritional status of hemodialysis patients. Further research needs to be done on nutritional support for hemodialysis patients, with the use of local food in Indonesia.

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REFERENCES :

1. Ghorbani A, Hayati F, Karandish M, Sabzali S. The prevalence of malnutrition in hemodialysis patients. J Renal Inj Prev 2020;9(2):1–6.
2. Ministry of the Health Republic of Indonesia. Hasil utama riskesdas 2018. Jakarta: Ministry of the Health Republic of Indonesia; 2018.
3. Rivai AT. Status albumin serum pasien penyakit ginjal kronik yang menjalani hemodialisis di rumah sakit cipto mangunkusumo pada bulan februari 2009 dan hubungannya dengan lama hemodialisis [Artikel Penelitian]. Jakarta: Universitas Indonesia; 2009.
4. Wi JW, Kim N. Assessment of malnutrition of dialysis patients and comparison of nutritional parameters of CAPD and hemodialysis patients. Biomed Sci Letters 2017;23(3):185–193.
5. Qureshi AR, Alvestrand A, Divino-Filho JC, Gutierrez A, Heimburger O, Lindholm B, Bergstrom J. Inflammation, malnutrition, and cardiac disease as predictors of mortality in hemodialysis patients. JASN 2002;13:S28–S36.
6. Suhardjono. Buku ajar ilmu penyakit dalam jilid 2 edisi 3. Jakarta: Balai Penerbit FKUI; 2001.
7. Sridhar NR, Josyula S. Hypoalbuminemia in hemodialyzed end-stage renal disease patients: risk factors and relationships - a 2-year single-center study. BMC Nephrol 2013;14(242):1–9.
8. Kim Y, Molnar MZ, Rattanasompattikul M, Hatamizadeh P, Benner D, Kopple JD, Kovesdy CP, Kalantar-Zadeh K. Relative contributions of inflammation and inadequate protein intake to hypoalbuminemia in patients on maintenance

hemodialysis. Int Urol Nephrol 2013; 45:215–227.
9. Widiany FL, Afriani Y. Pemantauan gizi dengan SMS reminder efektif meningkatkan kepatuhan diet pasien hemodialisis. Jurnal Gizi Klinik Indonesia 2019;15(3):89–94.
10. Widiany FL, Afriani Y. Pemberian SMS reminder efektif memperbaiki status gizi antropometri pasien hemodialisis. Ilmu Gizi Indonesia 2017;01(01):49–53.
11. Widiany FL, Afriani Y, Rohman A. The effectiveness of nutritional monitoring by short message service reminder to albumin level of hemodialysis patients. J Nutr Sci & Diet 2018;4(3):Summer.
12. Widiany FL. Faktor-faktor yang mempengaruhi kepatuhan diet pasien hemodialisis. Jurnal Gizi Klinik Indonesia 2017;14(2):72–79.
13. Lajuck KS, Moeis ES, Wongkar MCP. Status gizi pada pasien penyakit ginjal kronik stadium 5 yang menjalani hemodialisis adekuat dan tidak adekuat. Jurnal e-Clinic 2016;4(2).
14. Price SA, Wilson LA. Patofisiologi konsep klinis proses-proses penyakit edisi 6 volume 2. Jakarta: EGC; 2006.
15. Damayanti P, Apen, Lisa A. Karakteristik penderita gagal ginjal kronis yang dilakukan hemodialisis; 2014.
16. Kamaluddin R, Rahayu E. Analisis faktor-faktor yang mempengaruhi kepatuhan asupan cairan pada pasien gagal ginjal kronik dengan hemodialisis di RSUD Prof. Dr. Margono Soekarjo Purwokerto. Jurnal Keperawatan Soedirman (The Soedirman Journal of Nursing) [series online] 2009;4(1). Available from: URL: http://jks.fikes.unsoed.ac.id/index.php/jks/article/view/218.
17. Notoatmojo S. Metodologi penelitian kesehatan. Jakarta: Rineka Cipta; 2002.
18. Sacher, Ronald A. Tinjauan klinik hasil pemeriksaan laboratorium. Jakarta: EGC; 2004.
19. Mardiana N. Nutrisi pada penderita dialisis. Prosiding dalam Meeting and Symposium [series online] 2008. Available from: URL: http://www.b11nk.wordpress.com
20. Yuwono A. Kualitas hidup menurut spitzer pada penderita gagal ginjal terminal yang menjalani hemodialisis di RSU DR. Kariadi Semarang. Artikel [series online] 2000. Available from: URL: http://www.eprint.undip.ac.id
21. Sibuea H, Panggabean M, Gultom. Ilmu penyakit dalam. Jakarta: PT Rineka Cipta; 2005.
22. Arinta, Rihantoro T. Hubungan lama menderita gagal ginjal kronik dengan kadar albumin pada pasien yang menjalani hemodialisis. Jurnal Keperawatan 2015;11(1).
23. Almatsier S. Penuntun diet. Jakarta: PT Gramedia Utama; 2005.
24. Sustrani L, Syamsir A, Iwan H. Asam urat, informasi lengkap untuk penderita dan keluarganya edisi 6. Jakarta: Gramedia; 2004.
25. Sulistyowati E, Hadi H, Susetyowati, Atma G. Pemberian diet ekstra formula komersial dan diet ekstra filtrat ikan gabus intradialisis serta pengaruhnya terhadap kadar serum albumin dan kreatinin pasien dengan hemodialisis di RSUD Dr. Saiful Anwar Malang. Jurnal Gizi Klinik Indonesia 2008;5(2).
26. St-Jules DE, Goldfarb DS, Popp CJ, Pompeii ML, Liebman SE. Managing protein-energy wasting in hemodialysis patients: A comparison of animal- and plant-based protein foods. Seminars in Dialysis 2018:32(1).
27. Mustafa A, Widodo MA, Kristianto Y. Albumin and zinc content of snakehead fish (Channa striata) extract and its role in health. IEESE International Journal of Science and Technology (IJSTE) 2012;1(2):1–8.
28. Fatma N, Nurilmala M, Metusalach, Nurfaidah, Astuti N. Low economic benefits of fish as source of fish albumin besides cork fish. Advances in Economics, Business and Management Research 2019;141:56–59.

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Effects of mixture powder of black rice (Oryza sativa L indica), red beans (Phaseolus vulgaris L), and moringa leaves (Moringa oleifera L) on blood glucose concentration in hyperglycemic Rats

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ABSTRACT

Background: Diabetes mellitus, increased blood glucose or hyperglycemia, is associated with increased oxidative stress and cardiovascular diseases. This condition will further cause carbohydrate and fat metabolism change, resulting in the decreased antioxidant defense system. Black rice, red beans, and moringa leaves contain oleic acid, butyric amino acid, antioxidants, phytic acid, and arginine, which can improve insulin sensitivity, and blood glucose homeostasis.
Objective: This study aimed to analyze the effect of betamelor (black rice, red beans, and moringa leaves) on blood glucose in rats
Materials and Methods: The design of this research was experimental research with pre-and post-control group design. A total of 20 Sprague Dawley female rats were divided into four groups, namely standard feed (PS), 80% feed of betamelor (PB8), 50% feed of betamelor (PB5), and 20% feed of betamelor (PB2). Betamelor intervention was given as much as 5% of weight for 28 days. Fasting Blood Glucose (FBG) levels were measured using the GOD-PAP method. Blood glucose data were analyzed by Analysis of Variance (ANOVA) at a 95% confidence level and using Duncan's test.
Results: There were differences in FBG between groups after the intervention of betamelor. The results showed that after 28 days of intervention, betamelor decreased the serum glucose concentration from 122.69 mg / dL to 97.70 mg / dL (20.37%) in the PB8 group and from 123.91 mg / dL to 113.28 mg / dL (8.58%) in the PB5 group, but the standard diet (PS) increased by 5.73%. This result can be applied to reduce blood glucose levels in obese and patients with metabolic syndrome.
Conclusions: There was a significant effect of giving a mixture of black rice, red beans, and Moringa leaves on fasting blood sugar in rats.

Keywords: Black rice; Red beans; Moringa leaves; Fasting blood glucose

BACKGROUND

The changes in lifestyle and diet of carbohydrate-based foods into high-fat content foods leads to the rise of degenerative diseases such as coronary heart disease, hypertension, and diabetes mellitus. An increase in blood glucose or hyperglycemia is a sign of a metabolic disorder known as diabetes mellitus and is associated with the rise of oxidative stress and complications in the vascular system. It will further cause the alteration in carbohydrate and fat metabolism resulting in an impairment of the oxidation defense system. In which this condition stimulates the increased formation of Reactive Oxygen Species (ROS) induce β-pancreatic cell dysfunction.1

One of the antioxidant defense systems is Superoxide Dismutase (SOD), the enzyme involved in the earliest and most potent detoxification in cells.2 The antioxidant enzyme itself is an essential enzyme which is able to eliminate radicals, then may protect cells against toxic of aerobic metabolism byproducts.3 The use of oral medicine of diabetes mellitus accompanied by the use of natural ingredients already become common practice yet, and around 1050 anti-Diabetes Mellitus plants have been studied.4 Some plants commonly used as a source of functional foods and already developed to help in controlling blood sugar are black rice, red beans, and moringa leaves. Ingredients contain oleic acid, amino butyric acid, antioxidants, pitic acid and arginine which are proven to reduce the rate of oxidative stress, thereby increasing antioxidants in the body.

Following Walter and Marchesan (2011), phenolic compounds are concentrated higher in the black rice pericarp.5 Bioactive compounds work as antioxidants in rice, including phenolic, flavonoid,

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anthocyanin, proanthocyanidin, tocopherol, tocotrienol, γ- oryzanol and phytate.6 Aleuron and endospermic in black rice could produce the high-intensity anthocyanin result a deep purple to dark color.7 However, black rice contains less protein and fiber, and this lack can be corrected by substitution of red beans and Moringa leaves. Kidney beans contain fat (15.80%), dietary fiber (3.60%), and carbohydrates (49.00%).8 The soluble fiber in red beans may reduce blood glucose.9 These beans also possess a high antioxidant capacity. The antioxidant capacity of EC50 red bean skin extract reaches 294.78 mg/ml.10 Al-Malki et al. (2015) found that antioxidant and antidiabetic activity in Moringa leaf extract showed potential as an antidiabetic agent in streptozotocin induced mice.17 Tantipaiboonwong et al. (2017), who have reported the antihyperglycemic and anti-hyperlipidemic effects of red rice extract and black rice extract to streptozotocin induced mice. Kidney beans contain arginine about 600 mg/100g.25 Arginine acts as an antidiabetic by regenerating pancreatic β cells to improve the stimulation of insulin secretion.26 Moringa leaves contain 6.7% protein, 1.7% fat and 14.3% carbohydrates.

Rat is one of rodents often used as models in studying blood sugar levels. This is because rats have a significant similarity in hematology and genome with humans.11 Studies related to black rice, red beans and Moringa leaves, which associated with the potential and influence on health, are still rarely performed. It is critical to assess whether there is an impact of giving black rice, red beans, and moringa leaves on blood sugar levels in rats.

This study aims to determine the effect of feeding developed from black rice flour, red beans, and Moringa leaves on rat blood glucose. Precisely, the objectives of this study, i.e., 1) determine differences in body weight 2) determine the average consumption of rat feeding during the intervention period; 3) analyze the effect of treatment on reducing feed consumption; 4) investigate the impact of treatment on rat blood glucose.

MATERIALS AND METHODS

This was an experimental study using a complete random design (CRD) with a pre-post control group. This study was conducted in March-May 2020, involving several laboratories. Making the black rice, red beans, and moringa leaves flours were carried out in the Food Processing Laboratory of Kupang's Health Polytechnic of Health Ministry's. The feed production was carried out at the Animal Feed Industry Laboratory, Kupang Agricultural Polytechnic. The intervention towards the experimental animal and blood collection was executed at the Biosciences Laboratory of the University of Nusa Cendana Kupang, while blood analysis conducted at the Health Laboratory of East Nusa Tenggara. The proximate analysis was carried out at the Saraswanti Indo Genetech Laboratory in Bogor, West Java.

The number of experimental animals used in this study was 20 rats, which were calculated according to the Federer formula (1997): (n-1) (t-1) ≥15 (n is the number of samples needed and t is the number of treatments). All those 20 rodents were divided into four groups, namely five rats of the standart feed (PS), five rats intervened by 80% mixture powder (PB8), five rats intervened by 50% mixture powder (PB5), and five rats intervened by 20% mixture powder (PB2). The animals used were Sprague Dawley female rats met the inclusion criteria, such as normal and healthy, aged 5-6 months, the weight of 150-200 g, open eyes, reddish-white skin, agile, and have soft, clean, dense, smooth, not fall, and shiny-haired. The exclusion criteria were that the rat died during the intervention, behavior changes (did not want to eat, drink and limp), weight loss of > 5%. Equipment used for rat care consists of a cage filled with husks equipped with food containers and drinking bottles, iron cage enclosures, digital scales for weighing rats and leftovers, and a set of cage cleaning tools. This study had received ethical approval from the Animal Ethics Commission, Faculty of Veterinary Medicine, University of Nusa Cendana No.168 / UN15.8.1 / PP / 2020.

The first stage of the research was the feed formulations from black rice, red beans, and moringa leaves. The composition of ingredients was following the diet of people with diabetes mellitus. Producing mixture powder began with making black rice flour, red beans flour, and moringa leaves powder, then sieved with 80 mesh size. Feed material in the form of bravo 512 standard solid feed was mixed until homogeneous. After that, a little water added until the feed mixture was half wet, then formed to obtain pellet feed. The wet pellet then dried in an oven (T 40ºC), then put into a plastic bag and kept in a refrigerator until the intervention time to experimental rats.

During the adaptation period, intended to ensure the experimental animals were in a healthy condition before the intervention, which was carried out in individual cages, given standard feed, and ad libitum drinking water for about ten days. Bravo 512 standard feed contains 12% water, 19.5-21.5% crude protein, min 5% crude fat, max 5% crude fiber, max 7% ash, 0.9 - 1.1% calcium, 0.6 – 0.9% phosphorus, and energy

about 3125kcal/Kg. Intake and residual feed were weighed and recorded every day, and body weighing was done once a week. A high glucose feed of 2cc/200g of body weight/day was given through sonde for two weeks to create hyperglycemic rats. The treatment was performed for 28 days, i.e., subjected a standard feed, 80% mixture powder, 50% mixture powder, and 20% mixture powder. Furthermore, those rodents fasted for 10 - 12 hours, then two cc of blood samples per each were taken through the eye veins (ocular arteries) using a microhematocrit capillary tubes after anesthetized with ketamine 10 mg/kg rat. Blood serum glucose analysis was executed according to the GOD-PAP method. The data of blood glucose were analyzed by Analysis of Variance (ANOVA) at a 95% confidence level. If it shows the significance response, then Duncan's Multiple Range Test was applied. Data processing and analysis were performed using Microsoft Excel and SPSS for Windows version 22.0.

RESULTS AND DISCUSSION

Proximate Composition

The results of the proximate analysis of black rice, red beans, and moringa leaves can be seen in Table 1. It shows that the ingredients used complement nutritional value each other mainly on the parameters of carbohydrate, protein, fat and dietary fibre.

Table 1. Proximate Composition of Mixture Powder

Parameters	PB Mixture 80%	PB Mixture 50%	PB Mixture 20%
Protein (g)	6.3 ± 0.91	10.1 ± 1.26	13.87 ± 2.03
Total fats (g)	0.8 ± 0.17	1.54 ± 0.49	2.28 ± 0.94
Carbohydrate (g)	25.3 ± 3.21	36.5 ± 2.64	47.9 ± 5.16
Fibre (g)	18.6 ± 2.33	13.8 ± 1.75	9.1 ± 1.02
Energy (kcal)	133.5 ± 18.01	200.2 ± 20.01	267.6 ± 37.22

Body Weight

Body weighing was done weekly to determine the amount of standard feed, high glucose feed (weeks 1 and 2), and mixture powder feed (weeks 3 to 6). The effect of feeding treatment on the body weight of rats can be seen in table 2. At the end of acclimatization, the average body weight of the rats did not have a significant difference between treatment groups. During the high-fructose diet for 7 days, the body weight of the rats increased significantly compared to the acclimatization period, but not significantly between treatments. During the mixture powder intervention period for 21 days, the body weight of the rats experienced a significant increase compared to the high fructose diet and there were also significant differences between the treatment groups. It was probably caused by the influence of blood drawn through the eye veins (ocular arteries), which results in pain and low appetite, recognized from the decrease in feed intake. In the following days, there was a slow improvement in body weight in line with an increase in feed intake.

Table 2. Body weight of rats fed a high-fat diet and treated with mixture powder for 10 weeks

Group	Acclimatization mean ± SD (g)	HGD mean ± SD (g)	Intervention of mixture powder mean ± SD (g)	P[2] (Acclimatization)	P[1] HGD1-Intervention
PS	190.82 ± 8.81	202.33 ± 5.79	227.62 ± 4.95	0.048	0.000
PB8	192.27 ± 6.47	201.48 ± 7.40	210.13 ± 3.19		
PB5	190.38 ± 6.72	202.18 ± 4.51	219.68 ± 3.57		
PB2	189.91 ± 7.63	203.76 ± 6.24	224.27 ± 5.51		
P[1]	0.821	0.294	0.008		

HGD : High Glucose Diet, [1]One way Anova test, [2] Paired t-test

Table 3. Concentration (mg/dL) of fasting blood glucose of female Sprague dawley rats that were administered graded doses of mixture powder

Group	Acclimatization mean ± SD (mg/dl)	HGD mean ± SD (mg/dl)	P[2]	△Increase (mg/dl) (%)	P[1]
PS	78.41 ± 2.60	124.10 ± 3.71	0.000	45.69 ± 1.51 (58.27%)	0.071
PB8	80.12 ± 1.53	122.69 ± 2.06	0.000	42.57 ± 2.07 (53.13%)	
PB5	79.64 ± 1.29	123.91 ± 3.49	0.000	44.27 ± 1.92 (55.58%)	
PB2	78.29 ± 2.31	124.53 ± 3.84	0.000	46.24 ± 1.89 (59.06%)	
P[1]	0.369	0.493			

HGD : High Glucose Diet, [1] One way Anova test, [2] Paired t-test

Table 4. Concentration (mg/dL) of fasting blood glucose of female Sprague dawley rats that were administered graded doses of betamelor.

Group	Acclimatization mean ± SD (mg/dl)	HGD mean ± SD (mg/dl)	P[2]	△Increase (mg/dl) (%)	P[1]
PS	124,10 ± 3,71	131,21 ± 2,46	0,004	7,11 ± 2,18 (5,73%)	0,007
PB8	122,69 ± 2,06	97,70 ± 1,92	0,000	-24,99 ± 1,84 (-20,37%)	
PB5	123,91 ± 3,49	113,28 ± 1,17	0,000	-10,63 ± 2,35 (-8,58%)	
PB2	124,53 ± 3,84	126,96 ± 2,38	0,042	2,43 ± 1,72 (1,95%)	
P[1]	0,493	0,493			

HGD: High Glucose Diet, [1] One-way Anova test, [2] Paired t-test

Fasting Blood Glucose Level Before and After Observation

Blood glucose levels of the rats at the end of the aclimatization stage and after HGD feeding for each group can be seen in Table 3. Significantly increased blood sugar levels occurred in all groups after intervened by HGD with a significant value (p> 0.05) based on the analysis of one-way test Anova. The average value of fasting blood glucose levels in the intervention of mixture powder 80% (PB8) and mixture powder 50% (PB5) experienced a significant decrease (p <0.05). While the response by standard feed as control and mixture powder 20% (PB2) experienced an increase in blood glucose levels. There were differences in rat blood glucose levels between groups after the administration of mixture powder based on the *Kruskal Wallis test*. The PB8 treatment group experienced the most substantial decline in blood glucose levels which was -24.99 ± 1.84 (-20.37%) mg/dl (Table 4). Concentration (mg/dL) of fasting blood glucose of female Sprague dawley rats that were pre and post administered graded doses of *mixture powder*.

Body weight in mice increased significantly for all groups from the acclimatization period to the high fructose diet phase and the high fructose diet phase to the betamelor intervention phase.Weight loss in the experimental animal was significantly increased for all groups on the acclimatization, high glucose diet, and mixture powder intervention. There was no significant difference in body weight among the groups in either phase of acclimatization or a diet high in glucose, but there were substantial differences in the intervention period. The bodyweight improvement was due to the consumption of standard and high glucose feed before the intervention. The differences in the different treatments during the intervention cause the alterations that have a significant effect on the amount of energy intake so that excess consumption will be stored as fat

reserves and increase bodyweight. In the acclimatization phase, the standard feed intake of each group was not significantly different. In the phase of the provision of a high glucose diet, there were significant differences between the treatment groups and the standard feed, the feeding intake of the control group had the highest compared to other groups.

Significant differences in feed intake occurred between groups. The control group had a higher feed intake than the treatment group. The consumption of the control group tended to remain since acclimatization. The treatment group of 50% and 20% mixture powder increased but not significant, and the 80% mixture powder group experienced a decrease in feed intake. It was due to the administration of mixture powder in the treatment groups that contains a high fibre composition, lead to full quickly compared to the standard feed group. Reducing intake in the intervention phase compared to the conditioning phase of treatment groups was due to the high fibre effect of diet caused by the rise of the hormone leptin, so the appetite decreased in the intervention phase.12 In the treatment groups, the standard feed intake was lower than in the conditioning period because it was in the adaptation stage. During the intervention phase, the usual feed intake of treatment groups was lower than the other groups because the stomach was quickly filled due to high fibre feeding.

Blood glucose levels were elevated in the test animals as the result of conditioning the provision of a high glucose diet after a period of adaptation completed with values > 120 mg/dl. It can be interpreted as metabolic syndrome, according to Crescenzo (2014) in Italia, the metabolic syndrome can be realized by high glucose feeding to increase blood glucose levels.13 Excess fructose intake in the liver will be metabolized into fat. Fructose may lead to a failure of insulin signal to reduce the synthesis of glycogen and increase gluconeogenesis, resulting in blood glucose elevation.13

Furthermore, this study revealed that the average fasting blood glucose values in the PB8 and PB5 groups experienced a significant slope after mixture powder intervention ($p < 0.05$). The results of this study were in accordance with Mbikay's literature review study (2012), the administration of moringa leaves for diabetes mellitus causes a potentially reducing blood sugar levels.14 Compounds that was an essential key allegedly in the antihyperglycemic activity in extracts of moringa (M. oleifera) is oleic acid. Oleic acid, or commonly called omega-9, is one of the best types of fat. Oleic acid also is known as one of monounsaturated oil (MUFA) because the body can synthesize from the nutritional compounds consumed, so it also including in non-essential fatty acids.14

In the research conducted by Aly et al. (2016) using GC-MS analysis, identified eight fatty acids contained in Moringa seed oil including palmitic acid, stearic acid, arachidonic acid, behenic acid, palmitoleic acid, linoleic acid, linolenic acid, and oleic acid.15 Besides, there were more than 50 phytochemical contents possessed in Moringa seed oil. According to Busari et al. (2015), the hypoglycemic activity of Moringa seed oil is caused by the presence of fatty acids in the oil.16 Monounsaturated fatty acids have a tendency to improve the function of beta-cell secretors, reduce the disruption of beta-cell activity and proliferation. Al-Malki et al. (2015) found that antioxidant and antidiabetic activity in Moringa leaf extract showed potential as an antidiabetic agent in streptozotocin-induced mice.17

Blood glucose levels in this study also found a decrease might be caused by the presence of black rice extract in the mixture flour. It is in line with the study by Tantipaiboonwong et al. (2017), who have reported the antihyperglycemic and anti-hyperlipidemic effects of red rice extract and black rice extract to streptozotocin-induced mice. The results of the study revealed that consumption of black rice extracts of 50 mg/kg body weight or red rice extract of 100 mg/kg body weight significantly reduced blood sugar levels after eight weeks.18 Observation by Chaiyasut et al. (2017) showed that germinated black rice extract could increase the aminobutyric acid content, total antioxidant capacity, and levels of antioxidant enzymes in diabetic rats and showed antidiabetic activity both before and after administration of streptozotocin in rats as animal models.19

A study conducted by Chung, et al. (2016) also found that pigmented rice has inhibitory enzymes responsible for diabetes. The results showed that those pigments contained phenol and had a significant enzyme inhibitory activity. Bioactive compounds that cause pigmentation in rice are anthocyanin and proanthocyanidin.20 Kasim et al. (2005) revealed that black rice has the highest antioxidant activity compared to the red and white rice. Naturally, black rice contains pigments with high antioxidant activity (anthocyanin).21 Hosoda, et al (2018) reported that black rice and brown rice contain anthocyanin and proanthocyanidin, which are potentially used as antioxidants sources other than as a starch source in ruminants.22 Bioactive compounds in pigmented rice

may reduce the oxidative stress, prevent cancer, cardiovascular, complications and potential diabetes, and others.5,22

In this study, mixture powder as a mixture of flours in an intervention feed in which one constituent is red beans, so the slope of fasting blood sugar values in an animal model may be related to the red beans. Kidney beans (*Vigna angularis*) are good sources of fiber, every 100 grams of dried red beans provides about 4 grams of fiber consisting of both soluble and insoluble fiber.23 Soluble fiber significantly lowering blood sugar, due to the soluble fiber is able to substantially reduce the glycemic response 23,24. This beans like other legumes, contain several components of inhibitor substances such as phytic acid, tannin, trypsin inhibitors, and oligosaccharides. The diet treatment using this legume may reduce glucose absorption by 48.43%. As for the soy diet, it can reduce absorption by 45.84% .25 Water-soluble fiber can form a viscous solution; the higher viscosity in the intestine leads the slower absorption of glucose by the small intestine. The viscous soluble fiber can reduce postprandial blood sugar and insulin levels. Based on the results of several studies, three different doses of red beans might lower blood glucose levels in male Wistar rats that given glucose load.24

Kidney beans contain arginine about 600 mg/100g.25 Arginine acts as an antidiabetic by regenerating pancreatic β cells to improve the stimulation of insulin secretion.26 Arginine as an amino acid of hormonal terminal GLP-1 (Glucagon-Like-Peptide-1) served as proinsulin gene expression and insulin synthesis stimulator. After insulin secretion occurs, glucose levels in the circulation immediately decrease; thus, the effect of GLP-1 will disappear by itself.27 Kidney beans; a staple food recommended for diabetic patients contains high protein and fiber, and low carbohydrates. Starch granules from these legumes have a unique structure and hydrolysis of starch in the small intestine relatively slow, thus postprandial glycemic response will be delayed.27 Yao's (2014) finding that the mechanism of red beans in hyperglycemia related to the effect of protein and phenolic compounds in it. Besides, this experiment also proved that extruded red bean protein and polysaccharides play a role in α-glucosidase inhibition.28 The subjects used were mice with *Sparague Dawley* strains, could not study the mechanism of diabetes mellitus. in further research, humans can use as samples. Further research is necessary to test the complete phytochemical contents of mixture powder of black rice, red beans, moringa leaves and to analyze its effect on other anti-inflammatory and antioxidant parameters in diabetes mellitus to deepen our understanding on the role and mechanisms of mixture powder of black rice, red beans, moringa leaves in diabetes mellitus

CONCLUSIONS

According to this study, the mixture of black rice, red beans, and moringa leaves as mixture flour reduce the fasting blood glucose reduction in rats significantly. Further research is needed to examine the complete phytochemical content of mixed black rice, kidney bean, moringa leaf powder and analyze their effect on anti-inflammatory and other antioxidant parameters in diabetes mellitus to deepen understanding of the role and mechanism of mixed powder black rice, kidney beans, moringa leaves in diabetes mellitus.

REFERENCES

1. Retnaningsih C, Darmono, Widianarko B, Muis SF. Peningkatan aktivitas antioksidan superoksida diamutase pada tikus hiperglikemi dengan asupan tempe koro benguk (Mucuna pruriens L).Agritech. 2013: 33(2):154–161.
2. Ighodaro OM, Akinloye OA. First line defence antioxidants-superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPX): Their fundamental role in the entire antioxidant defence grid. Alexandria Med J. 2017;54(4):287–293
3. Mansuroglu B, Derman S, Yaba A, Kizilbey K. Protective effect of chemically modified SOD on lipid peroxidation and antioxidant status in diabetic rats. Int J Biol Macromol. 2014;72:79–87
4. Subramoniam, A. Anti-Diabetes Mellitus Plants. Boca Raton: CRC Press; 2016
5. Walter M, Marchesan E.Phenolic Compounds and Antioxidant Activity of Rice. *Brazilian A Bio Tech*. 2011; Vol. 54 (2): 371–377.
6. Goufo P, Trindade H. Rice Antioxidants: Phenolic Acids, Flavonoids, Anthocyanins, Proanthocyanidins, Tocopherols, Tocotrienols, γ-Oryzanol, and Phytic Acid. *F Sci Nutr*. 2013; Vol. 2 (2): 75–104.
7. Wanti S, Andriani M.A.M, Parnanto N.H.R. Pengaruh Berbagai Jenis Beras terhadap Aktivitas Antioksidan pada Angkak oleh *Monascus purpureus. Biofarmasi. 2015;*Vol. 13 (1): 1–5
8. Audu SS, Aremu MO. Effect of process- ing on chemical composition of red kidney bean

(*Phaseolus vulgalris* L.) flour. PJN. 2011;10(11):1069-1075.
9. Messina. Nutrition and health benefit of dried bean. Am J Clin Nutr. 2014; 100 (suppl): 437S-442S.
10. Fidriannyi I, Nuraini P, Marlia SW. Antioxidant activities, total flavonoid, phenolic, carotenoid of various shells extracts from four species of legumes. Asian J Pharm Clin Res. 2014; 7(4):42-46.
11. Dieman VV, Trindale EN, Trindale MRM. Experimental model to induce obesity in rats. Acta Cir Bras. 2006;21(6):425-429.
12. Izadi, V., Saraf-Bank, S., & Azadbakht, L. Dietary intakes and leptin concentrations. *ARYA atherosclerosis.*2014;*10*(5), 266–272.
13. Crescenzo R, Bianco F, Coppola P, Mazzoli A, Tussellino M, Carotenuto R, et al. Fructose supplementation worsens the deleterious effects of short-term high-fat feeding on hepatic steatosis and lipid metabolism in adult rats. Exp Physiol. 2014;99(9):1203-13
14. Mbikay, Majambu. Therapeutic Potential of *Moringa oleifera* Leaves in Chronic Hyperglycemia and Dyslipidemia: A Review. Front Pharmacol. 2012; 3:24
15. Al-Malki, A.L, dan El Rabey, H.A. The Antidiabetic Effect of Low Doses of Moringa oleifera lam. Seeds on Streptozotocin Induced Diabetes and Diabetic Nephropathy in Male Rats. Biomed research International. 2015: 1-13.
16. Tantipaiboonwonga P, Pinthaa K, Chaiwangyena W, Chewonarinb T, Pangjitc K, Chumphukama O,et.al. Anti hyperglycaemic and Anti hyperlipidaemic Effects of Black and Red Rice in Streptozotocin Induced Diabetic Rats. *ScienceAsia.* 2017;Vol. 43: 281– 288.
17. Chaiyasut C, Sivamaruthi B.S, Pengkumsri N, Keapai W, Kesika P, Saelee M, Tojing P, et.al. Germinated Thai Black Rice Extract Protects Experimental Diabetic Rats from Oxidative Stress and Other Diabetes-Related Consequences. *Pharmaceuticals*. 2017; Vol. 10 (3): 2–16.
18. Chung S.I, Kang M.Y, Lee S.C. Inhibitory Effect of Germinated Pigmented Rice on Key Metabolic Enzymes Associated with Diabetes and Hyperglycemia. *In JF Nutr Scie.* 2016; Vol. 5 (4): 32–40.
19. Kasim E, Astuti S, Nurhidayat N. Karakterisasi Pigmen dan Kadar Lovastatin Beberapa Isolat *Monascus purpureus*. *Biodiversitas*. 2005;6 (4): 245–247.
20. Hosoda K, Sasahara H, Matsushita K, Tamura Y, Miyaji M, Matsuyama H. Anthocyanin and proanthocyanidin contents, antioxidant activity, and *in situ* degradability of black and red rice grains. Asian-Australas J An Scie. 2018; Vol. 31 (8): 1213–1220.
21. Rimbawan.Indeks glikemik pangan: cara mudah memilih pangan yang menyehatkan. Jakarta: Penebar Swadaya;2004.
22. Audu SS, and MO Aremu. Effect of Processing on Chemical Composition of Red Kidney Bean (Phaseolus vulgaris L .) Flour. Pakistan J Nutr. 2011;10 (11):1069–75.
23. Monti LD, MCC asiraghi, E Setola, E Galluccio, MA Pagani, L Quaglia, et al,. L-Arginin Enriched Biscuit Improve Endothelial Function and Glucose Metabolism: A Pilot Study in Healthy Subjects and A Cross-over Study in Subjects with Impaired Glucose Tolerance and Metabolic Syndrome. Met Clin Ex. 2013;62: 255-264.
24. Yao Y, Cheng X, Ren G. Alpha-Glucosidase inhibitory activity of protein-rich extracts from extruded adzuki bean in diabetic KK-Ay mice. Food Func. 2014;5(5):966–971.
25. Liu, Y., Wang, Q., Li, S., Yue, Y., Ma, Y., & Ren, G. Convenient food made of extruded adzuki bean attenuates inflammation and improves glycemic control in patients with type 2 diabetes: a randomized controlled trial. Ther clin risk man.2018;14, 871–884
26. Yao Y, Cheng X, Wang L, Wang S, Ren G. A determination of potential alpha-glucosidase inhibitors from Azuki Beans (Vigna angularis) Int J Molecular Sci. 2011;12(10):6445–6451.

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The effectiveness of Islamic comic media in increasing the attitude of healthy breakfast among students

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ABSTRACT

Background: As much as 73.4% of children aged ≥ 5 years in Indonesia have breakfast with the low nutritional quality of food consumption, as evidenced by the prevalence of 95.5% of children's less consumption of vegetables and fruits. Breakfast habits affect fiber and micronutrient levels. Communication and nutritional information can increase accuracy in breakfast behavior from childhood. Effective and efficient media are needed. So far, the media for children has prioritized illustrations without paying attention to the cultivation of moral values.
Objectives: This study aimed to determine the effect of Islamic comic media on improving healthy breakfast attitudes among students.
Materials and Methods: The research design was a quasi-experimental type. Subjects were taken by a simple random sampling method. Ninety-six students were divided into three groups. Group A was given nutrition education using Islamic comic media about health breakfast (n = 32). Group B was given nutrition education using Islamic comic strip media about health (n = 32). Group C was a control group (n = 32). The study used two types of nutrition promotion media: comics and comic strips, which are stories about healthy eating in Islam. The time for nutrition education intervention through comics and comic strips is 2x ± 35 minutes. The pretest was ± 25 minutes with ± 10 minutes explaining the instructions for filling out the questionnaire. The posttest was ± 25 minutes with a discussion for ± 10 minutes—nutritional attitudes related to health breakfast by answering a validated questionnaire. We used the Lickert scale to categorized nutritional attitudes. Statistical test was done by Wilcoxon and Mann Whitney test.
Results: The distribution of nutrition attitudes of respondents increased after the provision of Islamic comics, both comics and comic strips about breakfast. The nutritional attitudes increased significantly in the Islamic comic media group (p = 0.000) and the Islamic comic strip media group (p = 0.000), from 78.75 to 92.96 and 78.61 to 92.88, respectively.
Conclusions: There was a relationship between counseling and Islamic comic media regarding healthy breakfast towards the nutritional attitude of elementary school-age students.

Keywords: Nutritional attitude; Islamic comics; Breakfast

BACKGROUND

In developing countries, the prevalence of obesity in children is known to have increased by 60% since 1980.[1] The habit of not having breakfast causes the child to experience hypoglycemia, dizziness, trembling, fatigue and difficulty concentrating.[2] Breakfast also affects appetite control, satiety, and energy expenditure, and weight management.[3] Long-term impact, resulting in nutritional status, decreasing health and stamina of children, and hindering the improvement of the nation's human resources In Indonesia. It is known that 69.6% of Indonesian children have not had breakfast according to the recommended balanced nutrition guidelines and 73.4% have breakfast with the nutritional quality of food consumption is low. The consumption of various foods greatly affects nutritional status.[4]

Breakfast has a positive influence on cognitive and academic outcomes.[5] Breakfast is important for elementary school age were a period of high brain productivity in absorbing various lessons.[6] The habit of not having breakfast also affects fiber and micronutrient levels. Children who regularly eat a healthy breakfast with vegetables and fruit have higher levels of fiber and micronutrients.[7] Low consumption of fruits and vegetables can result from a lack of motivation.[8]

Habits and appropriateness of children's breakfast can be formed with nutritional education.[9] Nutritional education with a medium can accelerate understanding, resulting in motivation according to the message received.[10] Therefore, an educational process needs to be supported by appropriate media.[11] Elementary school-aged students are more interested

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in media and picture books or cartoons than media and books that contain only writing.[12] Cartoon media has been shown to increase children's fruit or vegetable intake.[13]

Media is very important in the educational process. This is because a medium can increase motivation and enthusiasm for learning psychologically.[14] Someone who has good nutritional knowledge, then dominantly has a good nutritional attitude too. Student learning intentions can be shaped by using learning comics. Meanwhile, in comic media, besides being preferred by students, it is also proven to be effective in increasing students' knowledge. The effectiveness of comics as a learning medium was proven positive with a significant increase in the level of knowledge in the experimental group.[15] A significant increase also occurred in the attitude of the experimental group. Students' interest in learning media in the form of audiovisuals illustrates good acceptance.[16] Educational media for children nowadays put forward good illustrations without paying attention to the cultivation of moral values.[17] The learning media should not only prioritize changes in knowledge but also contain moral values.[18] Cultivation of noble morals through comic book characters also plays an important role, because children like to imitate and look after. [19] This study aims to analyze the influence of Islamic comic media on healthy breakfast on elementary school students' attitudes.

MATERIALS AND METHODS

This study used a quasi-experimental design. Location of research at Public Madrasah Ibtidaiyah (MIN) 1 Teluk Lingga, East Kutai district, East Kalimantan, Indonesia. This study was carried out in December 2018 - February 2019, which was adjusted to the active schedule of students of MIN 1 Teluk Lingga, Sangata City, East Kutai district, East Kalimantan, Indonesia. The school criteria taken were Islamic elementary (Madrasah Ibtidaiyah / SDIT), had A accreditation by the official madrasah accreditation organizing body, and had never received counseling or nutritional education. MIN 1 Teluk Lingga is a school that meets the above criteria with the largest student population.

The population in this study were all students of MIN 1 Teluk Lingga, as many as 647 students. Consists of 21 classes with various levels. The inclusion criteria for the selection of research subjects were subjects who had never received nutritional education about healthy breakfast, were willing to take part in the research until it was finished, and were aged 10-12 years. Sampling calculation using the Slovin formula to obtain a sample size of 96 students, then formed 3 groups, namely, group A given nutritional education with Islamic comic media about healthy breakfast (n = 32), group B was given nutritional education using Islamic comic strip media about healthy breakfast and group C was a control group (n = 32).

The variables studied included elementary school students' attitudes regarding healthy breakfast before and after the comic education intervention using a questionnaire that had been tested for validation. The questionnaire totaled 16 attitude statements. Lickert used to determine the level of a person's attitude by providing "agree" and "disagree" statements.[20] Attitude data was categorized as good if the answer is 76% -100% correct, categorized sufficient if the answer is correct 51% -76%, and categorized as less if the answer is correct 25-50%.[21]

This study used two types of nutrition promotion media, in the form of comics and comic strips, which are stories about healthy breakfast according to Islamic eating habits. The main characters used are elementary school-age children with additional characters in the form of mothers and teachers at school. Characters are designed with noble morals with a good speech to provide good role models for respondents.

The types of images used in both media were 2D animation based on Graphical User Interfaces (GUIs). The tools used in the media are the Samsung Galaxy Tab A.8.0 Tablet and the rubber nib Stylus Pen with applications such as Medibang Pro and Adobe Photoshop. The comic was later printed on A4 ivory paper.

This research was conducted in five stages which were carried out for five consecutive days. The first stage was pretest data collection on the three groups carried out on the first day. The second stage was carried out with nutrition education intervention in group 1 using comic strip media and in group 2 using comic strip media which was conducted on the second day. The third stage was posttest data collection from the three groups with the same questions in the three groups which were carried out on the third day. The fourth stage was the repetition of the second stage, while the fifth stage was the repetition of the third stage.

The time for nutrition education intervention through comics and comic strips was 2x ± 35 minutes. The pretest was carried out for ± 25 minutes with ± 10 minutes of explaining the instructions for filling out the questionnaire. Posttest was carried out for ± 25 minutes with a discussion for ± 10 minutes.

The analysis used the Wilcoxon test with Mann-Whitney post hoc because the data was not normal. The Wilcoxon test aims to determine whether there is a significant difference between the attitude values before and after the provision of comic strip media, comic strips, and without giving media to each treatment group. Furthermore, the Mann-Whitney post hoc test was conducted to determine which intervention or treatment was the most effective. This study was endorsed by the health research ethics committee of Dr. Moewardi Hospital, Faculty of Medicine, Sebelas Maret University with number 395 / III / HREC / 2019

Figure 1. Comic cover

The nutrition promotion media created were Islamic comics in the form of comics and comic strips, with the title "Sarapan Sehat, yuk!" (Let's have healthy breakfast!), The first edition. The place of manufacture was in Sangata on December 30th, 2018. The total page consists of 9 pages for comics. The language used is Indonesian. The story setting is Home and School. The main characters of this comic are Ali and Aisyah, while the additional characters are Budi, Umi, and Bu Guru. Summary of story: Every morning, Umi always told Ali and Aisyah to have breakfast, but they didn't know why they had to have breakfast every morning. One day at school, their friend Budi fainted during a ceremony because he wasn't having breakfast. After being explained by the teacher, Ali and Aisyah knew the benefits of breakfast. Since then, Ali and Aisyah always had breakfast happily and also helped Umi cook breakfast. Comic bonus: the 'Kata Nabi' comic about the prohibition of denouncing food and blowing hot food. Reference comics: Comic "Ayo Sarapan Sehat" (Come have a healthy breakfast), by PERGIZI, and comic "Sarapan, yuk!" (Let's have breakfast) by the POMPI club (BPOM).

Figure 2. Islamic Comic Characters and Storylines

The main characters used are elementary school-age children with additional characters in the form of mothers and teachers at school. Parents are the first and foremost educators, whose roles are role models and provide the best care and education for children. Characters are designed with noble morals with a good speech to provide good role models for respondents.

On the first day, the pretest questions were distributed to the research group at different lesson hours. The pretest time lasted for 35 minutes, the first 10 minutes, the researcher used to explain the aims and objectives of the study by filling out the questionnaire instructions. The time to answer the pretest questions was 25 minutes. The nutrition promotion media provision intervention was given the next day.

Nutrition promotion media were given in 2 days with 2 hours of subjects per day. The time used was 70 minutes. On the first day, students were asked to listen to several students who led the story (storytelling) in front of the class and matched the conversation with the comic that each student had held. This was done to add an audio function that makes it easier to remember during the presentation of the material. Because comics are printed media that do not have an audio function. Then, the researcher asked three students to retell the essence of the story they understood by bringing their respective comics.

The researcher asked each student to read their comics without being guided by anyone at the second meeting. This is to train the critical feeling that students have in understanding the implicit message in the story presented. The researcher gave 30 minutes to read the comic and asked the students to read it twice to strengthen students' understanding of the story content. At the end of the meeting, the researcher asked three students to give their opinion about the message conveyed by comics and provide the essence of the story, this time without bringing comic media.

RESULTS

Based on the data analysis carried out, the distribution of respondents age and gender characteristics
was described in the following table:

Table 1. Distribution of Respondents Age and Gender Characteristics

Variable	A%	B%	C%	Total%
Age				
10	65.7	34.3	53.1	51
11	34.3	65.7	46.9	49
Gender				
Male	50	50	43.7	48
Female	50	50	56.3	52

Note: A = The group that was given nutrition education using Islamic comic media about healthy breakfast (n = 32); B = The group that was given nutrition education using Islamic comic strip media about healthy breakfast (n = 32); C = Control group (n = 32).

Differences in Attitude Values Between Research Groups

The data was not normally distributed related to the results of the distribution on the normality test of the mean data difference between the pretest and posttest attitude values. Furthermore, the analysis was carried out with the Wilcoxon test with the Mann Whitney post hoc test, to determine which groups had differences. The results showed that between groups A and B the result was p = 0.000 (<0.05).

Table 2. Attitudes of Respondents Before and After Intervention

	Pengukuran	n	Mean	Median (Minimum-Maximum)	p Value
A	Before the Intervention	32	78.75	90.62 (50.00-100.00)	0.000
	After the Intervention	32	92.96	95.31 (76.56-100.00)	
B	Before the Intervention	32	78,61	88.81 (50.00-100.00)	0.000
	After the Intervention	32	92,88	95.31 (68.75-100.00)	
C	Before the Intervention	32	80,46	85.15 (48.43-98.43)	0.148
	After the Intervention	32	82,46	84.37 (50.00-100.00)	

DISCUSSION

Based on the results obtained, it can be seen that there were differences in attitude scores in groups A and B compared to group C which is the control group. This result was in line with previous research conducted by Widayanti et al. which states that there was a significant increase in attitude scores (<0.05) in elementary school students who were given nutrition promotion media in the form of comics.[22] This result was also supported by previous research conducted by Jefri and Kharis (2017) in which there was a significant increase in attitude levels (> 0.05) in elementary school students who were given promotional media in the form of comic strips.[6] In line with the research of Ridha et al. (2009) stated that comics have a strong influence on social information processing to create changes in attitudes.[23]

The main role of comics in instruction is their ability to create interest in students. The effectiveness of comics as a learning medium was proven positive with an increase in the level of knowledge that significantly shaped attitudes in the experimental group.[15] Students' interest in learning media in the form of audiovisuals illustrates good acceptance.[16] Comic media and comic strips have several main functions as follows: 1) Attention function, which attracts and directs students' attention to concentrate on the content of the lesson related to the visual meaning displayed; 2) The compensatory function, visual media that provides context for understanding the text, helps students who are weak in reading to organize information in the text and recall and understand the content of the lesson that is presented with text or presented verbally;[24] 3) Affective function, where visual images or symbols can evoke emotions and attitudes of students, for example, information relating to social or racial issues;[25] 4) Cognitive function, where visual symbols or images facilitate the achievement of goals to understand and remember the information contained in images.[26]

Based on the results of the mean difference between the pre and post-education attitude values in group A was 14.21 with a pretest value of 78.75 increasing to 92.96 and the mean difference between the pre and post-education attitude values in group B was 14.27 with a value of 78.61 increasing to 92.88. The results of the Mann Whitney test in groups A and B were p = 0.000. It can be concluded that there was a significant increase in attitudes after the provision of nutrition promotion media in the form of Islamic comic strips and comic strips in groups A and B. In group C, the difference in the mean value of attitude increased by 2.0 but based on the Wilcoxon test there was no increase in attitude in group C, which is the control group.

Changes in the attitude value of the experimental group that increased significantly also reflected an increase in the learning achievement of respondents. Motivation to learn is the point the respondent's result is obtained by each individual as a level of ability expressed in the form of cumulative values or numbers. The importance of increasing the respondent's nutritional attitude after giving the media is an indication that the moral values of the characters conveyed in Islamic comics have succeeded in giving a significant effect in increasing the respondent's attitude.[27] This is based on the nature of the respondents who are children of primary school age who like to imitate and nurture.[27] Factors that influence attitude formation include the amount of information a person has.[28] The more attractive the educational media used, the higher the increase in knowledge that shapes the respondent's attitude.[29]

CONCLUSIONS

The respondent's attitude increased after counseling by providing Islamic comic media about healthy breakfast. There was an effect of the provision of Islamic comic media and Islamic comic strips regarding healthy breakfast on increasing attitudes.

REFERENCES

1. Gupta N, Goel K, Shah P, Misra A. "Childhood obesity in developing countries: epidemiology, determinants, and prevention," *Endocr. Rev.*, vol. 33, no. 1, pp. 48–70, 2012, DOI: 10.1210/er.2010-0028.
2. Huang CJ, Hu HT, Fan YC, Liao YM, Tsai PS. "Associations of breakfast skipping with obesity and health-related quality of life: Evidence from a national survey in Taiwan," *Int. J. Obes.*, vol. 34, no. 4, pp. 720–725, 2010, doi: 10.1038/ijo.2009.285.
3. Gwin JA, Leidy HJ. "A review of the evidence surrounding the effects of breakfast consumption on mechanisms of weight management," *Adv. Nutr.*, vol. 9, no. 6, pp. 717–725, 2018, DOI: 10.1093/advances/nmy047.
4. Perdana F, Hardinsyah d. "Analisis Jenis, Jumlah, Dan Mutu Gizi Konsumsi Sarapan Anak Indonesia (Analysis of Type, Amount, and Nutritional Quality of Breakfast among Indonesian Children)," *J. Gizi Dan Pangan*,

vol. 8, no. 1, pp. 39–46, 2013, [Online]. Available: http://download.portalgaruda.org/article.php?article=127426&val=199&title=ANALISIS JENIS, JUMLAH, DAN MUTU GIZI KONSUMSI SARAPAN ANAK INDONESIA.

5. Yaslam Babaeer L, Wraith D. "A systematic review of the effect of habitual breakfast for A systematic review of the effect of habitual breakfast for adolescents aged 11-19 years on academic performance adolescents aged 11-19 years on academic performance," *J. Adolesc. Fam. Heal.*, vol. 9, no. 1, p. 7, 2018, [Online]. Available: https://scholar.utc.edu/jafh/vol9/iss1/7/.
6. Jefri M, Kharis W. "Pengaruh Pendidikan Kesehatan Menggunakan Media Komik Strip Terhadap Pengetahuan," *J. Penelit. Keperawatan*, vol. 3, no. 2, pp. 96–99, 2017.
7. Coulthard JD, Palla L, Pot GK. "Breakfast consumption and nutrient intakes in 4-18-year-olds: UK National Diet and Nutrition Survey Rolling Programme (2008-2012)," *Br. J. Nutr.*, vol. 118, no. 4, pp. 280–290, 2017, doi: 10.1017/S0007114517001714.
8. Albani V, Butler L, Traill W, Kennedy O. "Breakfast consumption and nutrient intakes in 4–18-year-olds: UK National Diet and Nutrition Survey Rolling Program," *Br. J. Nutr*, vol. 120, no. 2, pp. 398–408, 2017.
9. Damayanti AY, Setyorini IY, Fathimah F. "Gambaran Tingkat Pengetahuan Gizi Seimbang pada Santriwati Remaja Putri di Pondok Pesantren," *Darussalam Nutr. J.*, vol. 2, no. 2, p. 1, 2018, doi: 10.21111/dnj.v2i2.2437.
10. Cita DA. "Pengembangan Materi dan Daya Terima Pendidikan Gizi Melalui Media Visual Terhadap Pengetahuan Gizi Anak Usia Sekolah Dasar," 2014.
11. Kementrian Kesehatan RI. "Pedoman PGSKesehatan," *Pedoman Gizi Seimbang*, pp. 1–99, 2014.
12. Daryanto. *Manajeman Pemasaran : Sari Kuliah*. Bandung: Satu Nusa, 2013.
13. Kraak VI, Story, M. "Influence of food companies' brand mascots and entertainment companies' cartoon media characters on children's diet and health: A systematic review and research needs," *Obes. Rev.*, vol. 16, no. 2, pp. 107–126, 2015, DOI: 10.1111/obr.12237.
14. Rusman. *Belajar dan Pembelajaran Berbasis Komputer*, 1st ed. Bandung: Alfabeta, 2013.
15. Pritandhari M, Ratnawuri T. "Penerapan Komik Strip Sebagai Media Pembelajaran Mata Kuliah Manajemen Keuangan Mahasiswa Univeristas Muhammadiyah," *J. Pendidik*, vol. 3, no. 1, pp. 1–13, 2015.
16. Johan H, Pradana TD. "Pengaruh Penyuluhan Media Audio Visual Video Terhadap Perilaku Cuci Tangan Pakai Sabun Pada Siswa Kelas III Di SDN 027 Samarinda," *J. Husada Mahakam*, vol. 4, no. 6, pp. 352–356, 2018.
17. Yuliana L. *Urgensi Penanaman Nilai-Nilai Moral Pada Anak Usia Dini*. Bandung: Alfabeta, 2014.
18. Najuah. "Strategi Pembelajaran Berbasis Pendidikan Nilai Dalam Pembelajaran IPS Di Sekolah," in *Prosiding : Seminar Nasional Tahunan*, 2017.
19. Al-Ayouby MH. "Dampak Penggunaan Gadget Pada Anak Usia Dini (Studi di PAUD dan TK.Handayani Bandar Lampung)," *Skripsi*, vol. 1, no. 1, pp. 1–10, 2017, doi: 10.1037/0022-3514.51.6.1173.
20. Swarjana K. *Metode Penelitian Kesehatan*, 1st ed. Yogyakarta: ANDI, 2015.
21. Arikunto S. *Prosedur Penelitian Suatu Pendekatan Praktik*, Revisi 7. Jakarta: Arikunto, 2013.
22. Widajanti L, Suryawati C, Sugihantono A. "The Effect of Healthy and Nutritious Snack Food Comic to Increase Knowledge and Attitudes of Elementary School Children Ages, Indones.," *J. Public Heal.*, vol. 6, no. 1, pp. 22–27, 2009.
23. Ridha A, Dian P, Mayarestya NP. "Pengaruh media komik terhadap pengetahuan kesehatan mata pada anak," *J. Perikan (J. Fish. Sci.)*, vol. XII, no. 1, pp. 20–27, 2009.
24. Ajizah N, Suherman HA, Susilawati D. "Penggunaan Media Pembelajaran Untuk Meningkatkan Gerak Dasar Lari Sprint Pada Siswa Kelas V Sdn Sukajadi Kecamatan Situraja Kabupaten Sumedang," *Sp Vol 1*, vol. 1, no. 1, pp. 35–44, 2016.
25. Doron E. "Fostering Creativity in School-

Aged Children Trought Perspective Taking and Visual Media Based Short Term Intervention Program," *J. Think. Ski. Creat.*, vol. 23, no. 27, pp. 150–160, 2016, DOI: https://doi.org/10.1016/j.tsc.2016.12.003.

26. Dean J. "Sorted for Memes and Gifs: Visual Media and Everyday Digital Politics.," *Polit. Stud. Rev.*, vol. 17, no. 3, pp. 255–266, 2019, DOI: 10.1177/1478929918807483.
27. Naujah. "Strategi Pembelajaran Berbasis Pendidikan Nilai Dalam Pembelajaran IPS Di Sekolah," in *in Seminar Nasional Tahunan*, 2017, p. 1.
28. Azwar S. *Sikap Manusia : Teori dan Pengukurannya*, 2nd ed. Yogyakarta: Liberty, 2016.
29. Hartono NP, Wilujeng CS, Andarini S. "Nutritional Education of Healthy Snack Choice Between Lecture and Comic Method," Indonesia," *J. Hum. Nutr*, vol. 2, no. 2, pp. 76–84, 2015.

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Application of the general message of balanced nutrition during the pandemic coronavirus disease 2019 (COVID-19) in Mataram City

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ABSTRACT

Background: Coronavirus Disease 2019 (COVID-19) in 2020 became the limelight worldwide, including in Indonesia. Infection caused by this virus can be prevented by social distancing, physical distancing, and a healthy lifestyle. Maintaining nutritional intake by implementing balanced nutrition guidelines is very important to enhance the immune system.
Objectives: This study aimed to determine the description of the application of balanced nutrition guidelines during the COVID-19 pandemic in the community in Mataram City.
Materials and Methods: This research was a descriptive study with a survey approach, and the data taken were cross-sectional. This research was conducted in May-June 2020 in the community in the Mataram City taken by purposive sampling with inclusion criteria, namely women and men of productive age, domiciled in the city of Mataram, married status, and at least elementary school education. Respondents in this study were 460 respondents who were calculated using the Slovin formula. Data on the application of balanced nutrition guidelines were obtained using a questionnaire. Data from the results of this research were analyzed using descriptive methods.
Results: There are as many as >90% of the community has implemented a general message of balanced nutrition in addition to the fifth message. As many as 89.9% of respondents did not apply the fifth general message of balanced nutrition in limiting consumption of sweet, salty, and fatty foods. The majority of applications >50% of general messages of balanced nutrition were applied by respondents aged 21-29 years, female, high education and work status.
Conclusions: The application of the general message of balanced nutrition during the epidemic COVID-19 in Mataram City was implemented optimally, but this application was not carried out in the fifth message in limiting consumption of sweet, salty, and fatty foods.

Keywords: COVID-19; Application; General Message for Balanced Nutrition

BACKGROUND

In early 2020, various parts of the world including Indonesia were shocked by the pandemic caused by the Coronavirus Disease 2019 (COVID-19). Coronavirus disease 2019 (COVID-19) is a disease caused by the acute respiratory syndrome coronavirus (SARS-CoV-2). On March 11, 2020, the World Health Organization (WHO) declared the Coronavirus outbreak in 2019 to be a pandemic [1]. This is because this disease has rocked more than 200 countries in various parts of the world including Indonesia. In Indonesia, on June 18, 2020, there were 42.762 cases infected with this virus with a cure rate of 39,3% [2], while in West Nusa Tenggara Province there were 1.022 cases infected by this virus. This happened because the spread of Coronavirus Diseases 2019 was relatively fast and difficult to detect [3]. Various efforts have been recommended to cut the spread of the virus through social distancing and physical distancing so that all activities such as work, discussions, and meetings are normally carried out online [4].

The restrictions that occur during the pandemic have an impact on the decline in the economy, food availability, nutritional intake, and people's lifestyle. Efforts and strategies continue to be made by the government to stabilize this condition, either by providing direct cash assistance or providing food supplies [1]. Availability of foodstuffs, a healthy

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lifestyle, and fulfillment of balanced nutritional intake are now things that need to be prioritized and carried out optimally. However, as before known, the problem of unhealthy food consumption habits among Indonesians including West Nusa Tenggara Province before the COVID-19 pandemic is still quite high and has not been resolved optimally. This showed in the results of the 2018 Basic Health Research in West Nusa Tenggara Province which noted that as many as 7.1% of the people did not consume fruits and vegetables, 31.2% had less activity, 53.1% did not wash their hands properly, > 30% consuming sweet and fatty foods and 10% with the habit of consuming salty foods [5]. The application of general guidelines for well-balanced nutrition during the COVID-19 pandemic plays an important role in the health of the body. Fulfilling the need for micronutrients, micro (micronutrients), food safety, physical activity and adequate water consumption in the body can help the hormones forming, leukocytes, immunoglobulins, and lymphocytes in the immune system improving the function of the heart, lungs and maintaining balance in the body during the COVID-19 pandemic [6].

According to the Indonesian Ministry of Health (2020), the infection caused by the Coronavirus in 2019 has an impact on increasing body temperature, so that compliance of balanced and healthy nutritional intake tends to increase to support the immune system. The adoption of unhealthy eating habits and lifestyles by not washing hands properly, unbalanced food consumption, and inadequate physical activity during the pandemic will have an impact on decreasing the body's defense system and increasing viral infections. Therefore, understanding and implementing a healthy lifestyle that refers to the guidelines for well-balanced nutrition is the main asset for obtaining a healthy, strong body defense and avoiding various diseases caused by viruses during the COVID-19 pandemic [7].

Previous research has shown that the majority of toddler mothers in Babakan District do not apply the four pillars of balanced nutrition obtained by using the interview method [8]. However, further, in this study, research will be carried out about the description of the application of general guidance for balanced nutrition in more detail on the application of 10 general messages of balanced nutrition, especially during the COVID-19 pandemic. The application of the general message of balanced nutrition is obtained by using a questionnaire in the form of scoring on each general message of balanced nutrition. The goal of this research is that people notice and apply a healthy lifestyle based on balanced nutrition guidelines in their daily life. In addition, it is also hoped that health workers and the government will continue to give education and socialization about general guidance for balanced nutrition to improve the health status of the community. This study aims to describe the application of balanced nutrition guidelines during the COVID-19 pandemic in the people of Mataram City, West Nusa Tenggara.

MATERIALS AND METHODS

This research was a descriptive study using a survey approach method. This research was conducted from May to June 2020 in Mataram City, West Nusa Tenggara. The populations in this study were all people in West Nusa Tenggara Province with the number of respondents calculated using the Slovin formula (α = 5%) as many as 460 respondents. The sampling technique in this study was carried out by using purposive sampling with inclusion criteria consisting of women and men of productive age, live in Mataram city, married and or have been married and at least primary school education. Data on the application of balanced nutrition general messages were collected using a questionnaire for the application of 10 general messages of balanced nutrition which were applied during the COVID-19 pandemic with the help of google form media. The questionnaire used in this study refers to a balanced nutrition behavior questionnaire with a Cronbach α reliability value of 0.725 and has been valid [9]. The general message of balanced nutrition contained in this study consists of (1) being grateful and enjoying the diversity of food, (2) consuming lots of vegetables at least 3 times a day and consuming fruit at least 2 times a day, (3) get used to consuming side dishes that contain high protein, (4) get used to consuming a variety of staple foods, (5) limit consumption of sweet, salty and fatty foods, (6) get used to breakfast, (7) get used to drinking enough and safe water in the amount of 8 glasses or ± 2 liters per day, (8) getting used to reading labels on food packaging, (9) washing hands with soap and running water and, (10) doing adequate physical activity and maintaining a normal body

weight with a duration of activity and exercise as much as 30 minutes per day. The data obtained from the results of this study were analyzed descriptively on each general message in the guidelines for balanced nutrition.

RESULTS

Respondent Characteristics

Data on the characteristics of respondents in this study consisted of age, gender, education, and occupation. The results of this research show that most of the respondents in this research were in the age range 21-29 years and were female. In terms of education, most of the respondents are at the middle level with a working status. The data on the characteristics of respondents in this research can be seen in Table 1.

Application of The General Message of Balanced Nutrition

Data collection on the application of balanced nutrition general messages in this research refers to 10 general messages of balanced nutrition in the balanced nutrition guidelines regulated by the Ministry of Health of the Republic of Indonesia in 2014. The application data for balanced nutrition general messages are categorized in the category of applied and not applied. The applied category is given to respondents who apply according to the instructions on every general message of balanced nutrition, while in the not applied category it is given to respondents who do not apply according to the instructions in every general message of balanced nutrition. The results of application 10 general messages on balanced nutrition in the guidelines for balanced nutrition can be seen in Table 2.

Table 1. Respondent Characteristics

Category	n	%
Age		
≤ 20 years	75	16,3
21 - 29 years	238	51,7
30 – 49 years	142	30,9
> 49 years	5	1,1
Sex		
Male	155	25,0
Female	345	75,0
Education		
Primary	7	1,6
Middle	80	17,4
Higher	373	81,0
Work Status		
Work	316	68,7
Not Work	144	31,3

Based on the results of the analysis in Table 2 it is known that during the COVID-19 pandemic 449 (97.6%) respondents had applied the general message of balanced nutrition in being grateful and enjoying a variety of foods. The application of getting into the habit of consuming vegetables and fruit also shows that most of the 454 (98.7%) respondents have adopted the habit of consuming vegetables and fruit every day. This happened because most respondents said that during the COVID-19 pandemic respondents gained a lot of knowledge from various information sources that advocated a healthy lifestyle with enough consumption of fruits and vegetables to maintain endurance and avoid viruses and various diseases, besides that the habit of consuming fruits and vegetables has also become a habit that has been done before, but during this pandemic, the consumption of fruits and vegetables has increased.

The general message of balanced nutrition on the habit of consuming side dishes that contain high protein and the habit of consuming a variety of staple foods has mostly been applied as many as 456 (99.1%) and 454 (98.7%) respondents have applied this general message. This is because most respondents said that the availability of high protein and staple foods such as chicken, sea fish, rice, noodles, and bread in West

Nusa Tenggara Province is quite available and easy to find in traditional markets and other shopping centers so that access to these foods is easy to get. Respondents did not apply restrictions on consuming sweet, salty, and fatty foods during the COVID-19 pandemic. As many as 98.9% of respondents are accustomed to consuming sweet, salty, and fatty foods, so the general message of balanced nutrition in getting used to limiting consumption of sweet, salty, and fatty foods is not applied. Most of the reason respondents did not apply this message was that it had become a habit. Sweet foods that are often consumed include syrup, flavored drinks, jam, candy, and the use of sugar, salty foods can be salted fish and salted eggs, while fatty foods consumed are fried and fast food.

Table 2. Application of 10 General Messages on Balanced Nutrition in The Guidelines for Balanced Nutrition

General Messages on Balanced Nutrition	Application				Total	
	Applied		Not Applied			
	n	%	n	%	n	%
1. Being grateful and enjoying the diversity of food	449	97,6	11	2,4	460	100
2. Consuming vegetables and fruit	454	98,7	6	1,3	460	100
3. Consuming side dishes that contain high protein	456	99,1	4	0,9	460	100
4. Get used to the consumption of a variety of staple foods	454	98,7	6	1,3	460	100
5. Limit consumption of sweet, salty and fatty foods	5	1,0	455	98,9	460	100
6. Get used to breakfast	450	97,8	10	2,2	460	100
7. Get used to drinking enough and safe water	446	96,9	14	3,1	460	100
8. Get used to reading labels on food packaging	445	96,7	15	3,3	460	100
9. Get used to washing hands with soap and running water	458	99,6	2	0,4	460	100
10. Doing enough activity and maintaining a normal body weight	421	91,3	39	8,6	460	100

The habit of drinking enough and safe drinking water of ± 8 glasses (2 liters) during the COVID-19 pandemic has been implemented by 446 (96.9%) respondents in this study, this habit has also been practiced before the COVID-19 pandemic. In addition to the habit of drinking enough and safe water, breakfast habits have also been carried out during this pandemic as many as 450 (97.8%) of respondents have adopted the habit of having breakfast. This happens because most respondents said that the habit of eating breakfast is done because to maintain a healthy lifestyle and this breakfast habit accidentally occurs because all activities and work are done at home so that the opportunity to have breakfast can be done.

Judging from the application of the general message of balanced nutrition in the habit of reading labels on food packaging during the COVID-19 pandemic, it is known that 445 (96.7%) respondents applied the general message, but a small proportion did not apply it, while in the application it was used to wash their hands using soap, and running water during the COVID-19 pandemic, it is known that most of the 458 (99.6%) respondents applied this general message. In the application of general messages in getting used to doing adequate physical activity and maintaining normal body weight, there were 39 (8.6%) respondents who did not apply this general message during the pandemic. This is because many respondents have the reason that office work and online assignments and meetings make physical activity limited.

Data on the application of the general message of balanced nutrition were also analyzed descriptive based on the characteristics of the respondents consisting of age, gender, education, and occupation. The application of this balanced nutrition general message is categorized in the category of applying

message, apply 50% of messages, and didn't apply the message. The category of applying messages is given to respondents if they have applied >50% - 100% or 6-10 messages of balanced nutrition according to the advice, while in the category of applying 50% of the messages are given to respondents if they have applied 50% of messages or 5 of 10 general nutritional messages balanced according to recommendations and the category of not applying message is given to the respondent if they do not apply the 10 general messages of balanced nutrition according to the advice. The results of descriptive analysis of the application of the general message of balanced nutrition based on the characteristics of the respondent during the COVID-19 pandemic can be seen in Table 3.

Based on the results of the analysis in Table 3, it shows that the majority of respondents who applied the general message of balanced nutrition during the COVID-19 pandemic were in the age range of 21-29 years as many as 235 (98.7%) respondents and as many as 6 (8.0%) respondents in ≤ 20 years old who apply 50% of the general message of balanced nutrition. Judging by gender, as many as 341 (9.8%) female respondents applied the general message of fully balanced nutrition, and seen at the education level, most of the higher education levels implemented the general message of fully balanced nutrition as many 365 (97.8%). The application of the general message of balanced nutrition during the COVID-19 pandemic has also been fully implemented by most respondents with a working status of 140 (97.3%) of respondents.

DISCUSSION

The infection caused by the coronavirus disease 2019 (COVID-19) causes many life changes, including lifestyle, so that the nutritional balance before and after infection is very important to pay attention to [7]. The implementation of the general guidance for balanced nutrition established by the Ministry of Health of the Republic of Indonesia in 2014 is one way that can be done to improve body health. These guidelines aim to offer behavioral guidelines in consuming food and beverages and living a healthy life. The results of these studies indicate that > 90% of respondents have implemented the general message of balanced nutrition properly during the COVID-19 pandemic except for the fifth message by limiting sweet, salty, and fatty foods. In general, the application of general guidelines for well-balanced nutrition during the COVID-19 pandemic can offer benefits for the health of the body.

Table 3. Application of Balanced Nutrition General Message based on Respondent Characteristics

Characteristics	Application						Total	
	Applying Messages		Applying 50% of Messages		Didn't applying Message			
	n	%	n	%	n	%	n	%
Age								
≤ 20 years	69	92.0	6	8,0	0	0	75	100
21 - 29 years	235	98.7	3	1.3	0	0	238	100
30 – 49 years	139	98.6	3	1.4	0	0	141	100
> 49 years	5	100	0	0	0	0	5	100
Sex								
Male	107	93.0	8	7.0	0	0	115	100
Female	341	98.8	4	1.2	0	0	345	100
Education								
Primary	7	100	0	0	0	0	7	100
Middle	76	95.0	4	5.0	0	0	80	100
Higher	365	97.8	8	2.2	0	0	373	100
Job Status								
Work	308	97.5	8	2.5	0	0	316	100
Not Work	140	97.3	4	2.7	0	0	144	100

The habit of consuming a variety of foods, both staple foods, and fruits and vegetables, getting used to breakfast, and reading packaging labels on food during the COVID-19 pandemic, the need for macronutrients, micronutrients, and food safety in the body are met. In addition, the fulfillment of protein intake, physical activity, and adequate water consumption in the body can effectively help hormones forming, leukocytes, immunoglobulins, and lymphocytes which can support the immune system, improve the function of the heart, lungs and maintain balance in the body during the COVID-19 pandemic [10].

Prevention of deficiency and excess due to nutrition caused by bad food habits and lifestyle during a pandemic requires an understanding and application of good nutrition with the principle of balanced nutrition [11,12]. In general, actions in the application of food consumption and healthy living carried out by each person will pass through various stages including response, mechanism, and adaptation [13]. The difference that occurs in each action is consuming everyone's foods is strongly influenced by the presence of internal factors and external factors. Psychological conditions, availability of foodstuffs, and the area of residence are risk factors for changes in eating behavior for everyone [14]. This is in line with the application of the general message of balanced nutrition carried out during the COVID-19 pandemic in this study, the psychological state and the availability of food ingredients during this pandemic period have made people pay more attention to a healthy lifestyle that is carried out daily so that body health is maintained. In addition, the government has made an appeal and education to wash hands with soap and clean water as well as improve the clean and healthy lifestyle of the community. However, the application of limiting the consumption of sweet, salty, and fatty foods has not been implemented optimally with >90% of respondents in this study still consuming large amounts, so this is a special concern that needs to be done to maintain a healthier intake.

The limitation made on the consumption of sweet, salty, and fatty foods in the general message of balanced nutrition is an effort to support health. Excessive consumption of sweet, salty, and fatty foods will have an impact on the emergence of various degenerative diseases such as diabetes mellitus, heart disease, and hypertension as well as changes in nutritional status so that with these diseases the body's defenses will become weak in the face of the virus during the COVID-19 pandemic [15,16,17]. The results of this study show that 98.9% of respondents do not practice limiting their consumption of sweet, salty, and fatty foods. This is in line with the results of the 2018 Basic Health Research which showed that consumption of sweet, salty, and fatty foods respectively was 37.6%, 10.0%, and 32.4%, which are still quite high in West Nusa Tenggara [6]. The emergence of various types of food and beverages on some televisions, shopping centers, habits, preferences, and environmental factors makes it easy to access these foods, thus affecting food consumption. In addition to food restrictions, the application of the habit of washing hands using soap and running water during the COVID-19 pandemic is also highly recommended. Washing hands using soap and running water is one of the efforts that can be done in reducing the spread of the coronavirus, so this application is very important.

In general, food consumption behavior and habits that are implemented by a person are strongly influenced by various factors, including internal and external factors. Internal factors consisting of age, gender, education, and occupation can affect a person's healthy behavior. The age factor can affect the knowledge a person has [18]. With increasing age, it greatly affects the development, needs, and mindset that exists in him, so that this situation affects the habit of implementing the general message of balanced nutrition [19]. The results of these studies indicate that as many as 98.7% of respondents aged 21-29 and gender apply the general message of well-balanced nutrition. One of the perceptions and patterns of thought that exist in each person is greatly influenced by the increase in age. Everyone will play an active role in social life and prepare for the adjustment to old age that occurs at the age of 20 to 35 years. Throughout that age, everyone will tend to spend more time acquiring knowledge, so that intellectual abilities, problem-solving, and knowledge acquisition tend to increase and result in good behavior in everyday life [20].

The educational factor is one of the tools used to produce a change in a person, therefore through education, a person can find out everything that has not been known before. Education is a process carried out to gain knowledge, understanding, and how to behave according to your needs [21]. The results of this

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study show that as many as 365 (97.8%) respondents with a higher education level have implemented >50% of the general message of well-balanced nutrition. This research is in line with research on mothers with children in Manado who reported that mothers who are at the higher education level have knowledge of well-balanced nutrition and will continue to learn and apply this knowledge related to nutrition to their children and their families [22]. In addition, the results of research in South Kalimantan also show that providing education related to good nutrition to a person will improve healthy behavior [23].

An internal characteristic that factors into a person's behavior is work. The work that is owned by each person influences lifestyle and consumption behavior, including behavior in implementing general guidance for balanced nutrition [15]. The results of this study show that as many as 308 (97.5%) respondents with working status have implemented> 50% general messages of well-balanced nutrition. This research is in line with research at SMK Surabaya which reports that the work factor of parents is one of the factors that can influence food consumption behavior in the household so that the habit of providing and implementing food for each individual and family is very much determined by education, work and income [24].

CONCLUSIONS

Based on the results of this research it was concluded that >90% of the people in Mataram City applied the general message of balanced nutrition in the first to fourth messages and the sixth to tenth messages properly during the pandemic period. However, >90% of people in Mataram City do not apply the fifth general message of balanced nutrition, namely limiting the consumption of sweet, salty, and fatty foods. Judging by the characteristics, most of the people aged 21-29 years, female sex, middle education, and work applying >50% general nutrition messages during the COVID-19 pandemic. The application of healthy living habits based on general guidelines for balanced nutrition for both self and family during this pandemic is very necessary to be maintained to support a healthy body. The role of health workers and the government is very much needed to continue to offer education and information related to balanced nutrition to support and become a habit in having a healthy lifestyle.

REFERENCES

1. Djalante, R., Lassa, J., Setiamarga, D., Sudjatma, A., Indrawan, M., Haryanto, B., Warsilah, H. Review and Analysis of Current Responses to COVID-19 in Indonesia: Period of January to March 2020. Progress in Disaster Science Journal: 2020; 6 (100091): 1-9.
2. Kementerian Kesehatan Republik Indonesia. Situasi Covid-19, Kondisi 18 Juni 2020. Kementerian Kesehatan Republik Indonesia; 2020.
3. Sekertariat Daerah Pemerintah Provinsi Nusa Tenggara Barat. Press Release Covid-19. Nusa Tenggara Barat; 2020.
4. Lingam, L., & Suresh, S. R. COVID-19, Physical Distancing and Social Inequalities: are we all Really in This Together. The International Journal of Community. 2020; 2(2): 173-190.
5. Badan Penelitian dan Pengembangan Kesehatan Kementerian Kesehatan RI. Laporan Nasional Riskesdas 2018. Jakarta; 2018.
6. Wu, D., Lewis, E. D., Pae, M., & Meydani, S. Nutritional Modulation Of Immune Function: Analysis Of Evidence, Mechanisms, and Clinical Relevance. Frontiers in Immunology Journal. 2019; 9(3160): 1-19.
7. Kementerian Kesehatan Republik Indonesia. Panduan Gizi Seimbang Pada Masa Pandemi Covid-19. Jakarta; 2020.
8. Fitri, Z., & Salam, A. Penerapan Pedoman Gizi Seimbang Pada Ibu Balita di Kelurahan Babakan. Jurnal Gizi Prima. 2017; 2(2): 148-153.
9. Nuryani. Validitas dan Reabilitas Kuesioner Pengetahuan, Sikap dan Perilaku Gizi Seimbang Pada Remaja. Jurnal Gizi dan Kesehatan. 2019; 3(2): 37-46.
10. Kementerian Kesehatan Republik Indonesia. Pedoman Gizi Seimbang. Jakarta; 2014.
11. Pratami, T. J., Widajanti, L., & Aruben, R. Hubungan Penerapan Prinsip Pedoman Gizi Seimbang denga Status Gizi Mahasiswa S1 Departemen Ilmu Gizi Universitas Diponegoro. Jurnal Kesehatan Masyarakat. 2016; 4(4): 561-570.
12. Agustina, & Permatasari, P. Hubungan Pengetahuan dan Penerapan Pesan Gizi seimbang Remaja dalam Pencegahan Anemia Gizi Besi. Jurnal Ilmiah Kesehatan Masyarakat. 2019; 11(1): 1-9.

13. Perdani, Z. P., Roswita, H., & Nurhasanah. Hubungan Praktik Pemberian Makan dengan Status Gizi Anak usia 5 Tahun di Pos Gizi Desa Tegal Kunir Lor Mauk. Jurnal KFT. 2016; 1(2): 17-29.
14. Stankevich, A. Explaining the Consumer Decision-Making Process: Critical Literature Review. Journal of International Businees Research and Marketing. 2017; 2(6): 7-14
15. Adriaansz, P. N., Rottie, J., & Lolong, J. Hubungan Konsumsi Makanan dengan kejadian Hipertensi pada lansia di Puskesmas Ranomuut Kota Manado. Jurnal Keperawatan. 2016; 4(1): 1-9.
16. Anjangsari, K. N., & Isnawati, M. Hubungan Konsumsi Softdrink, lingkar pinggang dan aktivitas fisik dengan kadar glukosa darah puasa pada wanita dewasa. Jurnal Of Nutrition College. 2015; 4(2): 162-170.
17. Soraya, D., Sukandar, D., & Sinaga, T. Hubungan Pengetahuan gizi, tingkat kecukupan zat gizi dan aktivitas dengan status gizi pada guru SMP. Jurnal Gizi Indonesia. 2017; 6(1): 29-36.
18. Rahman, N., Dewi, N. U., & Armawaty, F. Faktor-faktor yang berhubungan dengan perilaku makan pada remaja SMA Negeri 1 Palu. Jurnal Preventif. 2016; 7(1): 1-64.
19. Yusra, V. D., Machmud, R., & Yenita. Faktor-faktor yang berhubungan dengan tingkat pengetahuan wanita usia subur tentang SADARI di Nagri Painan. Jurnal Kesehatan Andalas. 2016; 5(3): 697-704.
20. Silalahi, E., Syarifuddin, & Sudibyo, M. Faktor-faktor yang mempengaruhi terhadap pengetahuan tentang lingkungan pada siswa tingkat SMP/MTs dan SMA/MAN Adiwiyata di Kota Labuhanbatu. Jurnal Pendidikan Biologi. 2016; 5(3): 146-153.
21. Putri, R. M., & Lasri. Pekerjaan, Tingkat Pendidikan dan Pengetahuan Ibu Pra Sekolah tentang Gizi Seimbang. Jurnal Care. 2016; 4(3): 78-87.
22. Wowor, P., Engkeng, S., & Kalesaran, A. F. Faktor faktor yang berhubungan dengan perilaku konsumsi jajanan pada pelajar di Sekolah Dasar Negeri 16 dan Sekolah Dasar Negeri 120 Kota Manado. Jurnal Kesmas. 2018; 7(5): 17-23.
23. Irnani, H., & Sinaga, T. Pengaruh Pendidikan Gizi terhadap pengetahuan, praktik gizi seimbang dan status gizi pada anak sekolah dasar. Jurnal Gizi Indonesia. 2017; 6(1): 58-64.
24. B, V. N., & Wahini, M. Faktor- Faktor yang mempengaruhi pola konsumsi makan remaja (Kasus di sekolah Menenengah kejuruan Negeri 8 Surabaya). Jurnal Boga. 2014; 3(3): 47-50.

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Antibacterial potential of red dragon fruit peel yogurt (hylocereus spp.) against bacillus subtilis bacteria in hypercholesterolemic wistar rats

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ABSTRACT

Background: Fruit peel is a part of red dragon fruit that weighed 30-35% of the fruit weight and has not been used optimally. Red dragon fruit peel contains fiber, vitamin, flavonoid, tannin, alkaloids and has the potential as an antibacterial. Red dragon fruit peel can be processed into yogurt.
Objectives: This research examined the antibacterial potential of red dragon fruit peel yogurt against Bacillus subtilis in hypercholesterolemic Wistar rats.
Materials and Methods: Materials tested in this study were negative control, positive control, and caecum of hypercholesterolemic Wistar. This study used chloramphenicol as the positive control (K+) and DMSO 10% as the negative control (K-). The K1; K2; K3 were orally administered with 1.8 mL; 2.7 mL; 3.6 mL of red dragon fruit peel yogurt, respectively. Red dragon fruit peel yogurt was administered daily for 28 days. Caecum was collected and tested for antibacterial activity using disk diffusion (Kirby Bauer). The Bacillus subtilis was obtained from the Microbiology Laboratory of Center for Food and Nutrition Studies Universitas Gadjah Mada.
Results: The average inhibition zone in K-; K+; K1; K2; K3 were 0.00±0.00 mm; 11.5±1.41 mm; 11.5±0.96 mm; 10.13±0.66 mm; 10.38±1.12 mm, respectively. The experimental animal groups, which received 2.7 mL and 1.8 mL of red dragon fruit peel yogurts, showed a significant difference compared to the positive control group (p= 0.026 and p=0.021, respectively). When the dose was increased to 3.6 mL, it showed no statistical difference in results (p=1.000).
Conclusions: Red dragon fruit peel yogurt has an antibacterial potential against Bacillus subtilis.

Keywords: Antibacterial; Bacillus subtilis; Caecum; Red dragon fruit peel; Yogurt.

BACKGROUND

Antibacterial is a substance produced by microbes or other sources that can inhibit growth and even eradicate pathogenic bacteria that are harmful to humans. Bacterial infection is one of the health problems in Indonesia. Gastrointestinal disorders are complaints caused by bacterial infection.[1] Pharmacological therapy using antibiotics is often chosen to treat bacterial infection problems. Bacterial infection therapy using antibiotics can cause problems. Antibiotics are components produced by microbes that can kill or inhibit the growth of other microbes. Antibiotics are also defined as a natural molecular inhibitor.[2] The problem that arises from antibiotic therapy is the occurrence of resistance. This resistance event is triggered by the use of antibiotics that are not following the proper dosage, thus causing pathogenic microorganisms to become resistant and infection therapy to be ineffective. Bacillus subtilis can survive in various environmental conditions on land, water, and even in anaerobic conditions inside the digestive tract. These bacterial spores can even be blown by the wind to allow long-distance migration.[3] Bacillus subtilis can activate its host defense system so that the host becomes resistant to pathogens. Bacillus subtilis is a gram-positive bacterium commonly found in rotten bread.[4]

The discovery of new antibacterial compounds derived from natural ingredients can be an alternative solution to overcome the problem of antibiotic resistance. The sensitivity of each microorganism to a drug determines the lowest drug levels that can inhibit the growth of microorganisms in vitro. These antibacterial compounds can be obtained from plants. Plants contain compounds that have antibacterial potential with a new mechanism of action and have not yet experienced resistance. One of the plants that can function as an antibacterial is red dragon fruit.

Red dragon fruit is one type of dragon fruit species widely found and consumed in Indonesia. Dragon fruit is commonly known as pitaya, comes from the *Cactaceae* family [5] and is a tropical fruit.

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Pitaya has a CAM (photosynthetic crassulacean acid metabolism pathway) which allows efficient use of water.[6] Dragon fruit peel is a part of dragon fruit that weighed 30-35% of the fruit weight and has not been used optimally.[7] Red dragon fruit peel contains polyphenols and other active compounds. Active compounds other than polyphenols that can be found on red dragon fruit peel are alkaloids, terpenoids, flavonoids, saponins, tannins, carotenes, and phytoalbumins. Polyphenols such as flavonoids are more commonly found in the peel of red dragon fruit than in the flesh. Polyphenols as phenol components have many antioxidant activities. Betalain is a component associated with anthocyanins and is a reddish pigment found in red dragon fruit peel.

Processing of products from red dragon fruit peel which are considered as waste has already begun. To date, people know that red dragon fruit peel has many properties such as antioxidants, anti-hypercholesterolemia, and anti-diabetics. In research on antibacterial activity of n-hexane fraction from red dragon fruit peel against Staphylococcus aureus ATCC 25923, it was known that the inhibition zone diameter of the n-hexane fraction at concentrations of 20 and 40 mg/mL are 11.17±1.69 mm and 12.80±1.11 mm respectively.[8] Another research using ethanol extract of red dragon fruit peel (Hylocereus polyrhizus) on Staphylococcus aureus ATCC 25923 in vitro showed that the minimum inhibitory concentration was obtained at a concentration of 25% and the best inhibition zone was produced by an extract with a concentration of 100%.[9] Other research on red dragon fruit peels also stated that red dragon fruit peel extract can inhibit the growth of Salmonella pullorum with an average inhibition zone diameter at concentrations of 60 mg/mL, 40 mg/mL, and 20 mg/mL are 9.6 mm, 9.4 mm and 9.3 mm, respectively.[10] Research on the potential of red dragon fruit peel has not been done much, and it can be a great opportunity to explore red dragon fruit peel. Septiana's pre-clinical research shows that steeping dragon fruit peel can reduce plasma MDA levels in Sprague Dawley dyslipidemic rats. MDA is one of the parameters for free radical compounds. An increase in MDA value indicates an increase in oxidative stress in body cells.[11] Mardiana's research on red dragon fruit peel shows that red dragon fruit peel can be processed into yogurt and has the potential to be a functional food.[12] Yogurt can support human health and survival and improve dysbiosis due to aging.[13] Red dragon fruit added to yogurt with concentrations of 3%, 5%, and 7% showed antimicrobial activity with moderate to high inhibition zones because the growth of E. coli reached the range of 6.06 - 15.29 mm. [9]Research on the antibacterial activity of red dragon fruit peel is only limited to the extraction process and there has been no further research related to processed dragon fruit peel products. This study aims to determine the antibacterial properties of red dragon fruit skin yogurt against Bacillus subtilis in hypercholesterolemic rats.

MATERIALS AND METHODS

Materials and Tools

This research uses experimental animal handling and facilities of CNFS laboratory, Universitas Gadjah Mada, Yogyakarta based on the Guidelines for Care and Use of Laboratory Animals. The ethical letter was obtained from the Committee on Ethics of Health, Universitas Negeri Semarang number 140/KEPK/EC/2019. House of Experimental Rats CNFS, Universitas Gadjah Mada, Yogyakarta Indonesia is the provider of Wistar rats used in this study. The inclusion criteria were male Wistar rats, active, aged 8-12 weeks, weighed 150-240 grams with fasting blood glucose level <110 mg/dL, while the exclusion criteria were Wistar rats with anatomical abnormalities and had a weight loss of more than 10% during the study. During the study, no Wistar rats were dropped out because all animals were able to follow the intervention until the end.

The laboratory environment is conditioned to have a dark/light cycle with a ratio of 12 hours:12 hours with a room temperature of 25±1°C to maintain room humidity. The cages were always kept clean by removing feces every day, to minimize the stress of Wistar rats during the study. Wistar rats were placed in individual cages made of stainless steel and received standard feed with ad libitum water access. Wistar rats had an adaptation period of seven days before the study was carried out.

Hypercholesterolemia Induction

Hypercholesterolemic conditions in Wistar rats were obtained through single intraperitoneal injection of feed powder made of 1% cholesterol and 0.5% cholic acid given for 14 days.[14]Cholesterol and cholic acid powder were obtained from Sigma Aldrich, Japan.

Experimental Design

Materials used in this study were caecum from each group of hypercholesterolemic Wistar rats. The study used were K-: DMSO 10% as negative control; K+: chloramphenicol as positive control; K1: caecum of hypercholesterolemic Wistar rats received red dragon fruit peel yogurt as much as 1.8 mL/kg b.wt/day; K2: caecum of hypercholesterolemic

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Wistar rats received red dragon fruit peel yogurt as much as 2.7 mL/kg b.wt/day and K3: caecum of hypercholesterolemic Wistar rats received red dragon fruit peel yogurt as much as 3.6 mL/kg b.wt/day. Other materials required in this study were aqua dest, nutrient agar, chloramphenicol, and Dimethylsulfoxide (DMSO).

The tools used were separating funnel, rotary evaporator, ose needle, petri dish, vennier caliper, incubator, and autoclave. Tools such as separating funnels, ose needles, petri dishes, and vernier caliper were sterilized in an autoclave for 15 minutes at 121°C by regulating a pressure of 1.5 atm having previously been washed clean, dried, and wrapped in paper.

Procedures for Producing Red Dragon Fruit Peel Yogurt

Red dragon fruit peel yogurt was produced based on research by Mardiana[13] and Putriningtyas.[15] Production of red dragon fruit peel yogurt begins with making a starter. Culture rejuvenation or yogurt starter making was carried out using Lactobacillus bulgaricus or Streptococcus thermophilus, each of which was inoculated in full cream pasteurized milk for 18 hours at 42°C. The next step is selecting or sorting the red dragon fruit peel. Fresh red dragon fruit peel is separated from the pulp and cleaned using running water. Red dragon fruit peel is steamed for 15 minutes to remove the distinctive aroma of the peel and reduce the pectin content in the red dragon fruit peel. Red dragon fruit peel is blended with full cream milk in a ratio of 1:4. Suspension of steamed red dragon fruit peel with full cream milk is then added with 10% sucrose, and covered with aluminum foil for the next pasteurization process at a temperature of 75-85^{o}C for 15 minutes. The result of the pasteurized mixture of steamed red dragon fruit peel, full cream milk, and sucrose is cooled to a temperature of 35-37^{o}C then added with 5% Lactobacillus bulgaricus and 5% Streptococcus thermophilus yogurt starter. The last step in making red dragon fruit peel yogurt is incubation at 42^{o}C for seven hours.

Testing the Antibacterial Activity of Red Dragon Fruit Peel Yogurt

The antibacterial activity test of the sample was carried out on Bacillus subtilis obtained from the Microbiology Laboratory of the Center for Food and Nutrition Studies, UGM. Bacillus subtilis was rejuvenated by inoculating pure culture into nutrient agar and incubated at 37°C for 24 hours. Antibacterial activity was measured using the Disc diffusion method (Kirby Bauer).[16] Bacteria from the rejuvenation media were spread on nutrient agar. Empty test discs that have been soaked for 15 minutes in each caecum group of hypercholesterolemic Wistar rats were hygienically placed on the surface of the nutrient agar. The empty discs were immersed in 10 mL of caecum from each group of hypercholesterolemic rats. Media containing test discs were incubated at 37°C for 24 hours and then the diameter of the clear zone was measured using a vernier caliper. The negative control used was DMSO 10%, which was dripped as much as 20 µL on a paper disc. The positive control used in this study was chloramphenicol 0.5 mg. Chloramphenicol is a broad-spectrum antibiotic so it is appropriate to inhibit the growth of gram-positive and negative bacteria. All numerical data were expressed as mean±standard deviation from twice measurements. One-way analysis of variance (ANOVA) with Tukey's test was used to determine significant differences ($p < 0.05$) between means.

RESULTS

Determination of the antibacterial activity of red dragon fruit peel yogurt was carried out by the Kirby-Bauer disc diffusion method, which is the determination of the sensitivity of bacteria to certain substances that may have antibacterial activity, using paper discs. The results of the antibacterial activity of red dragon fruit peel yogurt against Bacillus subtilis in each group can be seen in table 1.

The average inhibition zone of K+; K-; K1; K2; K3 are each 11.50±1.41 mm; 0.00±0.00 mm; 11.50±0.96 mm; 10.13±0.66 mm; 10.38±1.12 mm. DMSO 10% as negative control has no inhibition zone. The categories of antibacterial inhibitory strength are inhibition zone diameter of 5 mm or less is categorized as weak, inhibition zone diameter of 5-10 mm is categorized as a medium, inhibition zone diameter of 10-20 mm is categorized as strong while the diameter of more than 20 mm is categorized as very strong.[10] The positive control using chloramphenicol (K1), which is the caecum of the experimental animal group which received 1.8 mL of red dragon fruit peel yogurt for 28 days, shows the widest inhibition zone.

The DMSO negative control group shows significant differences in the inhibition zone of each treatment group ($p<0.05$). Based on these results, it can be concluded that the red dragon fruit peel yogurt has antibacterial activity against Bacillus subtilis. The difference in the results of the antibacterial activity test in each group can be seen in table 2. Table 2 shows that there is no difference between the positive control group and the experimental animal group that received red dragon fruit peel yogurt at a dose of 1.8 mL and 3.6 mL ($p> 0.05$). The

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experimental animal group that received red dragon fruit peel yogurt with a dose of 1.8 mL shows a significant difference when compared to the group that received yogurt with a dose of 2.7 mL (p= 0.021). Table 2 also shows that there is no statistical difference when the dose is increased to 3.6 mL (p> 0.05).

Table 1. Difference Mean

Group	Deuteronomy1 (mm)	Deuteronomy2 (mm)	Mean±SD (mm)
K-	0	0	0.00±0.00
K+	12.0	11.0	11.50± 1.41
K1	11.25	11.75	11.50± 0.96
K2	10.0	10.25	10.13± 0.66
K3	10.5	10.25	10.38± 1.12

K- (DMSO); K+ (chloramphenicol); K1 (caecum derived from red dragon fruit peel yogurt at a dose of 1.8 mL); K2 (caecum derived from red dragon fruit peel yogurt at a dose of 2.7 mL); K3 (caecum derived from red dragon fruit peel yogurt at a dose of 3.6 mL)

Table 2. Post Hoc

Group	Group	p*
K-	K1	0.001*
	K2	0.001*
	K3	0.001*
K+	K-	0.001*
	K1	0.219
	K2	0.026*
	K3	0.084
K1	K2	0.021*
	K3	0.370
K2	K3	1.000

p significant post hoc test (p<0,05); K- (DMSO); K+ (Chloramphenicol); K1 (caecum derived from red dragon fruit peel yogurt at a dose of 1.8 mL); K2 (caecum derived from red dragon fruit peel yogurt at a dose of 2.7 mL); K3 (caecum derived from red dragon fruit peel yogurt at a dose of 3.6 mL)*

DISCUSSION

Red dragon fruit peel yogurt in this study was made using Lactobacillus bulgaricus and Streptococcus thermophilus as starters. Probiotic activity in yogurt is largely determined by nutrient availability, inoculation levels, incubation temperature, fermentation time, storage conditions, pH, sugar concentration, and content of milk solids. Lactic acid bacteria in yogurt can break down complex carbohydrates into simple carbohydrates thus producing the final product in the form of lactic acid. Lactic acid as a result of fermentation metabolites shows the ability to inhibit microbial growth.[17]

This study used DMSO in negative control and chloramphenicol in positive control. DMSO is a good extract solvent because it can dissolve without affecting the growth of the bacteria so that it does not interfere with the results of the observation. Polar and nonpolar compounds can be dissolved by DMSO.[18] Chloramphenicol gives an inhibitory area, so this shows that chloramphenicol is still sensitive to Bacillus subtilis.[19]Gram-positive bacteria have better antibacterial sensitivity than gram-negative bacteria. The composition and structure of cell walls of gram-positive bacteria are more susceptible to chemical components than gram-negative bacteria.[20] This sensitivity is due to differences in the structure of cell walls. Gram-positive bacteria have a relatively simple cell wall structure, making it easier for antimicrobial compounds to enter the cell and find targets to work. The structure of the cell wall of gram-negative bacteria is relatively more complex and consists of three layers, the outer layer is a lipoprotein, the middle layer is a lipopolysaccharide, and the inner layer is peptidoglycan. The middle layer in the form of lipopolysaccharide acts as a barrier against various antibacterial bioactive materials while the inner layer in the form of peptidoglycan has a high-fat content.[1] The results in table 2 show that there is no difference in the three administration of dragon fruit peel yogurt with chloramphenicol positive control. There was no

significant difference in the group that received 2.4 mL and 3.6 mL of red dragon fruit peel yogurt, so it can be assumed that administering 1.4 mL of red dragon fruit peel yogurt can also be used as antibacterial because it has the same antibacterial activity as the 3.6 mL. The process of making yogurt can trigger the production process of proteolytic enzymes by lactic acid bacteria. This proteolytic enzyme can cut peptide bonds in milk protein to form free amino acids and peptides.

Some peptides show antibacterial activity. Potential peptides that have antibacterial activity derived from milk proteins include Leu-Arg-Leu-Lys-Lys-Tyr-Lys-Val-Pro-Gln-Leu. These peptides are produced by the hydrolysis of pepsin in cow casein.[21] Antibacterial activity of these peptides also depends on various properties such as peptide charges, amphipathicity, and hydrophobic and hydrophilic conditions. This antibacterial activity can also be caused by the presence of several cationic peptides which are believed to interact with lipopolysaccharide anion bonds in cell membranes. This peptide can replace divalent cations such as Ca^{2+} and Mg^{2+}, distorting to the cell's outer membrane bilayer because these two minerals are important to support the integrity of the cell's outer membrane. This distortion in the outer membrane results in membrane lysis and ultimately cell death.[22]

Antibacterial effectiveness of red dragon fruit peel yogurt is possible due to the presence of polyphenols in the peel of red dragon fruit, alkaloids, and terpenoids. The antibacterial activity of phenol compounds is carried out through a protein denaturation process and results in cell lysis. Flavonoids are derivatives of phenolic compounds and act as protein coagulators. The toxicity of phenol compounds to bacteria adjusts the number of hydroxyl groups and the concentration given. Phenol can bind to proteins through hydrogen bonds resulting in damage to the protein structure. [23]

Terpenoids have antibacterial activity by reacting with porin (a transmembrane protein) on the outer membrane of the bacterial cell wall to form strong polymer bonds that cause damage to porin. Porin is the entrance and exit of compounds so that when porin is damaged it will reduce the permeability of bacterial cell walls. This cell wall permeability will interfere with the entry of nutrients and other compounds, thus inhibiting bacterial growth and can even cause bacterial death. Luo stated that red dragon fruit peel contains β-amirin (15.87%) and α-amirin (13.90%) which are the terpenoid group.[22] Other than terpenoids, the antibacterial activity of dragon fruit peel yogurt is also caused by alkaloids. Alkaloids carry out the antibacterial activity by disrupting the peptidoglycan that makes up bacterial cells so that the cell wall layer is not formed completely and causes cell death. Another bioactive substance that is believed to have an antibacterial role in red dragon fruit peel yogurt is flavonoids. Mardiana's research stated that red dragon fruit peel yogurt using Lactobacillus bulgaricus and Streptococcus thermophilus bacterial isolates had higher levels of flavonoids compared to dragon fruit peel yogurt using commercial starters.[12] Flavonoids are known to have antibacterial activity due to their complex formation process with bacterial cell walls. Flavonoids work by damaging bacterial cell membranes in the phospholipid portion to further reduce membrane permeability. The decrease in membrane phospholipid permeability due to the activity of phenolic components causes lysis of bacterial cell membranes. Saponin activity mechanism as an antibacterial is possible because saponins can cause proteins and enzymes leakage in cells. Tannins also play a role in inhibiting bacterial growth by damaging bacterial cell proteins.[20] Other alkaloid compounds found in the peel of red dragon fruit are betacyanins (betanin and isobetanin). The fermentation process in making yogurt of red dragon fruit peel causes an increase in the content of betacyanin.[24] Alkaloids act as antibacterial through an inhibitory mechanism of DNA synthesis. Alkaloid compounds have alkaline groups that contain nitrogen which will react with amino acid compounds that make up the bacterial cell walls and DNA. This reaction will result in changes in the structure and arrangement of amino acids, causing changes in the genetic balance of the DNA chain. This will cause bacterial cell lysis which will then cause cell death in bacteria. [25]Alkaloids can also inhibit bacterial growth through the action mechanism of the dihydrofolate reductase enzyme so that it inhibits nucleic acid synthesis. [26]

The limitations of this study are that the phytochemical (tannins, alkaloids, and terpenoids) tests on red dragon fruit peel yogurt were not carried out.

CONCLUSIONS

Red dragon fruit peel yogurt with a dose of 2.7 mL has the potential to inhibit the growth of Bacillus subtilis bacteria.

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REFERENCES

1. Waglechner, N. & Wright, G. D. Antibiotic resistance : it’s bad, but why isn't ’ t it worse ?

BMC Biology. 2017; 15(84): 1–8.
2. Davies, J. & Davies, D. Origins and Evolution of Antibiotic Resistance. Microbiol. Mol. Biol. Rev. 2010; 74 (3): 417–433.
3. Earl, A. M., Losick, R. & Kolter, R. Ecology and Genomics of Bacillus subtilis. Trends Microbiol. 2008; 16(6): 1–11.
4. Rahayu, W. P. & Nurwitri, C. Mikrobiologi Pangan. IPB Press; 2012.
5. Jaafar, R. A., Ridhwan, A., Mahmod, N. Z. C. & Vasudevan, R. Proximate Analysis of Dragon Fruit (Hylocereus polyhizus). Am. J. Appl. Sci. 2009; 6(7): 1341–1346.
6. Jaya, I. K. D. Morphology And Physiology Of Pitaya And It Future Prospects In Indonesia. Crop Agro 2010; 3 (1): 44–50.
7. Nurliyana, R., Zahir, S., Suleiman, M., Aisyah, M. & Rahim, K. Antioxidant study of pulps and peels of dragon fruits : a comparative study. International Food Research Journal. 2010; 17: 367–375.
8. Amalia, S., Wahdaningsih, S. & Untari, E. K. Antibacterial Activity Testing Of N-Hexane Fraction Of Red Dragon (Hylocereus polyrhizus Britton & Rose) Fruit Peel On Staphylococcus aureus ATCC 25923. Traditional Medicine Journal 2014; 19(2): 89–94.
9. Fitratullah, A., Maruddin, F., Yuliati, F., Prahesti, K. & Taufik, M. Addition of red dragon fruit (Hylocereus polyrhizus) on yogurt : Effect on lactic acid content, pH, and the inhibition of Escherichia coli growth. in *IOP Conf. Series: Earth and Environmental Science 2019; 343:* 1–7 (IOP Publishing). doi:10.1088/1755-1315/343/1/012034.
10. Maulana, I. *et al.* Antibacterial Test of Red Dragon Fruit Extract Peel (Hylocereus polyrhizus) Against Bacteria Salmonella pullorum. Jurnal Medika Veterinaria 2018; 12 (1): 9–14.
11. Septiana, W. C. & Ardiaria, M. Efek Pemberian Seduhan Kulit Buah Naga Merah (Hylocereus polyrhizus) terhadap Kadar Malondialdehyde (MDA) Tikus Sprague Dawley Dislipidemia. Journal of Nutrition College 2017; 5 (4): 344–352.
12. Mardiana, Budiono, I. & Putriningtyas, N. D. Comparison of organoleptic, protein, lipid and flavonoid content of commercial starter and isolated culture red dragon fruit peel yogurt. Food Research. 2020: 4(3): 920–925.
13. Ayala, F. R. *et al*. Microbial flora, probiotics, Bacillus subtilis and the search for a long and healthy human longevity. Microbial Cell 2017; 4 (4): 133–136.
14. Murase, T., Yamada, N. & Uchimura, H. Accumulation of intermediate-density lipoprotein in the plasma of cholesterol-fed hypothyroid rats. Metabolism 1983; 32 (2): 146–150.
15. Putriningtyas, N. D. & Wahyuningsih, S. Potensi yogurt kacang merah (Phaseolus vulgaris L) ditinjau dari sifat organoleptik, kandungan protein, lemak dan flavonoid. Jurnal Gizi Indonesia (The Indonesian Journal of Nutrition). 2017*;* 6 (1): 37–43.
16. Prayoga, E. Perbandingan Efek Ekstrak Daun Sirih Hijau (Piper betle L.) dengan Metode Difusi Disk dan Sumuran terhadap Pertumbuhan Bakteri Staphylococcus aureus. [Tesis]. Jakarta: Universitas Islam Negeri Syarif Hidayatullah; 2013. doi:10.1023/B:FOOP.0000019620.04821.a2.
17. Maruddin, F., Malaka, R. & Taufi, M. Characteristics and antimicrobial activity of dangke whey fermentation with sugar addition. Bulgarian Journal of Agricultural Science. 2019; 25 (2): 410–417.
18. Octaviani, M., Fadhli, H. & Yuneistya, E. Uji Aktivitas Antimikroba Ekstrak Etanol dari Kulit Bawang Merah (Allium cepa L.) dengan Metode Difusi Cakram. Pharm. Sci. Res. 2019*;* 6 (1): 62–68.
19. Fitriani, E. Uji Aktivitas Antibakteri Ekstrak Etanol Daun Sirsak (Annona muricata L.) terhadap Shigella flexneri Secara In Vitro. [Tesis]. Pontianak: Universitas Tanjungpura; 2014.
20. Ritarwan, K. & Nerdy, N. Antibacterial Activity of Red Dragon Fruit Leaves Extract and White Dragon Fruit Leaves Extract Against Meningitis Bacterial. Orient. J. Chem. 2018; 34 (5): 2534–2538.
21. Sah, B. N. P., Vasiljevic, T., Mckechnie, S. & Donkor, O. N. Antibacterial and antiproliferative peptides in synbiotic yogurt — Release and stability during refrigerated storage. J. Dairy Sci. 2016; 99: 4233–4242.
22. Luo, H., Cai, Y., Peng, Z., Liu, T. & Yang, S. Chemical composition and in vitro evaluation of the cytotoxic and antioxidant activities of supercritical carbon dioxide extracts of pitaya (dragon fruit) peel. Chemistry Central Journal. 2014; 8 (1): 1–7.
23. Karimi, S., Azizi, F., Nayeb-aghaee, M. & Mahmoodnia, L. The Antimicrobial Activity of Probiotic Bacteria Escherichia coli Isolated

from Different Natural Sources Against Hemorrhagic E. coli O157:H7. Electron. Physician 2018; 10 (3): 6548–6553.
24. Choo, K. Y., Kho C., Ong, Y. Y., Thoo, Y.Y., Lim, R.L. H., Tan, C.P., et al. Studies on the storage stability of fermented red dragon fruit (Hylocereus polyrhizus) drink. Food Sci. Biotechnol. 2018; 27: 1411–1417.
25. Wahdaningsih, S., Untari, E. K. & Fauziah, Y. Antibakteri Fraksi n-Heksana Kulit Hylocereus polyrhizus Terhadap Staphylococcus epidermidis dan Propionibacterium acnes. Pharm. Sci. Res. 2014; 1 (3): 180–193.
26. Astridwiyanti, A. A. B., Mahendra, A. N. & Dewi, N. W. S. Uji efektivitas ekstrak etanol kulit buah naga merah (Hylocereus polyrhizus) terhadap Staphylococcus aureus ATCC 25923 secara in vitro. Intisari Sains Medis 2019; 10 (3): 482–486.

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Differences in triglyceride levels before and after whey protein intervention in field workers exposed and unexposed to arsenic

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ABSTRACT

Background: A work environment that is exposed to heavy metals, such as a coal mining environment, can change fat metabolism in the body. Changes in fat metabolism will lead to cardiovascular disease. Consumption of dairy products, e.g. whey protein, can reduce the risk of metabolic disorders and cardiovascular disease.
Objectives: To test and analyze the differences in triglyceride levels before and after whey protein intervention to field workers with different working conditions.
Materials and Methods: This experimental research with pretest and posttest was conducted on field workers at PT Bukit Asam Tbk. Tarahan Port Unit, Lampung as an Arsenic exposed group and PTPN VII Way Berulu Business Unit as an Arsenic unexposed group. Both groups received 24 grams of whey protein daily for 28 days. The triglyceride levels before and after the intervention were measured by laboratory analysis using the enzymatic calorimetry method. The data were analyzed using independent t-test, Mann Whitney test, and Wilcoxon test.
Results: The average triglyceride levels increased by 50.48 ± 98.09 mg/dL in the exposed group and 16.78 ± 67.67 mg/dL in the unexposed group. There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups.
Conclusions: The whey protein intervention increased the triglyceride level in the exposed group and decreased it in the unexposed group.

Keywords: Triglycerides; Whey Protein; Arsenic (As).

BACKGROUND

With a good nutritional status and health status, the workforce will provide maximum results for the company. The work environment greatly influences their nutritional needs and health status. The work environment with exposure to chemical substances will affect the health status of the workers if it is not properly attended to by the manager.[1] Mining work environment, such as coal mining, is susceptible to the exposure of hazardous chemicals. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.[2,3] Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. It can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.[4]

Some epidemiological studies have shown that the consumption of dairy products can reduce the risk of metabolic disorders and cardiovascular disease.[5] Protein in cow's milk consists of 80% casein and 20% whey protein, both of which are rich in amino acids.[6] Whey protein is a food source that has high protein and essential amino acids, as well as branched-chain amino acids (BCAA) that play an important role in tissue growth and repair.[5] The results of the previous studies indicate that after the intervention of whey protein on 20 male subjects aged 30 to 50 years with relatively high serum cholesterol levels (≥ 200 mg/dL), their serum triglyceride levels decreased and their serum HDL levels increased after 8 weeks of intervention.[7]

MATERIALS AND METHODS

This research is an experimental study with pretest and posttest. The sample was assigned into two groups, Arsenic (As) exposed group (field workers of PT Bukit Asam Tbk. Tarahan Port Unit, Lampung, a coal mining product management company) and Arsenic (As) unexposed group (field workers of PT Perkebunan Nusantara VII Way Berulu Business Unit, a company engaged in the

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management of rubber plantations). Both groups received intervention in the form of taking whey protein for 28 days[8] with a dose of 24 grams per day dissolved in 300 ml of mineral water.[9] The variable measured was the subjects' triglyceride levels before and after the whey protein intervention.

This research was conducted after obtaining permission from the Health Research Ethics Committee of the Faculty of Medicine, Diponegoro University, Semarang, from January to March 2020. Whey protein intervention was carried out at the research site, PT Bukit Asam Tbk. Tarahan Port Unit in Lampung and PT Perkebunan Nusantara VII Way Berulu Business Unit. Subject characteristic data including name, age, and place of birth were collected through interviews using a questionnaire. The anthropometric data of the subjects consisted of body weight, measured using digital scales, and height, measured using a microtoise. Body mass index (BMI) was calculated using the BMI calculation formula based on measurements of weight and height. The physical activity data of the subjects were collected through interviews using the Physical Activity Level (PAL) questionnaire and then calculated with the PAL calculation formula. Food intake data were collected through interviews using a 24-hour food recall questionnaire 3 times each week during the study. The triglyceride levels before and after the intervention were measured by laboratory analysis using the enzymatic calorimetry method.

Data analysis was carried out through the SPSS program with the Shapiro-Wilk test to determine the normality of data distribution for each group, independent t-test, and Mann Whitney test to determine the average difference between the subject's characteristics and nutritional intake variables. Wilcoxon test was also used to determine differences in triglyceride levels before and after the intervention.

RESULTS

The data in this study are primary data taken from the research sample, field workers exposed to different work environments. The total sample consisted of 64 people (31 from the exposed group and 33 from the unexposed group).

Subject Characteristics

Subject characteristics data consisted of age, body weight (BW), body height (BH), body mass index (BMI), and physical activity level (PAL). They are shown in Table 1.

Table 1. Subject Characteristics in Both Groups

	Exposed (n = 31)		Unexposed (n = 33)		*p*
	Mean±SD	**minimum-maximum**	**Mean±SD**	**minimum-maximum**	
Age (year)	29.84±8.84	20-56	38.94±4.50	28-54	0.000[2]
Body weight (kg)	70.87±14.11	45-109	56.55±6.96	44.08-75.00	0.000[1]
Body height (cm)	168.03±5.89	154-179	162.01±4.84	153-171	0.000[1]
BMI (kg/m^2)	25.31±4.21	18.09-36.84	21.55±2.26	17.56-26.23	0.000[1]
PAL (unit)	1.57±0.13	1.41-1.87	1.99±0.24	1.44-2.78	0.000[2]

[1] difference test using Independent T-Test, [2] difference test using Mann Whitney

Table 1 shows significant differences in all variable subject characteristics ($p < 0.05$). The average weight, height, and BMI were higher in the exposed group compared to the unexposed group, whereas the average age and PAL were higher in the unexposed group.

Nutritional Intake

Nutritional intake data consists of total intake per day and intake adequacy. Nutritional intake data are presented in Table 2 and the data on the intake adequacy levels in both groups in Table 3.

Table 2. Overview of Total Nutritional Intake Per Day in Both Groups

	Exposed (n = 31)		Unexposed (n = 33)		*p*
	Mean±SD	**minimum-maximum**	**Mean±SD**	**minimum-maximum**	
Energy (kcal)	2018.72±452.88	1313.50-3314.90	2149.83±515.61	1213.20-3890.00	0.285[1]
Protein (g)	96.21±29.04	56.10-180.90	86.62±26.66	56.30-167.30	0.116[2]
Fat (g)	83.89±40.50	35.80-153.30	67.01±30.06	22.30-170.50	0.134[2]
Carbohydrate (g)	252.30±68.47	148.7-452.80	298.47±66.39	191.80-485.90	0.004[2]

[1] difference test using Independent T-Test, [2] difference test using Mann Whitney

Table 2 shows that the average energy and carbohydrate intake in the unexposed group was higher than those of the exposed group. The average protein and fat intake show a higher value in the exposed group. There was no significant difference in the average energy, protein, and fat intake, but there was a significant difference in the average carbohydrate intake (p = 0.004).

Table 3. Nutritional Intake Adequacy Level in Both Groups

Adequacy Level		Category	
		Exposed (n = 31)	Unexposed (n = 33)
Energy	Deficient	23 (74.2%)	26 (78.8%)
	Sufficient	8 (25.8%)	7 (21.2%)
Protein	Deficient	14 (45.2%)	21 (63.6%)
	Sufficient	17 (54.8%)	12 (36.4%)
Fat	Deficient	13 (41.9 %)	22 (66.7%)
	Sufficient	15 (48.4%)	9 (27.3%)
	Excessive	3 (9.7%)	2 (6.1%)
Carbohydrate	Deficient	28 (90.3%)	26 (78.8%)
	Sufficient	3 (9.7%)	7 (21.2%)

Table 3 shows that almost all of the adequacy levels of energy, protein, fat, and carbohydrate intakes in both groups were insufficient. Meanwhile, the fat adequacy of 9.7% of the total subjects in the exposed group and 6.1% in the unexposed group fell into the excessive category.

Triglyceride Levels Before and After Whey Protein Intervention

The triglyceride levels before and after whey protein intervention were tested for normality using the Shapiro Wilk test, resulting in the abnormal distribution of triglyceride levels with a p-value of < 0.05; thus, the Wilcoxon test was used to see the differences in triglyceride levels before and after the whey protein intervention.

Table 4. Triglyceride Levels Before and After Whey Protein Intervention in Both Groups

Triglyceride Levels (mg/dL)	Exposed (n = 31)		Unexposed (n = 33)		*p*
	Mean±SD	minimum-maximum	Mean±SD	min-max	
Before	125.32±68.97	35.00-310.00	157.18±104.82	38.00-562.00	0.202[1]
After	175.80±127.99	48.00-590.00	140.39±80.78	43.00-425.00	0.344[1]
Difference/delta (Δ)	50.48±98.09	414.00(-68.00)	-16.78±67.67	184.00(-137.00)	0.000[1]
p	0.003[2]		0.038[2]		

[1] difference test using Mann Whitney, [2] difference test using Wilcoxon

Table 4 shows that the average triglyceride level before the intervention in the unexposed group was higher than in the exposed group, and the average triglyceride level after the intervention in the exposed group was higher. The average triglyceride level in the exposed group increased by 50.48 ± 98.09 mg/dL and in the unexposed group decreased by 16.78 ± 67.67 mg/dL. The Wilcoxon test results showed differences in triglyceride levels in both groups (p < 0.05).

DISCUSSION

This study explores the differences in triglyceride levels in field workers with different work environment exposures before and after the whey protein intervention. The groups in this study consisted of field workers in a coal mining environment with exposure to Arsenic, an obesogenic substance, and field workers in a rubber plantation environment. Whey protein intervention was carried out for 28 days with a dose of 24 grams per person per day.

The results showed that the intervention could reduce triglyceride levels in the obesogen unexposed group. Meanwhile, in the exposed group, there was an increase in triglyceride levels after the intervention. The decrease in triglyceride levels in the unexposed group was due to several factors, such as lower BMI value, higher PAL value, and lower fat intake of the unexposed group compared to those of the exposed group.

On the other hand, the triglyceride level in the exposed group was found to increase after the whey protein intervention. This is due to several factors, including higher BMI value, lower PAL, and higher

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fat intake, as well as Arsenic exposure that occurs every day in the work environment of the exposed group.

The BMI value of the unexposed group was lower than that of the exposed group. Previous research has suggested that there is a positive correlation between BMI and triglyceride levels. High BMI is directly proportional to triglyceride levels in the body.[10] The linear relationship between BMI and triglyceride levels is seen in individuals with higher BMI between 18 to 35 kg/m^2, posing a 5.1% greater risk of having high triglyceride levels.[11] The PAL value of the exposed group was lower than that of the unexposed group. Previous research found that moderate to heavy PAL could increase the average HDL level between 0.89 to 1.71 mg/dL and reduce the average triglyceride level between 0.93 to 0.98 mg/dL.[12] Besides, fat intake also greatly affects triglyceride levels in the body. Based on the results of the 3×24 hour recall in the study, fat intake in the exposed group was higher than that in the non-exposed group. The results of statistical tests also indicate the same, where the average fat intake of the exposed group is higher than that of the unexposed group. Previous research has shown that an increase in fat by 10% of daily needs will lead to increased triglyceride levels by 0.06 mmol/L.[13]

Arsenic exposure that occurs every day in the work environment of the exposed group also influences changes in lipid metabolism in the body. Based on a study conducted on ApoE mice exposed to moderate Arsenic, it was found that mice with a normal diet and received 200 ppb-Arsenic exposure for 8 weeks had a significant increase in plasma triglyceride levels and triglyceride levels. The average triglyceride levels of mice exposed to 0 ppb, 200 ppb, and 1,000 ppb of Arsenic for 8 weeks were 138 ± 18 mg/dL, 233 ± 29 mg/dL, and 178 ± 26 mg/dL, respectively. This implies that chronic Arsenic exposure will increase triglyceride levels in the body.[14]

Triglycerides or triacylglycerols are formed from three fatty acids and mono glycerol. Triglycerides function as energy substances. As cells need energy, the lipase enzyme in fat cells breaks down triglycerides into glycerol and fatty acids to be released into the blood vessels.[15] Triglyceride synthesis is influenced by Growth Hormone (GH). GH increases the activity of Hormone Sensitive Lipase (HSL) which can break down triglycerides into free fatty acids in fat tissue.[15] Triglyceride synthesis occurs when the energy source from carbohydrates is sufficient, and then the fatty acids will experience esterification, forming esters with glycerol and triglycerides as long-term energy reserves. Once there is no energy reserve from carbohydrates, the fatty acids and triglyceride reserves in the tissues will be broken down through the lipolysis process. Triglyceride levels in the body are influenced by several factors including genes, age, gender, food intake, obesity, physical activity, and smoking habits. The accumulation of excess triglyceride levels in the body will increase the risk of dyslipidemia. Dyslipidemia is a condition of an abnormal lipid profile in the body that increases levels of total cholesterol, triglycerides, and LDL and decreases HDL levels in the blood.[16]

Milk consists of two protein contents, 80% casein and 20% whey. When casein is separated from whole milk, the whey will remain and contain 68% protein.[5] Whey protein is a liquid byproduct of cheese making. Whey protein consists of several biological components, including BCAA which are easily absorbed and used by the body, lactoferrin as a non-enzymatic antioxidant, immunoglobulin as an antibody, β-Lactoglobulin as BCAA that plays a role in retinol-binding and can modulate the lymphatic response in humans, α-Lactalbumin which can increase antibody response to systematic antigen stimulation and suppress T cell response in lymphocytes, lactoperoxidase which can remove harmful bacteria for the human body, glycomacropeptide, and serum albumin.[17]

The high protein and amino acid content in whey protein make it an alternative food source of protein and amino acids with many health benefits. Increased protein intake can lead to changes in circulating lipids and lipoprotein levels, such as decreased LDL, decreased serum triglycerides, and increased HDL concentrations.[18] Research on female Wistar rats showed that whey protein concentrate supplementation could reduce cholesterol levels and serum triglyceride concentrations after consumption for 8 weeks compared to casein supplementation.[19] The decreased lipid profile levels with whey protein consumption occur through the role of BCAA in whey which accelerates digestion and absorption rates in intestines. β-Lactoglobulin content helps the absorption of cholesterol in the intestine, inhibits the expression of genes in charge of the absorption and synthesis of intestinal fatty acids and cholesterol, and increases the excretion of fecal steroids.[20]

Global competition demands a productive workforce for companies. A productive workforce will be created through good nutritional and health status. Some epidemiological studies have shown that nutrient intake is positively associated with labor productivity.[1] The work environment condition is one factor that influences the health and nutritional status of the workforce. A work environment with exposure

to hazardous chemicals such as a mining environment can affect the health status of workers. Coal, one of the largest natural resources in Indonesia, creates many job opportunities in the coal mining environment. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.[2,3] Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. Arsen can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.[4]

LIMITATION

This study did not analyze the levels of Arsenic (As) in the body of the research subjects, so the exact amount of exposure that affects triglyceride levels in the body remains unknown. Further research on this topic still needs to be conducted.

CONCLUSIONS

There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups. Whey protein intervention led to an increase in triglyceride levels in the exposed group and a decrease in triglyceride levels in the unexposed group although only a slight decrease was produced with the whey protein intervention for 28 days.

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REFERENCES

1. Suma'mur. Higiene Perusahaan dan Kesehatan Kerja (Hiperkes). Jakarta: Sagung Seto; 2013.
2. Radi, F. Hasni, M.J. Obesogens As An Environmental Risk Factor For Obesity. Malays J Public Health Med. 2014;14 (3):63–70.
3. Baillie-Hamilton P. Chemical Toxins: A Hypothesis To Explain The Global Obesity Epidemic. J Altern Complement Med. 2002;8:185–92.
4. Ceja-Galicia ZA, Daniel A, Salazar AM, Panico P, Ostrosky-Wegman P, Diaz-Villasenor A. Effects of arsenic on adipocyte metabolism: Is arsenic an obesogen? Elsevier Mol Cell Endocrinol Xxx. 2017;1-8.
5. Marshall K. Therapeutic applications of whey protein. Altern Med Rev. 2004;9:136–156.
6. Furst P, Stehle P. What Are The Essential Elements Needed For The Determination of Amino Acid Requirements in Humans ? J Nutr. 2004;134:1558–65.
7. Kawase M, Hashimoto H, Hosoda M, Morita A, Hasono A. Effect of Administration of Fermented Milk Containing Whey Protein Concentrate to Rats and Healthy Men on Serum Lipids and Blood Pressure. J Diet Sci. 2000; Vol. 83, No. 2.
8. Bhathena S, Velasquez M. Beneficial Role Of Dietary Phytoestrogens In Obesity And Diabetes. Am J Clin Nutr. 2002;76:1191–1201.
9. Vatani, DS, Golzar, FAK. Changes in Antioxidant Status and Cardiovascular Risk Factors of Overweight Young Men After Six Weeks Supplementation of Whey Protein Isolate and Resistance Training. Elsevier Ltd. 2012;59(Appetite):673–8.
10. Shamai L, Lurix E, Shen M, Novaro GM, Szomstein S, Rosenthal R, et al. Association of Body Mass Index and Lipid Profiles: Evaluation of a Broad Spectrum of Body Mass Index Patients Including the Morbidly Obese. 2010;21 (1)(Obesity Surgery):42–47.
11. Hansen SEJ, Madsen CM, Varbo A, Nordestgaard BG. Body mass index, triglycerides and risk of acute pancreatitis: A population-based study of 118,000 individuals. 2019;(The Journal of Clinical Endocrinology&Metabolism.).
12. Silva Rc, Diniz M, Alvim S, Vidigal P, Fedeli L, Barreto S. Physical Activity and Lipid Profile in the ELSA-Brasil Study. 2016;107(1):10–9.
13. Kdekian A, Alssema M, Van Der Beek EM, Greyling A, Vermeer MA, Mela DJ, et al. Impact of isocaloric exchanges of carbohydrate for fat on postprandial glucose, insulin, triglycerides, and free fatty acid responses-a systematic review and meta-analysis. Eur J Clin Nutr. 2019.

14. Lemaire M, Lemarié CA, Flores Molina M, Schiffrin EL, Lehoux S, Mann KK. Exposure to Moderate Arsenic Concentrations Increases Atherosclerosis in ApoE−/− Mouse Model. Oxf Univ Press. 2011;122 (1)(Toxicological Sciences):211–221.

15. Karyadi E. Hidup Bersama Penyakit Hipertensi, Asam Urat, Jantung Koroner. Jakarta: PT. Intisari Mediatama; 2006.

16. Lichtenstein AH. A. H. Diet and Lifestyle Recommendations Revision 2006: A Scientific Statement From the American Heart Association Nutrition Committee. Dalam 2006. Page 82–96. (Circulation; vol. 114 (1)).

17. Anthony JC, Anthony TG, Kimball SR, Jefferson LS. Signaling Pathways Involved In Translational Control Of Protein Synthesis In Skeletal Muscle By Leucine. J Nutr. 2001;131:856S-860S.

18. Pal S, Radavelli-Bagatini S. The Effects Of Whey Protein On Cardiometabolic Risk Factors. Obes Rev. April 2012.

19. Zhang, X., Beynen, A. C. Lowering effect of dietary milk-whey protein v. casein on plasma and liver cholesterol concentrations in rats. 1993;70(British Journal of Nutrition):139.

20. Pal S, Ellis V, Dhaiwal S. Effects of Whey Protein Isolate on Body Composition, Lipids, Insulin and Glucose in Overweight and Obese Individuals. Br J Nutr. 2010;104:716-723.

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Differences in triglyceride levels before and after whey protein intervention in field workers exposed and unexposed to arsenic

Eka Putri Rahmadhani[1*], Ani Margawati[1], Apoina Kartini[2]

ABSTRACT

Background: A work environment that is exposed to heavy metals, such as a coal mining environment, can change fat metabolism in the body. Changes in fat metabolism will lead to cardiovascular disease. Consumption of dairy products, e.g. whey protein, can reduce the risk of metabolic disorders and cardiovascular disease.
Objectives: To test and analyze the differences in triglyceride levels before and after whey protein intervention to field workers with different working conditions.
Materials and Methods: This experimental research with pretest and posttest was conducted on field workers at PT Bukit Asam Tbk. Tarahan Port Unit, Lampung as an Arsenic exposed group and PTPN VII Way Berulu Business Unit as an Arsenic unexposed group. Both groups received 24 grams of whey protein daily for 28 days. The triglyceride levels before and after the intervention were measured by laboratory analysis using the enzymatic calorimetry method. The data were analyzed using independent t-test, Mann Whitney test, and Wilcoxon test.
Results: The average triglyceride levels increased by 50.48 ± 98.09 mg/dL in the exposed group and 16.78 ± 67.67 mg/dL in the unexposed group. There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups.
Conclusions: The whey protein intervention increased the triglyceride level in the exposed group and decreased it in the unexposed group.

Keywords: Triglycerides; Whey Protein; Arsenic (As).

BACKGROUND

With a good nutritional status and health status, the workforce will provide maximum results for the company. The work environment greatly influences their nutritional needs and health status. The work environment with exposure to chemical substances will affect the health status of the workers if it is not properly attended to by the manager.[1] Mining work environment, such as coal mining, is susceptible to the exposure of hazardous chemicals. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.[2,3] Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. It can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.[4]

Some epidemiological studies have shown that the consumption of dairy products can reduce the risk of metabolic disorders and cardiovascular disease.[5] Protein in cow's milk consists of 80% casein and 20% whey protein, both of which are rich in amino acids.[6] Whey protein is a food source that has high protein and essential amino acids, as well as branched-chain amino acids (BCAA) that play an important role in tissue growth and repair.[5] The results of the previous studies indicate that after the intervention of whey protein on 20 male subjects aged 30 to 50 years with relatively high serum cholesterol levels (≥ 200 mg/dL), their serum triglyceride levels decreased and their serum HDL levels increased after 8 weeks of intervention.[7]

MATERIALS AND METHODS

This research is an experimental study with pretest and posttest. The sample was assigned into two groups, Arsenic (As) exposed group (field workers of PT Bukit Asam Tbk. Tarahan Port Unit, Lampung, a coal mining product management company) and Arsenic (As) unexposed group (field workers of PT Perkebunan Nusantara VII Way Berulu Business Unit, a company engaged in the

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management of rubber plantations). Both groups received intervention in the form of taking whey protein for 28 days[8] with a dose of 24 grams per day dissolved in 300 ml of mineral water.[9] The variable measured was the subjects' triglyceride levels before and after the whey protein intervention.

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Data analysis was carried out through the SPSS program with the Shapiro-Wilk test to determine the normality of data distribution for each group, independent t-test, and Mann Whitney test to determine the average difference between the subject's characteristics and nutritional intake variables. Wilcoxon test was also used to determine differences in triglyceride levels before and after the intervention.

RESULTS

The data in this study are primary data taken from the research sample, field workers exposed to different work environments. The total sample consisted of 64 people (31 from the exposed group and 33 from the unexposed group).

Subject Characteristics

Subject characteristics data consisted of age, body weight (BW), body height (BH), body mass index (BMI), and physical activity level (PAL). They are shown in Table 1.

Table 1. Subject Characteristics in Both Groups

	Exposed (n = 31)		Unexposed (n = 33)		*p*
	Mean±SD	**minimum-maximum**	**Mean±SD**	**minimum-maximum**	
Age (year)	29.84±8.84	20-56	38.94±4.50	28-54	0.000[2]
Body weight (kg)	70.87±14.11	45-109	56.55±6.96	44.08-75.00	0.000[1]
Body height (cm)	168.03±5.89	154-179	162.01±4.84	153-171	0.000[1]
BMI (kg/m^2)	25.31±4.21	18.09-36.84	21.55±2.26	17.56-26.23	0.000[1]
PAL (unit)	1.57±0.13	1.41-1.87	1.99±0.24	1.44-2.78	0.000[2]

[1] difference test using Independent T-Test, [2] difference test using Mann Whitney

Table 1 shows significant differences in all variable subject characteristics ($p < 0.05$). The average weight, height, and BMI were higher in the exposed group compared to the unexposed group, whereas the average age and PAL were higher in the unexposed group.

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Nutritional intake data consists of total intake per day and intake adequacy. Nutritional intake data are presented in Table 2 and the data on the intake adequacy levels in both groups in Table 3.

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Protein (g)	96.21±29.04	56.10-180.90	86.62±26.66	56.30-167.30	0.116[2]
Fat (g)	83.89±40.50	35.80-153.30	67.01±30.06	22.30-170.50	0.134[2]
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Table 2 shows that the average energy and carbohydrate intake in the unexposed group was higher than those of the exposed group. The average protein and fat intake show a higher value in the exposed group. There was no significant difference in the average energy, protein, and fat intake, but there was a significant difference in the average carbohydrate intake (p = 0.004).

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Adequacy Level		Category	
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Fat	Deficient	13 (41.9 %)	22 (66.7%)
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Table 3 shows that almost all of the adequacy levels of energy, protein, fat, and carbohydrate intakes in both groups were insufficient. Meanwhile, the fat adequacy of 9.7% of the total subjects in the exposed group and 6.1% in the unexposed group fell into the excessive category.

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Triglyceride Levels (mg/dL)	Exposed (n = 31)		Unexposed (n = 33)		*p*
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After	175.80±127.99	48.00-590.00	140.39±80.78	43.00-425.00	0.344[1]
Difference/delta (Δ)	50.48±98.09	414.00(-68.00)	-16.78±67.67	184.00(-137.00)	0.000[1]
p	0.003[2]		0.038[2]		

[1] difference test using Mann Whitney, [2] difference test using Wilcoxon

Table 4 shows that the average triglyceride level before the intervention in the unexposed group was higher than in the exposed group, and the average triglyceride level after the intervention in the exposed group was higher. The average triglyceride level in the exposed group increased by 50.48 ± 98.09 mg/dL and in the unexposed group decreased by 16.78 ± 67.67 mg/dL. The Wilcoxon test results showed differences in triglyceride levels in both groups (p < 0.05).

DISCUSSION

This study explores the differences in triglyceride levels in field workers with different work environment exposures before and after the whey protein intervention. The groups in this study consisted of field workers in a coal mining environment with exposure to Arsenic, an obesogenic substance, and field workers in a rubber plantation environment. Whey protein intervention was carried out for 28 days with a dose of 24 grams per person per day.

The results showed that the intervention could reduce triglyceride levels in the obesogen unexposed group. Meanwhile, in the exposed group, there was an increase in triglyceride levels after the intervention. The decrease in triglyceride levels in the unexposed group was due to several factors, such as lower BMI value, higher PAL value, and lower fat intake of the unexposed group compared to those of the exposed group.

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fat intake, as well as Arsenic exposure that occurs every day in the work environment of the exposed group.

The BMI value of the unexposed group was lower than that of the exposed group. Previous research has suggested that there is a positive correlation between BMI and triglyceride levels. High BMI is directly proportional to triglyceride levels in the body.[10] The linear relationship between BMI and triglyceride levels is seen in individuals with higher BMI between 18 to 35 kg/m^2, posing a 5.1% greater risk of having high triglyceride levels.[11] The PAL value of the exposed group was lower than that of the unexposed group. Previous research found that moderate to heavy PAL could increase the average HDL level between 0.89 to 1.71 mg/dL and reduce the average triglyceride level between 0.93 to 0.98 mg/dL.[12] Besides, fat intake also greatly affects triglyceride levels in the body. Based on the results of the 3×24 hour recall in the study, fat intake in the exposed group was higher than that in the non-exposed group. The results of statistical tests also indicate the same, where the average fat intake of the exposed group is higher than that of the unexposed group. Previous research has shown that an increase in fat by 10% of daily needs will lead to increased triglyceride levels by 0.06 mmol/L.[13]

Arsenic exposure that occurs every day in the work environment of the exposed group also influences changes in lipid metabolism in the body. Based on a study conducted on ApoE mice exposed to moderate Arsenic, it was found that mice with a normal diet and received 200 ppb-Arsenic exposure for 8 weeks had a significant increase in plasma triglyceride levels and triglyceride levels. The average triglyceride levels of mice exposed to 0 ppb, 200 ppb, and 1,000 ppb of Arsenic for 8 weeks were 138 ± 18 mg/dL, 233 ± 29 mg/dL, and 178 ± 26 mg/dL, respectively. This implies that chronic Arsenic exposure will increase triglyceride levels in the body.[14]

Triglycerides or triacylglycerols are formed from three fatty acids and mono glycerol. Triglycerides function as energy substances. As cells need energy, the lipase enzyme in fat cells breaks down triglycerides into glycerol and fatty acids to be released into the blood vessels.[15] Triglyceride synthesis is influenced by Growth Hormone (GH). GH increases the activity of Hormone Sensitive Lipase (HSL) which can break down triglycerides into free fatty acids in fat tissue.[15] Triglyceride synthesis occurs when the energy source from carbohydrates is sufficient, and then the fatty acids will experience esterification, forming esters with glycerol and triglycerides as long-term energy reserves. Once there is no energy reserve from carbohydrates, the fatty acids and triglyceride reserves in the tissues will be broken down through the lipolysis process. Triglyceride levels in the body are influenced by several factors including genes, age, gender, food intake, obesity, physical activity, and smoking habits. The accumulation of excess triglyceride levels in the body will increase the risk of dyslipidemia. Dyslipidemia is a condition of an abnormal lipid profile in the body that increases levels of total cholesterol, triglycerides, and LDL and decreases HDL levels in the blood.[16]

Milk consists of two protein contents, 80% casein and 20% whey. When casein is separated from whole milk, the whey will remain and contain 68% protein.[5] Whey protein is a liquid byproduct of cheese making. Whey protein consists of several biological components, including BCAA which are easily absorbed and used by the body, lactoferrin as a non-enzymatic antioxidant, immunoglobulin as an antibody, β-Lactoglobulin as BCAA that plays a role in retinol-binding and can modulate the lymphatic response in humans, α-Lactalbumin which can increase antibody response to systematic antigen stimulation and suppress T cell response in lymphocytes, lactoperoxidase which can remove harmful bacteria for the human body, glycomacropeptide, and serum albumin.[17]

The high protein and amino acid content in whey protein make it an alternative food source of protein and amino acids with many health benefits. Increased protein intake can lead to changes in circulating lipids and lipoprotein levels, such as decreased LDL, decreased serum triglycerides, and increased HDL concentrations.[18] Research on female Wistar rats showed that whey protein concentrate supplementation could reduce cholesterol levels and serum triglyceride concentrations after consumption for 8 weeks compared to casein supplementation.[19] The decreased lipid profile levels with whey protein consumption occur through the role of BCAA in whey which accelerates digestion and absorption rates in intestines. β-Lactoglobulin content helps the absorption of cholesterol in the intestine, inhibits the expression of genes in charge of the absorption and synthesis of intestinal fatty acids and cholesterol, and increases the excretion of fecal steroids.[20]

Global competition demands a productive workforce for companies. A productive workforce will be created through good nutritional and health status. Some epidemiological studies have shown that nutrient intake is positively associated with labor productivity.[1] The work environment condition is one factor that influences the health and nutritional status of the workforce. A work environment with exposure

to hazardous chemicals such as a mining environment can affect the health status of workers. Coal, one of the largest natural resources in Indonesia, creates many job opportunities in the coal mining environment. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.[2,3] Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. Arsen can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.[4]

LIMITATION

This study did not analyze the levels of Arsenic (As) in the body of the research subjects, so the exact amount of exposure that affects triglyceride levels in the body remains unknown. Further research on this topic still needs to be conducted.

CONCLUSIONS

There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups. Whey protein intervention led to an increase in triglyceride levels in the exposed group and a decrease in triglyceride levels in the unexposed group although only a slight decrease was produced with the whey protein intervention for 28 days.

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REFERENCES

1. Suma'mur. Higiene Perusahaan dan Kesehatan Kerja (Hiperkes). Jakarta: Sagung Seto; 2013.
2. Radi, F. Hasni, M.J. Obesogens As An Environmental Risk Factor For Obesity. Malays J Public Health Med. 2014;14 (3):63–70.
3. Baillie-Hamilton P. Chemical Toxins: A Hypothesis To Explain The Global Obesity Epidemic. J Altern Complement Med. 2002;8:185–92.
4. Ceja-Galicia ZA, Daniel A, Salazar AM, Panico P, Ostrosky-Wegman P, Diaz-Villasenor A. Effects of arsenic on adipocyte metabolism: Is arsenic an obesogen? Elsevier Mol Cell Endocrinol Xxx. 2017;1-8.
5. Marshall K. Therapeutic applications of whey protein. Altern Med Rev. 2004;9:136–156.
6. Furst P, Stehle P. What Are The Essential Elements Needed For The Determination of Amino Acid Requirements in Humans ? J Nutr. 2004;134:1558–65.
7. Kawase M, Hashimoto H, Hosoda M, Morita A, Hasono A. Effect of Administration of Fermented Milk Containing Whey Protein Concentrate to Rats and Healthy Men on Serum Lipids and Blood Pressure. J Diet Sci. 2000; Vol. 83, No. 2.
8. Bhathena S, Velasquez M. Beneficial Role Of Dietary Phytoestrogens In Obesity And Diabetes. Am J Clin Nutr. 2002;76:1191–1201.
9. Vatani, DS, Golzar, FAK. Changes in Antioxidant Status and Cardiovascular Risk Factors of Overweight Young Men After Six Weeks Supplementation of Whey Protein Isolate and Resistance Training. Elsevier Ltd. 2012;59(Appetite):673–8.
10. Shamai L, Lurix E, Shen M, Novaro GM, Szomstein S, Rosenthal R, et al. Association of Body Mass Index and Lipid Profiles: Evaluation of a Broad Spectrum of Body Mass Index Patients Including the Morbidly Obese. 2010;21 (1)(Obesity Surgery):42–47.
11. Hansen SEJ, Madsen CM, Varbo A, Nordestgaard BG. Body mass index, triglycerides and risk of acute pancreatitis: A population-based study of 118,000 individuals. 2019;(The Journal of Clinical Endocrinology&Metabolism.).
12. Silva Rc, Diniz M, Alvim S, Vidigal P, Fedeli L, Barreto S. Physical Activity and Lipid Profile in the ELSA-Brasil Study. 2016;107(1):10–9.
13. Kdekian A, Alssema M, Van Der Beek EM, Greyling A, Vermeer MA, Mela DJ, et al. Impact of isocaloric exchanges of carbohydrate for fat on postprandial glucose, insulin, triglycerides, and free fatty acid responses-a systematic review and meta-analysis. Eur J Clin Nutr. 2019.

14. Lemaire M, Lemarié CA, Flores Molina M, Schiffrin EL, Lehoux S, Mann KK. Exposure to Moderate Arsenic Concentrations Increases Atherosclerosis in ApoE−/− Mouse Model. Oxf Univ Press. 2011;122 (1)(Toxicological Sciences):211–221.

15. Karyadi E. Hidup Bersama Penyakit Hipertensi, Asam Urat, Jantung Koroner. Jakarta: PT. Intisari Mediatama; 2006.

16. Lichtenstein AH. A. H. Diet and Lifestyle Recommendations Revision 2006: A Scientific Statement From the American Heart Association Nutrition Committee. Dalam 2006. Page 82–96. (Circulation; vol. 114 (1)).

17. Anthony JC, Anthony TG, Kimball SR, Jefferson LS. Signaling Pathways Involved In Translational Control Of Protein Synthesis In Skeletal Muscle By Leucine. J Nutr. 2001;131:856S-860S.

18. Pal S, Radavelli-Bagatini S. The Effects Of Whey Protein On Cardiometabolic Risk Factors. Obes Rev. April 2012.

19. Zhang, X., Beynen, A. C. Lowering effect of dietary milk-whey protein v. casein on plasma and liver cholesterol concentrations in rats. 1993;70(British Journal of Nutrition):139.

20. Pal S, Ellis V, Dhaiwal S. Effects of Whey Protein Isolate on Body Composition, Lipids, Insulin and Glucose in Overweight and Obese Individuals. Br J Nutr. 2010;104:716-723.

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The effect of additional protein, phosphatidylcholine, phosphatidylserine, and inulin on S100β levels of acute ischemic stroke patients at Dr. Kariadi Central Hospital, Semarang

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ABSTRACT

Background: The brain releases biochemical substrates, such as S100β protein, into circulation in response to ischemic conditions as a sign of damage in nerve cells and disruption of the blood-brain barrier's integrity. Thrombolytic therapy has led to the development of many neuroprotective therapies such as protein, phosphatidylcholine, phosphatidylserine, and inulin, which can be added to food products. Protein, phospholipids, and inulin, have a neuroprotective impact on nerve cells in the brain and blood-brain barrier.
Objective: To prove the effect of protein, phosphatidylcholine, phosphatidylserine, and inulin on S100β levels and clinical outcomes in patients with acute ischemic stroke.
Materials and Methods: This study was done in a single-blind RCT. Eighteen ischemic stroke patients were randomly divided into nine subjects for the intervention group and nine subjects for the control group. The Control group received 250 ml conventional formula milk (11.8 g protein) 3 times/day. The intervention group received 250 mL commercial milk 3 rimes/day which contained 15 g protein with 128 mg phosphatidylcholine, 32 mg phosphatidylserine, and 3 g inulin. All of the groups were given hospital-standard therapy for ischemic stroke. S100β levels were measured at pre and post-intervention.
Results: Pre and post S100β levels in intervention and the control group did not show any statistically difference (p = 0.777 and p = 0.096), but there was a trend of decreasing levels of S100β in the intervention group (-24.6 ± 252.0 pg/mL) versus control group (135.8 ± 216.2 pg/mL).
Conclusions: The addition of protein, phosphatidylcholine, phosphatidylserine, and inulin did not have a significant effect on S100β levels.

Keyword: Protein; Phosphatidylcholine; Phosphatidylserine; Inulin; S100β; Stroke

BACKGROUND

Ischemic stroke is the most common stroke that occurs when blood vessels in the brain are clogged by plaque/embolism. Clogged blood vessels interfere with blood flow going to the brain, reducing the supply of oxygen and glucose to the brain. This condition could lead to the death of nerve cells in the brain (apoptosis).[1,2] Damaged and dead nerve cells will cause some biochemical substrates to leave the brain into the circulatory system in response to ischemic conditions. This clog could also result in reduced proteins that play a role in maintaining the integrity of the blood-brain barrier membrane, thus the membrane increases its permeability. This, in turn, causes several biochemical substrates that should stay in the components of the brain to move into the circulatory system.[1–6]

Various biochemical substrates released in response to ischemic conditions play important roles in triggering brain tissue damages[6], one of them is S100β protein. This protein helps to regulate intracellular calcium levels[3], where excessive intracellular Ca^{2+} levels will lead to apoptosis (death of nerve cells).[2,5] Excessive intracellular Ca^{2+} caused by the decrease in the supply of blood that carries oxygen and nutrients to the brain, so the brain will lack energy (ATP) to be function normally. The Ca^{2+} ATPase pump which normally picks up Ca^{2+} into the

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organelle becomes inactive due to lack of ATP in the brain, resulting in the accumulation of intracellular Ca^{2+}. Accumulation of intracellular Ca^{2+} will removes Mg^{2+} ions in charge to keep N-methyl-D-aspartate (NMDA) receptors from being active. Activation of the NMDA receptor will cause stimulation of the post-synaptic membrane, where this membrane is 5 times higher in permeability for Ca^{2+}, so the intracellular Ca2+ ion becomes excessive, which will lead to nerve cell apoptosis. Apoptosis will cause the release of S100β protein in brain components to move into the circulation. S100β protein secretion increases along with the response of glial cells due to metabolic disorders such as head injury, damaged blood-brain barrier, and ischemia.[6]

Damage to cells in the brain will also trigger the activation of catabolic hormones that the patient will experience hypercatabolism.[7] Hypercatabolism, if not treated promptly, will worsen the patient's nutritional status.[7] Also, stroke patients also experience intestinal dysbiosis through immunological pathways.[8–11] Dysbiosis is a condition in which qualitative and quantitative changes occur in the composition, distribution, and metabolic activity of microbes in the intestines, causing adverse effects and worsening the patient's clinical outcome.

Appropriate therapy is the key in ischemic stroke patients.[12,13] One of the pharmacological therapies given is thrombolytic therapy (rTPa).[14] This therapy cannot be given to all acute ischemic stroke patients due to very strict indication criteria, especially in terms of duration (time window). The best timeframe for the administration of this therapy, which can provide the benefit of brain functional improvement and can reduce mortality, is <3 hours and ranged between 3-4.5 hours after symptom onset.[15,16] If the therapy is given not according to the guidelines, it can cause side effects such as the risk of bleeding in the brain and gastrointestinal tract, and angioedema.[14]

This situation has led to the development of many neuroprotective therapies, namely the addition of phospholipids which can be given directly in the form of supplements (cytolin) or fortified in food products such as milk. Phospholipids are a type of fat found in many nerve cell membranes. There are several types of phospholipids, the common ones are phosphatidylcholine and phosphatidylserine. Phosphatidylcholine increases the biosynthesis of membrane phospholipids which is degraded by an increase in free radicals during brain ischemia (neuroprotection).[12,17] Phosphatidylcholine also inhibits the activation of enzymes that trigger apoptosis of nerve cells,[12,17] thus affecting S100β levels. Also, the protein contained in milk will increase protein intake, muscle mass, and possibly increase body mass index (BMI). Therefore, it can improve the patient's motoric function and prevent the patient from experiencing a decrease in nutritional status.[18–20]

One of the nutrients that need to be given to ischemic stroke patients is inulin. Studies regarding the effect of inulin in improving ischemic conditions are still very limited in ischemic stroke patients and there are no studies about the direct effect of inulin on blood biomarkers of brain damage such as S100β protein. So far research has proven that Short-Chain Fatty Acid (SCFAs) acts as neuroprotective agents in the nerve cells. Inulin will be quickly fermented by *Bifidobacteria* and *Lactobacilli* (probiotic bacteria) and will produce SCFA in the form of acetic acid, propionic, butyrate, L-lactate, CO2, and hydrogen as fermentation products.[21,22] SCFA can be used as the source of energy (ATP) by the central nervous system because long-chain fatty acids cannot cross the blood-brain barrier membrane. SCFA has been proved to play a neuroprotective role, synthesizing neurotransmitters, and modulating the immune system. Thus, SCFA can improve dysbiotic conditions due to ischemic stroke.[23–25]

Dr. Kariadi Central Hospital is an "A-Accredited" Hospital that has a Stroke Unit with an average of two new stroke patients every day. The number of stroke patients in this hospital continues to increase.[26] This research aimed to prove the effect of additional protein, phosphatidylcholine, phosphatidylserine, and inulin on S100β levels and clinical outcomes of acute ischemic stroke patients at Dr. Kariadi Central Hospital, Semarang.

MATERIALS AND METHODS

A randomized control trial with single-blind was used as the design of this research. The effect of additional protein, phosphatidylcholine, phosphatidylserine, and inulin was seen through S100β levels. The research subjects were acute ischemic stroke patients in the inpatient room of Rajawali 1A of Dr. Kariadi Central Hospital Semarang amounted to 18 people (control = 9, intervention = 9), 10 subjects were male and 8 subjects were female. The research lasted for 4 months (February-May 2020). Acute ischemic stroke patients were selected to participate if their attack onsets were <72 hours and aged > 18 years.

Also, the researchers had randomized the subjects through Consecutive Sampling and provided informed consent to the patient/patient's family. During the implementation of the research, 7 patients dropped out or lost to follow-up with the following details: 4 patients not domiciled in Semarang, 2 patients died during the intervention, and 1 patient was forced to move out from hospital because of cost limitation.

The physical and neurological examinations had been performed by the doctor in charge of treating those patients. Supporting data, such as weight, blood pressure, blood glucose, lipid profile, smoking history, rTPa therapy status, and history of recurrent stroke had been taken from the patients' medical records once when the patient entered the stroke ward only to determine the risk factor data such as obesity, hypertension, diabetes mellitus, dyslipidemia, and smoking history. The researchers assessed the subject's dietary intake of energy, carbohydrate, protein, fat, phosphatidylcholine, phosphatidylserine, and inulin every day using the 24 hours recall method. The intake data consumed by the subjects will be compared with the patient's requirement to obtain the adequacy of nutritional intake which is expressed in percentage (%) with the following formula:

$$\text{Adequacy intake:} \frac{nutrient\ intake}{nutrient\ need} \text{x } 100\%$$

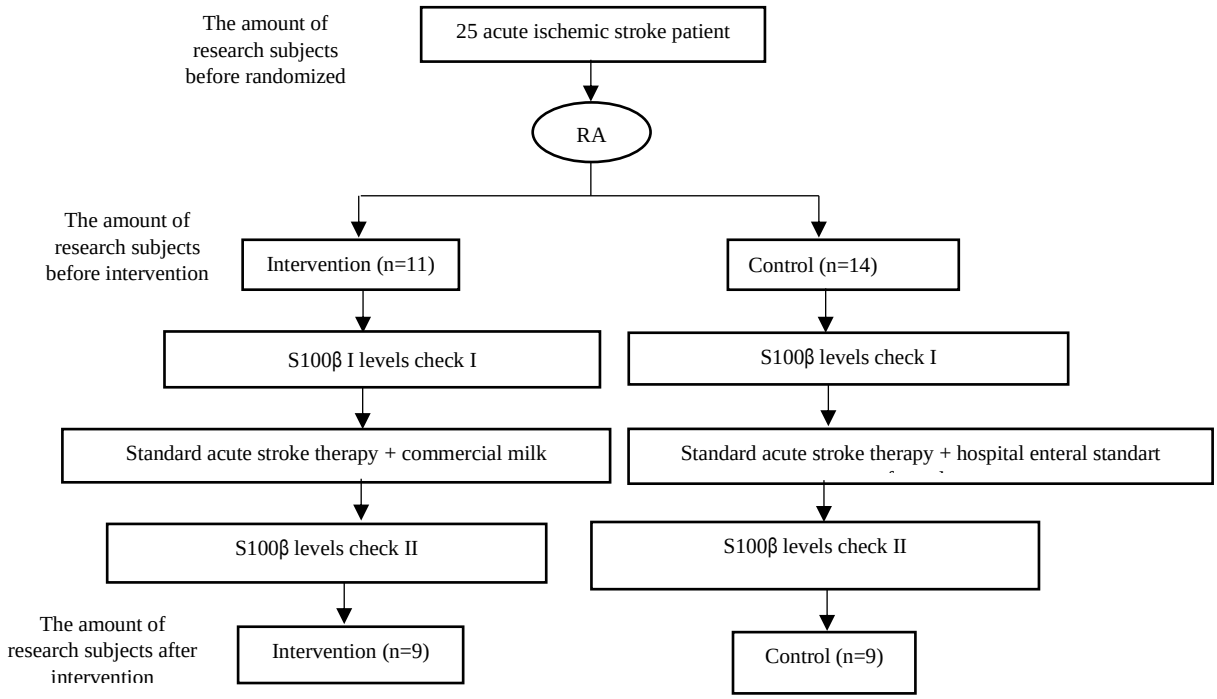

Figure 1. The Flow Chart of the Amount of Research Subjects (Start to Finish)

The nutritional requirement has been adjusted to the nutritional requirement for ischemic stroke patients: 30 kcal/kg BW for energy, 60% TEE for carbohydrate, 20% TEE for protein, 20% TEE for fat, 1000 mg/day for phosphatidylcholine, 100 mg/day for phosphatidylserine, and 12 mg/day for inulin. All data collection has been recorded on the research sheets that had been prepared by researcher.

The control group was given milk based on the enteral standard formula from the hospital that contains 11.8 g protein (from hospital database) without the addition of phosphatidylcholine, phosphatidylserine, and inulin as much as 250 ml.

The intervention group was given 69 g of commercial milk powder which contains higher protein than the control group (15 g) with the addition of 128 mg phosphatidylcholine, 32 mg phosphatidylcholine, and 3 g inulin. The commercial milk given to the intervention group contains a moderate glycemic index, therefore, it is necessary to adjust the dose and the duration of the intervention for subjects with diabetes. In the intervention group, subjects with diabetes mellitus received 34.5 g of commercial milk powder (half the original dose) so the duration of the treatment becomes longer (14 days). Researchers cannot exclude subjects with

diabetes because of the limited number of study subjects. All the treatment was given 6 times a day during the acute phase (1-3 days according to the patient's swallowing ability) by enteral tube and then given orally 3 times a day during the recovery phase until the end of treatment (day 7). During the administration of the intervention, the researchers observed functional and biochemical changes as well as possible side effects.

Whole blood vein samples were collected from the mediana cubiti vein. A sampling of 6 mL was carried out using a red *vacutainer* (blood clot without the addition of EDTA). The duration between blood sampling to storing should not exceed 8 hours. Blood samples were sent to the central laboratory of Dr. Kariadi Central Hospital Semarang for centrifugation and stored in a deep freezer (-80ºC) until the number of research, samples were met. After meeting the sample size, blood serums were sent to the GAKI Laboratory of the RSND-FK UNDIP. Serum S100β levels were checked using the Human S100β ELISA (Enzyme-Linked Immunosorbent Assay) kit Elabscience with catalog number E-EL-H1297. The inspection method was following the manual found in the kit. The analysis of serum S100β levels has been carried out twice; when the patients were admitted to Dr. Kariadi Hospital Semarang before given intervention (day 1) and after given intervention (day 7). This research has received Ethical Clearance approval by the Medical Research Ethics Commission of FK UNDIP / RSDK with registration No. 479 / EC / KEPK-RSDK / 2020.

Statistical analysis was performed using SPSS version 22.0 for windows. Data with categorical scales were expressed in the distribution of frequencies and proportions. The numerical scale data [levels of S100β and changes (Δ) of S100β] were tested for normality and then tested using Paired T-test and Independent T-Test. Also, different levels of S100β and changes (Δ) of S100β in the DM and Non-DM subgroups have been carried out using Paired T-Test/ Independent T-test for normally distributed data and Willcoxon/ Mann Whitney for data that were not normally distributed.

RESULTS

Research Subject Characteristics

There was no significant difference in the category of age (p = 0.347), gender (p = 0.637), mean BMI (p = 0.468), obesity status (p = 1.000), hypertension (p = 0.576), diabetes mellitus (p = 1.000), dyslipidemia (p = 1.000), smoking status (p = 1.000), rt-PA therapy (p = 1.000), inpatient duration (p = 0.637), intervention duration (p = 0.206), and history of recurrent stroke (p = 1.000).

Adequacy Level of Energy, Protein, Fat, and Carbohydrate Intake

The adequacy level was obtained from the total intake per day compared to the actual needs and was presented in percentage. Table 2 shows that there were significant differences in the mean level of adequacy of energy intake (p = 0.008), protein (p = 0.002), carbohydrate (p = 0.002), phosphatidylcholine (p=0.000), phosphatidylserine (p=0.000), and inulin (p=0.000) however there was no significant difference in the mean level of adequacy of fat intake between groups (p = 0.912). For the control group, all adequate levels of nutrient intake were deficits, whereas in the intervention group only adequate levels of intake of fat and phosphatidylcholine were deficits.

Serum S100β Levels Distribution Before and After Interventions

The results of statistical tests showed that there was no significant difference in the distribution of serum S100β levels both before (p = 1.000) and after the intervention (p = 0.576) between the intervention group and the control group. The serum S100β level category <236.7 pg/mL indicates that the patients tended to have a good improvement in clinical outcome. Serum S100β levels> 236.7 pg/mL are associated with poor clinical outcomes and even death. Based on the aforementioned results, one subject in the intervention group before treatment had a high-risk S100β level category, but it went down to the low-risk category after the treatment compared to one subject in the control group who had a low-risk S100β level category before treatment and went up to being categorized as high risk after the treatment. This shows that there was a trend of improvement in S100β levels in the intervention group compared to the control group.

Differences in S100β levels and changes (Δ) of S100β levels

The mean serum S100β levels in baseline of the control and intervention groups (358.8 $\pm$ 215.3 and 512.3 $\pm$ 343.9 pg / mL, respectively) were not statistically significant (p = 0.273). This illustrates that the condition of the two groups before the intervention was homogeneous. In table 4, it is shown that there was a trend of decreasing mean serum S100β levels in the intervention group from 512.3 $\pm$ 343.9 pg / mL to 487.7 $\pm$ 366.8 pg / mL, while in the

control group there was an increasing trend in the mean serum S100β level from 358.8 ± 215.3 pg / mL to 494.6 ± 296.4 pg / mL. Also, the pre and post S100β levels in both the intervention and control groups were not statistically significant (p = 0.777 and p = 0.096, respectively). There was no significant difference in the mean change (Δ) of serum S100β levels in the intervention group (-24.6 ± 252.0 pg / mL) and control group (135.8 ± 216.2 pg / mL), but in the intervention group, the tendency of decline was better than the control group although it was not statistically significant (p = 0.166).

Table 1. The Characteristics of Research Subjects in Research Groups

Characteristic	Group		*p*
	Intervention (n=9)	Control (n=9)	
Gender			
- Male	4 (44.4%)	6 (66.7%)	0.637[b]
- Female	5 (55.6%)	3 (33.3%)	
Mean age (year)	50.6 ± 14.8	62.6 ± 8.8	0.053[a]
Age category			
- >60	3(33.3%)	6(66.7%)	0.347[b]
- <60	6(66.7%)	3(33.3%)	
Mean BMI (kg/m^2)	25.3 ± 4.1	24.1 ± 2.9	0.468[a]
Obesity Status			
- Yes (BMI ≥ 25 kg/m^2)	4 (44.4%)	3 (33.3%)	1.000[b]
- No (BMI < 25 kg/m^2)	5 (55.6%)	6 (66.7%)	
Hipertension			
- Yes (BP ≥ 130/80 mmHg)	8 (88.9%)	6 (66.7%)	0.576[b]
- No (BP < 130/80 mmHg)	1 (11.1%)	3 (33.3%)	
Diabetes Mellitus			
- Yes	3 (33.3%)	4 (44.4%)	1.000[b]
- No	6 (66.7%)	5 (55.6%)	
Dislipidemia			
- Yes	7 (77.8%)	8 (88.9%)	1.000[b]
- No	2 (22.2%)	1 (11.1%)	
Smoking Status			
- Yes	5 (55.6%)	6 (66.7%)	1.000[b]
- No	4 (44.4%)	3 (55.6%)	
rt-PA Therapy			
- Yes	2(22.2%)	1(11.1%)	1.000[b]
- No	7(77.8%)	8(88.9%)	
Inpatient Duration			
- <7 days	6(66.7%)	4(44.4%)	0.637[b]
- ≥7 days	3(33.3%)	5(55.6%)	
Intervention Duration			
- 7 days	6(66.7%)	9(100%)	0.206[b]
- 14 days	3(33.3%)	0(0%)	
History of Recurrent Stroke			
- Yes	3(33.3%)	3(33.3%)	1.000[b]
- No	6(66.7%)	6(66.7%)	

[a] Independent T-Test; [b] Fisher-Exact Test
The values in table are: mean ± SD; n (%) Percentage shown in columns

Table 2. The Mean Adequacy Intake of Energy, Protein, Fat, and Carbohydrate between Groups

Adequacy Levels	Intervention Mean±SD	Control Mean±SD	*p*
Energy (%)	95.1±18.4	73.8±10.1	0.008[a]
Carbohydrate (%)	107.0±20.6	76.6±12.2	0.002[a]
Protein (%)	94.2±17.6	68.5±10.9	0.002[a]
Fat (%)	74.3±21.2	73.4±11.2	0.912[a]
Fosfatidilkolin (%)	72.7±8.1	25.3±3.4	0.000[a]
Fosfatidilserin (%)	112.4±12.3	48.7±10.3	0.000[a]
Inulin (%)	92.4±8.0	24.4±7.3	0.000[a]

[a] Independent T-Test

Table 3. The Distribution of S100β Levels before Intervention in Research Groups (n=18)

	Category of Serum S100β Levels (pg/mL)	Group		*p*
		Intervention (n=9)	Control (n=9)	
Before Intervention	Low risk (<236.7)	2(22.2%)	2(22.2%)	1.000[b]
	High risk (≥236.7)	7(77.8%)	7(77.8%)	
After Intervention	Low risk (<236.7)	3(33.3%)	1(11.1%)	0.576[b]
	High risk (≥236.7)	6(66.7%)	8(88.9%)	

[b]Fisher Exact Test

Table 4. The Difference in Serum S100β Levels (pg/mL)

Group	n	Before Mean±SD	After Mean±SD	*P*	Δ Mean±SD
Intervention	9	512.3±343.9	487.7±366.8	0.777[c]	-24.6±252.0
Control	9	358.8±215.3	494.6±296.4	0.096[c]	135.8±216.2
p		0.273[a]	0.965[a]		0.166[a]

[a]Independent T-Test; [c] *Paired T-Test*

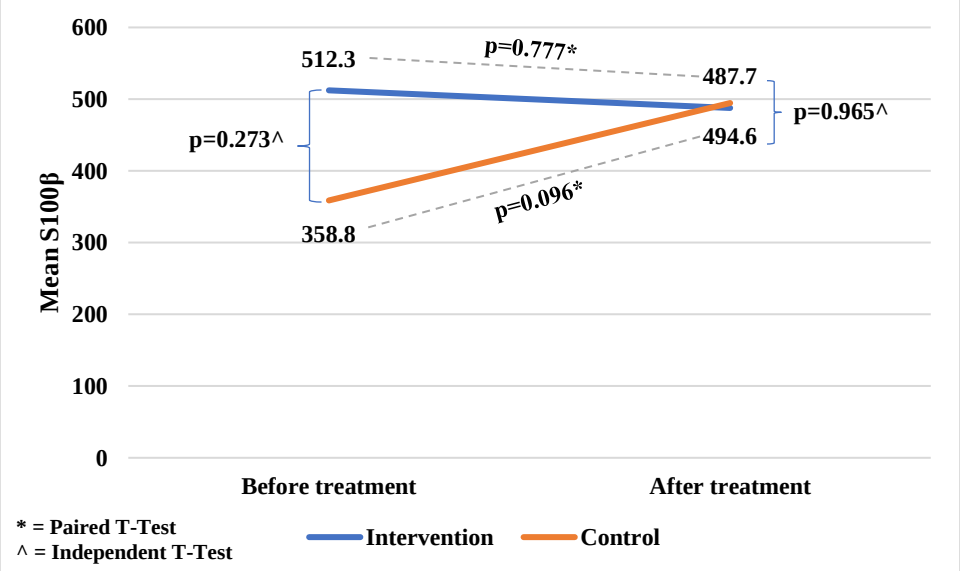

Figure 2. The Serum S100β Levels before and after Intervention

Sub-group analysis of S100β levels

In DM and non-DM sub-groups, the median serum S100β levels before treatment between the intervention and control groups were not statistically significant (p = 0.480 and p = 0.251, respectively). This illustrates that the condition of the two sub-groups before the intervention was homogeneous. There was no significant difference in the median

levels of S100β before and after the intervention in both DM (p intervention = 0.285; p control = 0.272) and non-DM sub-group (p intervention = 0.484; p control = 0.308).

In DM and Non-DM sub-groups, there was a trend of decreasing S100β levels in a better direction for the intervention group and the decrease (Δ) in the DM sub-group was bigger than Non-DM sub-group compared to the control group where there was an increasing trend (Δ) in S100β levels in DM and Non-DM sub-group and the increase (Δ) in DM sub-group was bigger than Non-DM sub-group.

DISCUSSION

The data of research subjects showed no significant difference in mean BMI, mean age, age category, gender, obesity, hypertension, diabetes mellitus, dyslipidemia, smoking status, rt-PA therapy, inpatient duration, intervention duration, and history of recurrent stroke (p>0.05).

Table 5. The *Sub Group* Analysis of S100β Levels between Groups

Sub Grup	Kelompok	n	Sebelum Median(Min-Max)	Sesudah Median(Min-Max)	*p*	Δ Median(Min-Max)
DM	Intervention	3	971.9(115.5-977.9)	521.8(146.6-771.8)	0.285[c]	-200.1(-456.1 – 31.1)
	Control	4	428.8(252.1-794.2)	641.0(253.4-1015.9)	0.272[d]	117.8(-83.1 – 494.9)
	P		0.480[e]	0.589[a]		0.102[a]
Non-DM	Intervention	6	475.1(28.5-660.7)	392.7(25.6-1004.8)	0.484[c]	-39.9(-117.5 – 344.5)
	Control	5	255.9(107.1-426.5)	417.8(187.7-610.9)	0.308[c]	75.0(-178.8 – 355.0)
	P		0.251[a]	0.593[a]		0.584[e]

[a] Independent T-Test; [c] Paired T-Test; [d] Willcoxon Test; [e] Mann Whitney Test

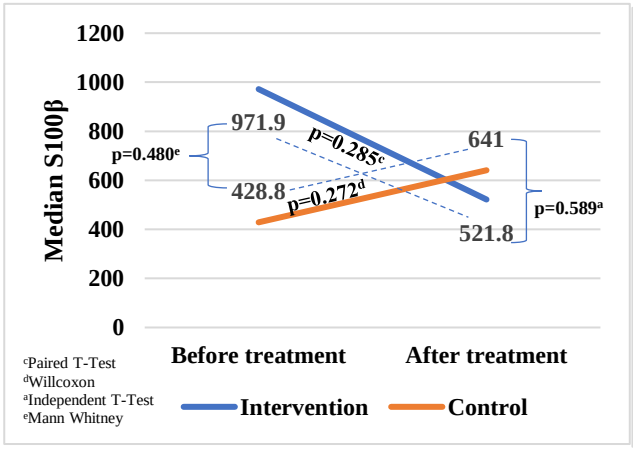

Figure 3. The S100β Levels before and after intervention in DM Sub Group

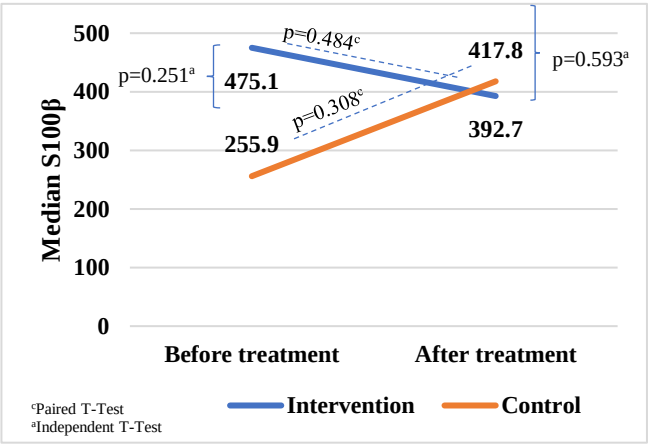

Figure 4. The S100β Levels before and after intervention in Non-DM Sub Group

Stroke does not only occur in the elderly, but also occurs in those at productive age under 45 years, and there are some cases where the sufferers are under 30 years old.[27,28] In this study, 2 subjects were under 30 years old. The habit of consuming food containing high fat and smoking has been shown to influence the incidence of ischemic stroke at an age less than 45 years.[29] People who consume high-fat foods ≥3 times/week have a 3.744 times more risk of suffering from ischemic stroke compared to people who consume high-fat foods <3 times/week.[29] High-fat food sources when consumed excessively can increase cholesterol levels in the blood. If the cholesterol level in the blood is abnormal (≥ 200 mg / dL) it will increase the accumulation of fat on the walls of blood vessels, causing narrowing of the blood vessels and disrupting the blood supply to the brain.[30] Cholesterol can form blood clots and plaques that clog the arteries and eventually cut off the blood flow to the heart and brain. This can lead to heart attacks and strokes.[31] Smoking is associated with a hypercoagulable state. It is characterized by an increase in thromboxane release which causes increased platelet activation and degeneration of the vascular endothelium, which promotes the formation of thrombus-producing plaque. Nicotine has no direct

effect on this mechanism, but it is possible that the burning of cigarette smoke causes thromboembolic formation, leading to ischemic stroke.[32]

The data on the adequacy of nutritional intake showed that there were significant differences in levels of the adequacy of energy, carbohydrate, and protein between groups ($p < 0.05$). There was no difference in the level of adequacy of fat between groups ($p > 0.05$). The nutritional content of commercial milk given to the intervention group had higher energy, carbohydrate, and protein content (Energy = 289.8 kcal, Protein = 15.18 grams, KH = 44.16 grams per 250 mL) compared to milk given to the control group (Energy = 222.2 - 246.4 kcal, Protein = 11 - 11.8 grams, CH = 28 - 29.2 grams per 250 mL), while milk given to the control group had higher fat content (9 gram / 250 mL) compared to the milk given to the intervention group (5.52 g / 250 mL). This was also influenced by nutritional requirement acceptance of each respondent, method of administration (enteral tube for acute phase and then orally for recovery phase), the role of medical team, especially nurses, and family members in motivating patients to eat, as well as the length of hospitalization of patients.

Also, there were six subjects (66.7%) in the intervention group and four subjects (44.4%) in the control group who returned home before the 7th day. Patients in both control and intervention groups who returned home before the 7th day still had poor appetite, but patients in the intervention group have received the commercial milk to be consumed at home while the control group did not. This led to a higher level of energy, protein, and carbohydrate intake in the intervention group even though the patient did not have a good appetite. Also, patients who returned home before the 7^{th} day tended to have an increase in fat intake based on the results of the recall of intake by telephone. They tended to eat food that contains high fat rather than at the hospital, where the menu has been adjusted with the requirements and conditions of each respondent.

The results of statistical tests showed that there was no significant difference in the mean serum S100β levels between before and after the intervention or between groups. The causes of this insignificant difference were multifactorial, one of which was that the subjects in this research have different comorbidities such as hypertension, diabetes mellitus, obesity, and dyslipidemia. According to research, S100β levels in subjects who have comorbid like diabetes mellitus, hypertension, central obesity, and dyslipidemia will tend to be increased.[33,34] Two subjects in the intervention group and one subject in the control group in this research received rt-PA therapy. Some research proved the effectiveness of citicoline as a neuroprotective therapy, showing that rt-PA could improve blood flow in the penumbra area. This leads to becoming soluble and having difficulty improving its performance due to the effects of thrombolysis. Also, the higher the severity of the stroke will affect the performance of citicoline because the penumbra areas that are damaged are not fully reached by citicoline.[12,35] The results of the difference test (Δ) S100β levels in this research were not statistically significant ($p = 0.166$) between groups. This shows that rt-PA therapy does not affect the intervention given. Another research also stated that rt-PA did not significantly affect S100β levels.[36] The reason was that the response of each individual to rt-PA therapy was different. rt-PA therapy can exert a significant neuroprotective effect only when thrombolysis and reperfusion are achieved earlier, whereas in cases where reperfusion is not immediately achieved there may be side effects such as blood-brain barrier (BBB) disruption and hemorrhagic transformation.[36]

A study shows that there is a positive and significant correlation between age and S100β levels in study subjects with mild TBI (Traumatic Brain Injury). The study reported that the subjects aged >65 years had higher levels of S100β (> 0.10 μg / L) compared to subjects aged <65 years. This is because older subjects usually have chronic diseases or neurological diseases such as Alzheimer's and Parkinson's, but further research is needed to explain this association.[37] In this study, the mean age of subjects in the control group was older (62.6 ± 8.8) than in the intervention group (50.6 ± 14.8). This is one of the possible causes of S100β levels in the control group which tended to increase compared to the intervention group.

Research on the effect of giving protein, neuroprotective substances (phosphatidylcholine and phosphatidylserine), and inulin on brain damage markers, especially S100β, is still very limited. In this research, the interventions were administered orally in the form of powdered milk containing neuroprotectants (phosphatidylcholine and phosphatidylserine) and prebiotic inulin. In oral administration, phosphatidylcholine and phosphatidylserine are almost completely absorbed efficiently. Once absorbed, phosphatidylcholine and phosphatidylserine spread widely throughout the body through the blood-brain barrier and reach the central nervous system (CNS).[38,39] Inulin is soluble in

water and difficult to be hydrolyzed by enzymes in the digestive tract, thus it can reach the large intestine mostly intact. In the large intestine, inulin will be fermented by probiotic bacteria into short-chain fatty acids (SCFA) and some specific microflora that produces lactic acid.[40]

The results of different tests for S100β levels were not statistically significant, but in the intervention group, there was a trend of decreasing S100β levels compared to the control group. According to research, a high-protein diet given from the beginning provides regulation at the molecular level, which helps to increase SOD expression, suppresses MDA expression, and restores iNOS expression.[41] This suggests that a high-protein diet given from the beginning can inhibit oxidative stress, thereby helping the body to eliminate free radicals that play a role in the pathophysiological process of stroke.[41]

Vitamin B supplementation (folic acid, riboflavin, vitamin B12) and choline are effective in improving stroke recovery by suppressing the transcription factor p53 mechanism which causes apoptosis (death of nerve cells) where p53 activation is triggered by an increase in homocysteine. Also, there is an increase in neuroplasticity through increased BDNF (Brain-derived neurotrophic factor) expression and activation of anti-oxidative mechanisms characterized by increased levels of the transcription factor Nrf2 (nuclear factor erythroid 2-related factor 2) and SOD2 (Superoxide dismutase 2) enzyme. This means an increase in the flexibility of cells to manage oxidative damage.[42]

Several kinds of research on the effects of phosphatidylserine supplementation in experimental animals and humans have been carried out on the improvement of cognitive and memory functions by largely unexplainable mechanisms. In essence, phosphatidylserine plays a role in the activation of signaling proteins and receptors that are important for neuron survival, differentiation, and regulation of neurotransmitters in the signaling process.[39]

Inulin is an example of a prebiotic. Prebiotics are components of food ingredients that the digestive tract cannot enzymatically digest. Thus, they will be fermented by microbiota in the large intestine. Inulin acts as a substrate to increase the diversity of beneficial microbiota in the intestine, increasing the production of SCFA (acetate, propionate, and butyrate) as a result of anaerobic fermentation.[43] SCFA can affect the gut-brain axis and brain function directly or indirectly. SCFAs are absorbed by colonic cells via H^+-dependent monocarboxylate transporter (MCT) or Na^+-dependent monocarboxylate transporter (SMCTs). SCFA then binds to G protein-coupled receptors (GPCRs) such as FFAR2, FFAR3, GPR109a / HCAR2 (hydrocarboxylic acid receptors), and GPR164 by inhibiting histone deacetylation that will affect intestinal mucosal immunity and intestinal barrier integrity. SCFA interaction with some of these receptors on enteroendocrine cells will cause indirect signaling to the brain through the systemic circulation by inducing the secretion of intestinal hormones such as glucagon-like peptide 1 (GLP1), peptide YY (PYY), γ-aminobutyric acid (GABA), and serotonin (5-HT). Colon-generated SCFAs reaches the systemic circulation and other tissues leading to activation of brown adipose tissue, regulation of mitochondrial function in the liver, increased insulin secretion by β-pancreatic cells, and energy homeostasis throughout the body. Systemically, SCFAs influence systemic inflammation by inducing differentiation of T regulatory (Treg) cells and regulating interleukin secretion. SCFA can cross the blood-brain barrier via monocarboxylate transporter (MCT) located on endothelial cells and improve the integrity of the blood-brain barrier by increasing the expression of tight junction proteins. Furthermore, in the central nervous system (CNS), SCFA fixes inflammation in the nerves by improving glial cell function and morphology, modulating levels of neurotrophic factors, increasing neurogenesis, contributing to serotonin biosynthesis, and increasing the function and homeostasis of nerve cells. SCFA interactions with the gut-brain axis can directly or indirectly affect the emotion, cognition, and pathophysiology of brain disorders.[44]

Since researchers can not exclude subjects with diabetes mellitus, then subgroup analysis was done. The results of DM and Non-DM sub-groups analysis showed that in DM and Non-DM sub-groups there was a trend of decreasing S100β levels in a better direction for the intervention group and a decrease (Δ) in the DM subgroup was bigger than Non-DM subgroup compared to a control group where there were a trend of increasing (Δ) levels of S100β in the DM and Non-DM subgroups and an increase (Δ) in DM subgroup was bigger than Non-DM subgroup. This reason was that in the DM subgroup the intervention duration was different from the Non-DM subgroup, especially in the intervention group. Milk given to the intervention group had a rather high glycemic index, so it was necessary to adjust the daily dose in patients with comorbid DM. This resulted in the intervention duration that was originally given for 7 days stretched to 14 days.

According to research, the pattern of S100β levels will increase from 0-2 hours of onset to 24-48 hours of onset (peak level) then S100β levels will decrease up to 2-3 weeks after onset.[45] This causes a decrease in (Δ) levels of S100β bigger in the DM subgroup compared to the Non-DM sub-group.

The limitation of this research was the number of samples which was 18 subjects (intervention = 9; control = 9). Dr. Kariadi Central Hospital is a referral hospital for COVID-19 patients, so the hospital limits the number of patients at the polyclinic every day. Also, many people are afraid of going to the hospital. Another reason might be the dose and duration of intervention. In some research, the minimum duration of 14 days[38,41,43,46,47] and a minimum dose that can provide a better clinical outcome is 1000 mg/day for phosphatidylcholine.[48,49] In this study, the mean phosphatidylcholine intake in the intervention group is still a deficit. The subjects receive the treatment by enteral tube in the acute phase (1-3 days after stroke onset) followed by oral administration. For administration through the enteral tube, it will be easier to ensure that the treatment is completed, whereas when given orally, some patients do not cooperate due to several factors such as the imperfect swallowing ability and the level of consciousness is still weak. Also, there were subjects in both the control and intervention group who returned home before the 7th day of treatment because some patients ask to continue the recovery phase at home after they have passed the acute period. This has to lead the researchers to recall those subjects regarding their intake by telephone. As a result, patients tend to forget about what they have consumed, also the (household size used by those subjects was different so that the recall results could be underestimating/overestimate. In the hospital, portion standards are already specified for each type of diet.

CONCLUSIONS

The addition of 15 g protein, 128 mg phosphatidylcholine, 32 mg phosphatidylserine, and 3 g inulin, three times a day, for seven days, in patients with acute ischemic stroke does not have a significant effect on S100β levels statistically, however, there is a decreasing trend of S100β levels for the intervention group.

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REFERENCES

1. Xing C, Arai K, Lo EH, Hommel M. Pathophysiologic cascades in ischemic stroke. Int J Stroke. 2012;7(5):378–85.
2. Guo Y, Li P, Guo Q, Shang K, Yan D, Du S, et al. Pathophysiology and biomarkers in acute ischemic stroke - A review. Trop J Pharm Res. 2013;12(6):1097–105.
3. Rezaei O, Pakdaman H, Gharehgozli K, Simani L, Vahedian-Azimi A, Asaadi S, et al. S100 B: A new concept in neurocritical care. Iran J Neurol. 2017;16(2):83–9.
4. Secades JJ. Citicoline: pharmacological and clinical review, 2016 update. Rev Neurol. 2016;63(S03): S1–73.
5. Álvarez-Sabín J, Román GC. The role of citicoline in neuroprotection and neuro repair in ischemic stroke. Brain Sci. 2013;3:1395–414.
6. Kadry H, Noorani B, Cucullo L. A blood-brain barrier overview on structure, function, impairment, and biomarkers of integrity. Fluids Barriers CNS [Internet]. 2020;17(1):1–24. Available from: https://doi.org/10.1186/s12987-020-00230-3
7. Kasim VN, Pateda SM, Hadju V, Jafar N. Suplementasi ekstrak albumin ikan gabus terhadap status gizi dan imunitas pasien stroke. J Gizi Klin Indones. 2017;13(3):91.
8. Singh V, Roth S, Llovera G, Sadler R, Garzetti D, Stecher B, et al. Microbiota dysbiosis controls the neuroinflammatory response after stroke. J Neurosci. 2016;36(28):7428–40.
9. Hennezel E, Abubucker S, Murphy LO, Cullen TW. Total Lipopolysaccharide from the. MBio. 2017;2(6):1–12.
10. Hotamisligil GS. Inflammation and metabolic disorders. 2019;7121(December):1–17.
11. Li H, Li H, Zhang X, Zhang X, Pan D, Pan D, et al. Dysbiosis characteristics of gut microbiota in cerebral infarction patients. Transl Neurosci. 2020;11(1):124–33.
12. Overgaard K. The effects of citicoline on acute ischemic stroke: A review. J Stroke Cerebrovasc Dis. 2014;23(7):1764–9.
13. Adibhatla RM. Citicoline in stroke and TBI clinical trials. Nat Rev Neurol. 2013;9(3):173.
14. Presley B. Penatalaksanaan Farmakologi Stroke Iskemik Akut. Buletin Rasional [Internet]. 2013;12(1):6–8. Available from:

http://repository.ubaya.ac.id/21378/1/Rasional Vol 12 No 1.pdf
15. Lansberg MG, O'Donnell MJ, Khatri P, Lang ES, Nguyen-Huynh MN, Schwartz NE, et al. Antithrombotic and thrombolytic therapy for ischemic stroke: Antithrombotic Therapy and Prevention of Thrombosis, 9th ed: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines. Chest. 2012;141(2 Suppl):e601S-e636S.
16. Jauch EC, Saver JL, Adams HP, Bruno A, Connors JJB, Demaerschalk BM, et al. Guidelines for the early management of patients with acute ischemic stroke: A guideline for healthcare professionals from the American Heart Association/American Stroke Association. Stroke. 2013;44(3):870–947.
17. Sahota P, Savitz SI. Investigational Therapies for Ischemic Stroke: Neuroprotection and Neuro recovery. Neurotherapeutics. 2011;8(3):434–51.
18. Zanini B, Simonetto A, Zubani M, Castellano M, Gilioli G. The effects of cow-milk protein supplementation in the elderly population: Systematic review and narrative synthesis. Nutrients. 2020;12(9):1–26.
19. Patel S. Functional food relevance of whey protein: A review of recent findings and scopes ahead. J Funct Foods [Internet]. 2015;19:308–19. Available from: http://dx.doi.org/10.1016/j.jff.2015.09.040
20. Mitchell CJ, McGregor RA, D'Souza RF, Thorstensen EB, Markworth JF, Fanning AC, et al. Consumption of milk protein or whey protein results in a similar increase in muscle protein synthesis in middle-aged men. Nutrients. 2015;7(10):8685–99.
21. Birkeland E, Gharagozlian S, Birkeland KI, Valeur J, Måge I, Rud I, et al. Prebiotic effect of inulin-type fructans on faecal microbiota and short-chain fatty acids in type 2 diabetes: a randomized controlled trial. Eur J Nutr [Internet]. 2020;59(7):3325–38. Available from: https://doi.org/10.1007/s00394-020-02282-5
22. Shoaib M, Shehzad A, Omar M, Rakha A, Raza H, Sharif HR, et al. Inulin: Properties, health benefits, and food applications. Carbohydr Polym [Internet]. 2016;147(October 2017):444–54. Available from: http://dx.doi.org/10.1016/j.carbpol.2016.04.020
23. Sherwin E, Sandhu K V., Dinan TG, Cryan JF. May the Force Be With You: The Light and Dark Sides of the Microbiota–Gut-Brain Axis in Neuropsychiatry. CNS Drugs. 2016;30(11):1019–41.
24. Braniste V, Al-Asmakh M, Kowal C, Anuar F, Abbaspour A, Tóth M, et al. The gut microbiota influences blood-brain barrier permeability in mice. Sci Transl Med. 2014;6(263):1–12.
25. Erny D, De Angelis ALH, Jaitin D, Wieghofer P, Staszewski O, David E, et al. Host microbiota constantly control maturation and function of microglia in the CNS. Nat Neurosci. 2015;18(7):965–77.
26. Arbayanto DN. Penyembuhan Stroke Perlu Kecepatan Waktu [Internet]. Ruang Publik. 2018. Available from: rri.co.id
27. Ghani L, Mihardja LK, Delima. Faktor Risiko Dominan Penderita Stroke di Indonesia. Bul Penelit Kesehat. 2016;44(1):49–58.
28. Laily SR. Hubungan Karakteristik Penderita dan Hipertensi dengan Kejadian Stroke Iskemik. J Berk Epidemologi. 2017;5(1):95–106.
29. Iskandar A, Hadisaputro S, Pudjonarko D, Suhartono, Pramukarso DT. Gaya Hidup Yang Berpengaruh Terhadap Kejadian Stroke Iskemik Pada Usia Kurang Dari 45 Tahun (Studi Di BLUD RSUD Cut Nyak Dhien Meulaboh Kabupaten Aceh Barat Provinsi Aceh). J Epidemiol Kesehat Komunitas. 2018;3(2):54–62.
30. Junaidi I. Stroke : Waspadai Ancamannya. Yogyakarta: Andi; 2011.
31. Mohr J, Wolf P, Moskowitz M, Mayberg M, Kummer R Von. Stroke: Pathophysiology, Diagnosis, and Management. 5th ed. Grotta J, editor. Philadelphia: Elsevier Saunders; 2011. 198–218 p.
32. Matsuo R, Ago T, Kiyuna F, Sato N, Nakamura K, Kuroda J, et al. Smoking status and functional outcomes after acute ischemic stroke. Stroke. 2020;846–52.
33. Lotosh NG, Savel'Eva EK, Selishcheva AA, Savel'Ev S V. Autoantibodies to neuron-specific proteins S100, GFAP, MBP, and NGF in the serum of rats with streptozotocin-induced diabetes. Bull Exp Biol Med. 2013;155(1):48–51.
34. Kheirouri S, Ebrahimi E, Alizadeh M. Association of S100B Serum Levels with

Metabolic Syndrome and its Components. 2018;31(4):201–6.
35. Dávalos A, Alvarez-Sabín J, Castillo J, Díez-Tejedor E, Ferro J, Martínez-Vila E, et al. Citicoline in the treatment of acute ischaemic stroke: An international, randomised, multicentre, placebo-controlled study (ICTUS trial). Lancet [Internet]. 2012;380(9839):349–57. Available from: http://dx.doi.org/10.1016/S0140-6736(12)60813-7
36. Woods SD, Flores R, Roberson PK, Lowery JD, Skinner RD, Culp WC. Decreased Serum Levels of S-100B Protein Reflect Successful Treatment Effects in a Rabbit Model of Acute Ischemic Stroke. Open Neurol J. 2011;5(1):55–7.
37. Calcagnile O, Holmén A, Chew M, Undén J. S100B levels are affected by older age but not by alcohol intoxication following mild traumatic brain injury. Scand J Trauma Resusc Emerg Med. 2013;21(1):2–7.
38. Wahyudi R, Hasmono D, Fitrina R, Armal K. Injected Citicoline Improves Impairment and Disability During Acute Phase Treatment in Ischemic Stroke Patients. Folia Medica Indones. 2016;51(4):245.
39. Glade MJ, Smith K. Phosphatidylserine and the human brain. Nutrition [Internet]. 2015;31(6):781–6. Available from: http://dx.doi.org/10.1016/j.nut.2014.10.014
40. Setiarto R, Widhyastuti N, Saskiawan I, Safitri R. Pengaruh Variasi Konsentrasi Inulin Pada Proses Fermentasi Oleh L. Acidophilus, L. Bulgaricus Dan S. Thermophillus. Biopropal Ind. 2017;8(1):1–17.
41. Ji M, Li S, Dong Q, Hu W. Impact of an early high-protein diet on neurofunctional recovery in rats with ischemic stroke. Med Sci Monit. 2018;24:2235–43.
42. Jadavji NM, Emmerson JT, MacFarlane AJ, Willmore WG, Smith PD. B-vitamin and choline supplementation increases neuroplasticity and recovery after stroke. Neurobiol Dis. 2017;103:89–100.
43. Hoffman JD, Yanckello LM, Chlipala G, Hammond TC, McCulloch SD, Parikh I, et al. Dietary inulin alters the gut microbiome, enhances systemic metabolism, and reduces neuroinflammation in an APOE4 mouse model. PLoS One. 2019;14(8):1–22.
44. Silva YP, Bernardi A, Frozza RL. The Role of Short-Chain Fatty Acids From Gut Microbiota in Gut-Brain Communication. Front Endocrinol (Lausanne). 2020;11:1–14.
45. Thelin EP, Nelson DW, Bellander BM. A review of the clinical utility of serum S100B protein levels in the assessment of traumatic brain injury. Acta Neurochir (Wien) [Internet]. 2017;159(2):209–25. Available from: http://dx.doi.org/10.1007/s00701-016-3046-3
46. Aquilani R, Sessarego P, Iadarola P, Barbieri A, Boschi F. Nutrition for brain recovery after ischemic stroke: An added value to rehabilitation. Nutr Clin Pract. 2011;26(3):339–45.
47. Richter Y, Herzog Y, Lifshitz Y, Hayun R, Zchut S. The effect of soybean-derived phosphatidylserine on cognitive performance in elderly with subjective memory complaints: A pilot study. Clin Interv Aging. 2013;8:557–63.
48. Martynov MY, Boiko AN, Kamchatnov PR, Kabanov AA, Yasamanova AN, Shchukin IA, et al. Neuroprotective therapy with citicoline (Ceraxon) in patients with ischemic stroke. Neurosci Behav Physiol. 2013;43(6):706–11.
49. Kato-Kataoka A, Sakai M, Ebina R, Nonaka C, Asano T, Miyamori T. Soybean-derived phosphatidylserine improves memory function of the elderly Japanese subjects with memory complaints. J Clin Biochem Nutr. 2010;47(3):246–55.

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The effect of addition protein, phosphatidylcholine, phosphatidylserine, and inulin on GFAP levels of acute ischemic stroke patients at Dr. Kariadi Hospital, Semarang

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ABSTRACT

Background: The occurrence of ischemia causes a loss of energy to switch to anaerobic processes resulting in acidosis due to reduced Adenosina Triphosphate (ATP). This condition makes neuron cells apoptotic. Apoptotic of several biochemical substrates in the brain, such as Glial Fibrillary Acidic Protein (GFAP) exit into the circulatory system which is associated with dysbiosis through immunological pathways.
Objectives: To determine the effect of giving enteral formula containing protein, phosphatidylcholine, phosphatidylserine, and inulin on GFAP levels in patients with acute ischemic stroke Dr. Kariadi Hospital.
Materials and Methods: This study was done in a single-blind RCT. Eighteen ischemic stroke patients were randomly divided into intervention (9 subjects) and control groups (9 subjects). The intervention group received 69 g of the powdered enteral formula three times a day for seven days. The formula contained protein (15 g), phosphatidylcholine (128 mg), phosphatidylserine (32 mg), and inulin (3 g). The subject who had diabetes mellitus received for 14 days at a dose of 34.5 g per day (7.5 g protein with additions 64mg phosphatidylcholine, 16mg phosphatidylserine, 1.5 g inulin). The control group received the standard enteral formula from the hospital, which contains (11.8 g protein without additions protein, phosphatidylcholine, phosphatidylserine, and inulin). GFAP levels by ELISA method (Enzyme-linked immunosorbent Assay) at pre and post-intervention.
Results: There was a trend of decreasing GFAP levels before and after in the intervention group towards a better direction from 8.37±4.25 to 8.30±4.9 compared with the control group which experienced an increasing trend from 5.4±1.8 to 7.5±4. There was no significant difference in GFAP levels after intervention between groups (p = 0.7).
Conclusions: The addition of protein, phosphatidylcholine, phosphatidylserine, and inulin had no significant effect on GFAP levels.

Keyword: GFAP; Protein; Phosphatidylcholine; Phosphatidylserine; Inulin

BACKGROUND

Ischemic stroke occurs when blood supply to the brain is interrupted or reduced due to a blockage. The occurrence of ischemia in cells and reduced glucose supply causes an increased glutamate expression. Glucose metabolism later switches to anaerobic processes so that it becomes acidosis due to reduced ATP. This situation prompts apoptosis or the death of nerve cells in the brain. In the event of apoptosis, some biochemical substrates may leave the brain and leak off into the circulatory system. In addition to apoptosis, there could be a reduction of proteins that play a role in maintaining the integrity of the blood-brain barrier membrane; hence membrane will encounter permeability. This results in GFAP which should be in the brain structure going out into the circulatory system.[1–4]

Glial Fibrillary Acidic Protein (GFAP) is one of the intermediate protein families, including vimentin, nestin, and is known as an intermediate filament (IF) which is a group of cell types found in the central nervous system and is responsible for maintaining the structure and migration of astroglia cells. GFAP plays a role in the mitotic process by adjusting the filamentous tissue in

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cells. Besides, GFAP also plays a role in neuron astrocyte interactions and communication between cells. Under physiological conditions, GFAP is not actively secreted by cells and is generally undetectable in the blood of healthy individuals. In a moment of brain injury, astroglia responds by producing more GFAP, which in turn will be released into the Central Nervous System (CNS) fluid and blood. GFAP is exclusively produced by astrocytes so that this protein is particularly observed in the brain. Examinations have shown that GFAP is released rapidly in cases of acute ischemic stroke, wherein levels peak within 12 hours of onset and are maintained for up to 72 hours of onset.[5]

A study by Dvorak et al which stated that serum GFAP has taken within 2-6 hours after stroke onset was significant in differentiating hemorrhagic stroke from ischemic stroke. GFAP reaches its highest level within 12 hours of stroke onset.[6] Supported by research by Puspitasari et al which states there is a significant correlation between GFAP serum levels with stroke severity scale after 1 month of stroke onset.[7] This is different from the study conducted by Neila et al, which aims to determine the difference between blood sampling time and serum GFAP levels within 24 hours of onset in patients with ischemic stroke and hemorrhagic stroke, which shows that there was no significant difference in GFAP levels in samples <6 hours, 6-12 hours, and 12-24 hours in ischemic strokes and ICH strokes.[8]

Factors that can influence GFAP include some cellular processes. In general, GFAP has an important role in the regulation between astrocytes and neurons. Changes in GFAP expression can affect synaptic function, interfere with glutamate metabolism, increase the volume of astrocytes that can expand to the surface area of neurons in contact with the astrocyte membrane, and hypoxia.[4]

Thrombolysis or *recombinant tissue plasminogen activator (*rTPA*)* may be one of his definitive therapies. However, this therapy cannot be given to all ischemic stroke patients. Given the strictness of criteria such as the time window, the time for rTPA therapy was <3 hours and a range of 3-4.5 hours after symptom onset. If the therapy is not performed according to established guidelines, one may experience a more severe risk, such as bleeding in angioedema, intracranial and gastrointestinal tract.[9] The limitations of the requirement for rTPA have made many pharmacological therapies emerged, such as neuroprotective therapies in which the addition of phospholipids given in supplement form (citicoline). Some of the most abundant types of phospholipids are phosphatidylcholine and phosphatidylserine. Phosphatidylcholine can improve the biosynthesis of membrane phospholipids which are deteriorated by an increment in free radicals during brain ischemia (neuroprotection). Besides that, the activation of the enzymes that trigger apoptosis in nerve cells will be inhibited by phosphatidylcholine so that it will affect GFAP levels.[10] Research on the effect of giving protein, neuroprotective substances (phosphatidylcholine and phosphatidylserine), and inulin on markers of brain damage, especially GFAP, is still very limited.

Animal studies with the administration of the drug cytoline have the potential to reduce acute brain damage and improve functional recovery, even when given several hours after an ischemic event because the phosphatidylcholine in cytoline can increase the biosynthesis of membrane phospholipids degraded by increased free radicals during brain ischemia (neuroprotection). Besides, cytoline has been shown to restore mitochondrial ATPase and Na1 / K1 ATPase activity to inhibit phospholipase A2 activation which triggers apoptosis of nerve cells.[11,12] so that will affect the levels of GFAP.

A Japanese study was conducted to investigate the effect of phosphatidylserine extracted from soybeans (Soy-PS) on the cognitive function of an elderly group with memory complaints. Oral administration of Soy-PS improves memory function, especially in the elderly with memory complaints. These effects were similarly observed at low doses (100 mg/day) and high doses (300 mg/day).[13]

Fulfillment of protein intake in stroke patients is often given enteral formula with high calories high protein diet which operates to preserve adequate nutritional intake to speed up the recovery period as the enteral formula contains high protein. Dysphagia, unconsciousness (coma), or what is known as enteral feeding is one alternative to meet the nutritional needs of stroke patients by providing nutrients in the form of enteral formula. The protein content in the enteral formula can improve body metabolism through appetite regulation and/or other mechanisms that can control body weight and body composition.[14] Several studies have proven the benefits of enteral formula protein in increasing protein consumption, muscle mass and possibly

increasing body mass index so that it can improve the motor function abilities of patients.[15–17]

Prebiotics is one of the nutrients needed in stroke patients in addition to phospholipids and protein. One type of which is named inulin. Prebiotic bacteria such as Bifidobacteria and Lactobacilli will ferment inulin to produce Short-Chain Fatty Acid (SCFA) and its fermented products in the form of propionate, butyrate, acetic acid, L-lactate, CO2, and hydrogen. Directly or indirectly, the gut-brain axis is influenced by SCFA. The blood-brain barrier can be penetrated by SCFA mediated by monocarboxylate transporter (MCT) and improves the integrity of the blood-brain barrier by increasing the expression of tight junction proteins. SCFA will improve inflammation in nerves by modulating levels of neurotrophic factors, improving morphology and glial cell function, and strengthening neurogenesis.[18] Previous research conducted by Liu et al explained that administering 10% inulin for 4 weeks can change the composition of the gut microbiota and improve gut function in obese mice.[19] The same thing was done by Chuncai et al which stated that the administration of NAC, inulin, and a combined therapy improved cognition in castrated rats. RX-fed rats treated with either the vehicle or testosterone had a significantly increased number of GFAP positive cells. This increment was attenuated by treatment with NAC, inulin, or the combined therapy.[20]

Enteral nutrition has been shown to reduce infectious complications and improve outcomes in patients with severe TBI. Increasing immune cell activity and the integrity of the intestinal barrier, most likely preventing immune dysfunction and bacterial translocation, are the benefits of early feeding, respectively. The nervous system and endocrine changes following enteral feeding can help reduce massive inflammation through vagus nerve-mediated mechanisms. In a journal published by Luyer in Bansal et al, it is shown that enteral feeding, especially dietary fat, stimulates the release of cholecystokinin and binds to cholecystokinin receptors which ultimately leads to activation of the efferent vagal system. This activation triggers an increase in acetylcholine at the gut synaptic level and decreases TNF-α production by way of acetylcholine binding to α-7 nicotinic receptors on macrophages and other immune cells. Therefore, the positive immunologic benefits of early enteral feeding after TBI may also be a result of decreased inflammatory cytokine production by way of intraluminal feeds inducing vagus nerve activity which can thus affect the effectiveness of GFAP levels.[21]

This study aimed to determine the effect of giving enteral formula containing protein, phosphatidylcholine, phosphatidylserine, and inulin on GFAP levels in ischemic stroke patients. The intervention group received 69 g of the powdered enteral formula containing 15 g of protein, 128 mg of phosphatidylcholine, 32 mg of phosphatidylserine, and 3 g of inulin dissolved in 250 ml of warm water, given three times a day for seven days.

Giving 69 g of powdered enteral formula because it contains high protein [22 g per 100 g in solid form][22]. The intake of phosphatidylcholine in the enteral formula also covers 60-90% of the daily requirement for phosphatidylcholine for adults and 3x1 enteral formula feeding for 7 days is following the therapeutic dosage for cytolin [23] where the administration of cytolin 2000 mg has a restorative effect.[24] According to the results of the study, it was shown that the administration of enteral nutrition within 7 days could reduce the mortality rate by 5.8% and survive more than the group who started giving nutrition late.[25] Feeding 69 g of enteral formula has been consistent with the therapeutic dose of phosphatidylserine, whereas administration of 100 mg/day of phosphatidylserine has provided improvement.[26] A dose of 0.5 g has been shown to increase the diversity of probiotic bacteria in the gut[27] which will produce short-chain fatty acids, namely SCFA. Inulin in the enteral formula has met the inulin dose for adults, which is 5-15 g /day and 0.5 g of inulin has been shown to increase the number of probiotic bacteria in the intestine which will produce short-chain fatty acids such as SCFA. In subjects suffering from diabetes, the treatment was carried out for 14 days at a dose of 34.5 g per day. The control group was given standard enteral formula from the hospital (11.8 g protein).

MATERIALS AND METHODS

This research is experimental research with a randomized control trial in a single-blind manner where the research subjects do not know what therapy is being obtained. The research subjects were 18 ischemic stroke patients aged >18 years, 9 men and 9 women who were divided into the intervention group (9) and the control group (9)

by randomization. This research was conducted for four months (February-May). The sample size was calculated by taking into account the minimum sample size by using the experimental sample size formula for clinical trials with the following formula:*****

$$n_1 = n_2 = 2\left(\frac{(Z_\alpha + Z_\beta)\ SB}{x1\text{-}x2}\right)^2$$

The results of previous studies showed that the level of GFAP in patients with ischemic stroke was 9.46 ng / mL (mean) and the standard deviation was 4.6 ng / mL.[28] The minimum difference considered significant (x1-x2) was 5.13 ng / mL.[28] If the error of type I is set at 15% (α=0.15) then Zα=1.440. The amount of type II error is set at 20% (β=0.20) then Zβ=0.842 and research power is 80%, the sample size calculation is as follows:

$$n = 2\left(\frac{(1.440 + 0.842)\ 4.6}{5.13}\right)^2$$
$$= 8.3 \approx 9 \text{ ischemic stroke patients/group}$$

When the sample was selected with the inclusion criteria of ischemic stroke patients with attack onset, not more than 72 hours as evidenced by an MSCT head scan and aged >18 years, and the exclusion criteria for ischemic stroke patients who had a head injury in the last 3 months, subarachnoid hemorrhage, brain tumor, infection of the central nervous system, psychiatric disorders, complications of acute or chronic renal failure, a history of reactions to enteral formula, hematemesis and / hematemesis + melena and those receiving citicoline. The researcher randomized the subjects by dividing the research subjects into the control group or the intervention group. Researchers provided an informed consent sheet for consent to be the study sample to the patient's family.

GFAP levels were checked at the beginning and the end of treatment. Examination of clinical characteristics is carried out by physical examination and a doctor examines neurological examination. The MSCT scan was performed to determine whether the patient had a head injury in the last 3 months, subarachnoid hemorrhage, brain tumor, infection of the central nervous system, psychiatric disorders, patient medical records to determine blood laboratory results (routine hematology, blood sugar, lipid profile) and complications. acute or chronic renal failure, history of reactions to enteral formula, hematemesis and / hematemesis + melena, and those receiving citicoline. During the study, researchers assessed the daily intake of food consumed by patients using the 24-hour recall method for 7 days. How to determine the patient's nutritional adequacy level through the average intake divided by the patient's requirement then multiplied by 100. The control group was given enteral formula based on the standard RS conventional enteral formula according to the character/comorbid of the ischemic patient (11.8 protein without the addition of phosphatidylcholine, phosphatidylserine, and inulin). The intervention group was given enteral formula containing 15 g of protein, 128 mg of phosphatidylcholine, 32 mg of phosphatidylserine, 3 g of 69 g of powdered enteral formula inulin, each dissolved in 250 ml of warm water and given three times a day for seven days. In the intervention group suffering from diabetes, the treatment was conducted for 14 days at a dose of 34.5 g per day. This provision is differentiated because the enteral formula given contains a high enough glycemic index which will have side effects in diabetes mellitus patients, so the administration will be reduced but it is carried out for a longer period. To avoid any side effects during the intervention, the researchers observed the patients every day.

The blood samples used were whole blood vein samples taken from the median cubital vein. The 6 ml blood sample was taken using a red vacutainer. The blood was then exported to the central laboratory of Dr. Kariadi for centrifuge and stored at –20ºC until testing. After the serum was successfully collected, the serum was sent to the Faculty of Medicine Undip GAKI laboratory for testing using the ELISA GFAP E-EL-H1888 kit.

This study has received Ethical Clearance approval by the Medical Research Ethics Commission FK UNDIP / RSDK with Number 503 / EC / KEPK-RSDK / 2020.

Statistical analysis was performed on a computer using the SPSS for windows version 20.0 program. Bivariate analysis was performed with the $\chi 2$ test or with the Fisher-Exact test to determine the change (Δ) in serum GFAP levels and the characteristics of the categorized subjects. The result is significant if $p < 0.05$. Multivariate analysis on confounding variables (age, sex, hypertension, diabetes mellitus, obesity status, dyslipidemia, history of stroke, rTPA therapy,

duration of intervention, and length of treatment) was carried out if any variables in the bivariate test were found to be different, with p <0.05 using General Linear Model.

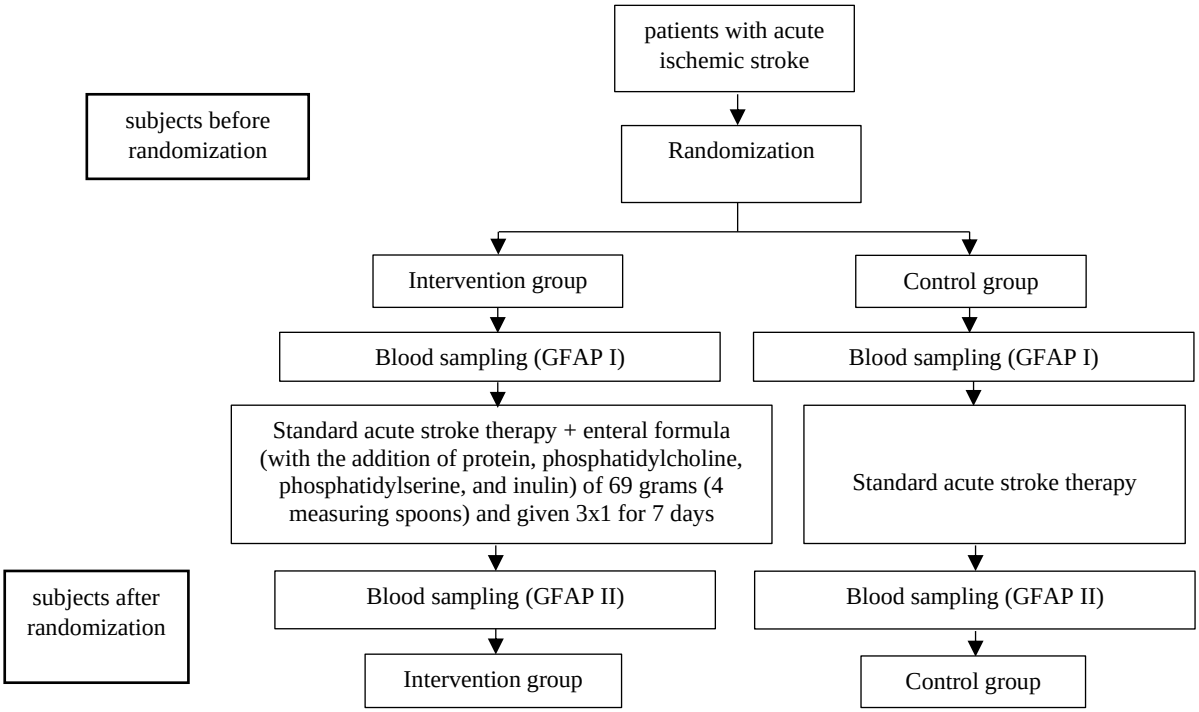

Figure 1. Flow chart of the research from the beginning to the end of the study

RESULTS

Characteristics of Research Subjects

In the implementation of the study, 8 out of 26 patients were found to have dropped out or lost to follow-up. Some of the things that caused the drop-out included patients who were not willing to continue the study (5) and died (3).

A total of 18 acute ischemic stroke patients were the subjects in the study. There were no significant differences in the categories of gender, age, BMI, obesity status, hypertension, diabetes mellitus, dyslipidemia, smoking, rTPA therapy, history of stroke, duration of intervention, and length of stay (p> 0.05). However, there were differences in the mean age category (p = 0.01).

Average Adequacy Level of Energy, Protein, Fat, Carbohydrates, Phosphatidylcholine, Phosphatidylserine, and Inulin Intake

Table 2 reveals that there is a significant difference in the adequacy of energy, protein, carbohydrate, phosphatidylcholine, phosphatidylserine, and inulin intake (p<0.05). There was no significant difference in the adequacy of fat intake between groups (p>0.05).

Difference of Mean and Change (Δ) GFAP levels

There was no difference in the mean GFAP level before the study between intervention group 8.37±4.25 and the control group 5.4±1.8 with a value (p = 0.08). These results designate that the conditions of the two groups before the intervention are corresponding (homogeneous). In the intervention group, the average GFAP level managed to decrease from 8.37±4.25 to 8.30±4.9, whereas in the control group, there was an increment from 5.4±1.8 to 7.5±4. This disparity shows that the intervention given affects even though this difference is not statistically significant (p = 0.9). Changes in Δ levels of GFAP in the intervention group also tended to decrease -0.07±2.26 compared to the control group which tended to increase 2.13±2.84 but not statistically significant (p> 0.05).

Table 1. Characteristics of Research Subjects in the Two Research Groups

Characteristics	Group		*p*
	Intervention (n=9)	Control (n=9)	
Gender			
a. Male	4 (44.4%)	6 (66.7%)	0.6[a]
b. Women	5 (55.6%)	3 (44.4%)	
Average age (years)	50.5 ± 14.8	65.5 ± 8.2	0.01[b]
Age Category (years)			
a. > 60	3 (33.3%)	7 (77.8%)	0.1[a]
b. <60	6 (66.7%)	2 (22.2%)	
Average BMI	25.2 ± 4	23.8 ± 2.6	0.3[b]
Obesity (BMI> 25)			
a. Yes	4 (44.4%)	2 (22.2%)	0.6[a]
b. Not	5 (55.6%)	7 (77.8%)	
Hypertension			
a. Yes	8 (88.9%)	7 (77.8%)	1[a]
b. Not	1 (11.1%)	2 (22.2%)	
Diabetes mellitus			
a. There is	3 (33.3%)	5 (55.6%)	0.6[a]
b. Not	6 (66.7%)	4 (44.4%)	
Dyslipidemia			
a. Yes	7 (77.8%)	8 (88.9%)	1[a]
b. Not	2 (22.2%)	1 (11.1%)	
Smoke			
a. Yes	4 (44.4%)	6 (66.7%)	0.6[a]
b. Not	5 (55.6%)	3 (33.3%)	
RTPA therapy			
a. Yes	2 (22.2%)	2 (22.2%)	1[a]
b. Not	7 (77.8%)	7 (77.8%)	
Stroke History			
a. Yes	3 (33.3%)	3 (33.3%)	1 [a]
b. Not	6 (66.7%)	6 (66.7%)	
Duration of Intervention			
a. 7 days	6 (66.7%)	9 (100%)	0.2 [a]
b. 14 days	3 (33.3%)	0 (0%)	
Length of Hospitalization			
a. <7 days	6 (66.7%)	3 (33.3%)	0.3 [a]
b. ≥ 7 days	3 (33.3%)	6 (66.7%)	

[a] *Fisher-Exact*; [b] *Independent T-Test*

Table 2. Adequacy Level of Patient Intake at Research

Variable (%)	Intervention	Control	*p*
	Mean ± SD	Mean ± SD	
Energy Adequacy Level	95 ± 18.4	75.7±11.9	0.018[b]
Protein Adequacy Level	94.1 ± 17.6	69.1±11.7	0.003[b]
Adequacy Level of Fat	74.3 ± 21.2	75.9±14.8	0.9[b]
Adequacy Level of Carbohydrates	106.9 ± 20.5	78.8±14.4	0.004[b]
Adequacy Level of Phosphatidylcholine	72.7±8	25.2±3.4	0.0[b]
Adequacy Level of Phosphatidylserine	112.3±12.3	49±10.2	0.0[b]
Adequacy Level of Inulin	92.4±7.9	24.1±7.6	0.0[b]

[b]*Independent T-Test*

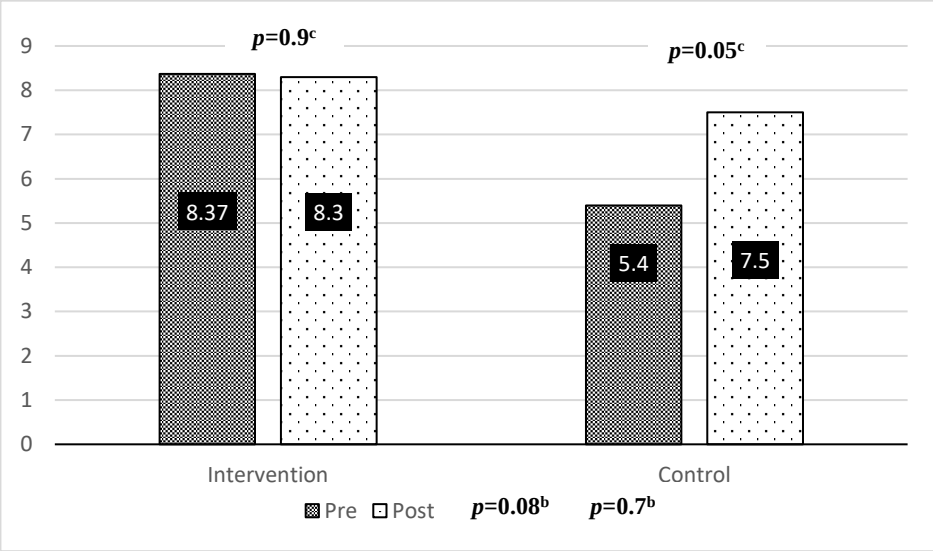

Figure 2. Average GFAP Levels Between Groups

[b] *Independent T-Test*; [c] *Paired T-Test*

Sub-Group Analysis of Inter-Group GFAP Levels

Median GFAP levels before the study in the intervention group DM subgroup 11.3 (2.4 – 12.4) with the control group 6 (3.9 – 8.1) was not statistically significant (p = 0.3). These results indicated that the state of the two groups before the intervention was in identical condition. In the DM sub-group in the intervention group, the median GFAP level increased from 11.3 (2.4 – 12.4) to 12.6 (2.1 – 12.8), it was the same as the control group which also increased from 6 (3.9 – 8.1) to 7 (3.4 – 15.6) and statistically, this difference was not significant (p>0.05). In the DM subgroup, the change in GFAP levels in the intervention group also decreased from 0.4 (-0.3 – 1.3) the same was the case with the control group 1 (-0.5 – 7.9) nonetheless, this difference was not statistically significant.

Median GFAP levels before the study in the non-DM subgroup of the intervention group 9.6 (1.1-12.3) with the control group 4.1 (2.8-6.7) were not statistically significant (p = 0.1). These results indicated that the condition of the two groups before the intervention is in the same condition. In the non-DM subgroup the intervention group median GFAP level there was a tendency to decrease from 9.6 (1.1-12.3) to 7.3 (0.8-14.2), in contrast to the control group which there was an increase from 4.1 (2.8-6.7) to 5.9 (3.8-8.2), and statistically, this difference was not significant (p>0.05). In the non-DM subgroup, the change in Δ GFAP levels in the intervention group also tended to decrease -0.2 (-3.5 – 3.4) is different the case with the control group which there was an increase of 0.8 (-0.5 – 4.9) however, this difference was not statistically significant.

Table 3. Sub-group analysis of GFAP levels between groups

Sub Group	Kelompok	n	Before	After	p	Δ
			Median (minimum-maximum)	Median (minimum-maximum)		Median (minimum-maximum)
DM	Intervention	3	11.3 (2.4 – 12.4)	12.6 (2.1 – 12.8)	0.2[d]	0.4 (-0.3 – 1.3)
	Control	5	6 (3.9 – 8.1)	7 (3.4 – 15.6)	0.1[c]	1 (-0.5 – 7.9)
	p		0.3[b]	0.8[e]		0.3[b]
Non DM	Intervention	5	9.6 (1.1-12.3)	7.3 (0.8-14.2)	0.7[c]	-0.2(-3.5 – 3.4)
	Control	5	4.1 (2.8-6.7)	5.9 (3.8-8.2)	0.2[c]	0.8 (-0.5 – 4.9)
	p		0.1[b]	0.4[b]		0.2[b]

[b] *Independent T-Test* ; [c] *Paired T-Test*; [d] *Wilcoxon*; [e] *Mann-Whitney*

GLM (General Linear Model)

Table 4 shows that there is no significant difference in GFAP levels controlled by the age variable (p = 0.3)

Table 4. General Linear Model

Variable	Covariat	*p*	R^2	Lower	Upper
(Δ) GFAP levels	Age	0.5	0.1	-0.14	0.079

DISCUSSION

Subject characteristics data revealed that there were no significant differences in the categories of gender, age, BMI, obesity status, hypertension, diabetes mellitus, dyslipidemia, smoking, rTPA therapy, history of stroke, duration of intervention, and length of stay (p>0.05). However, there were differences in the mean age category (p = 0.01). Stroke is often referred to as a disease suffered by older people. Currently, there was a trend that young people suffer from a stroke. The rising number of stroke cases at a young age can be prompted by a not-healthy lifestyle, such as smoking, alcohol consumption, frequent eating low-fiber foods such as fast food or junk food, and lack of exercise which will lead to hypertension. The habit is usually worsened by people in Central Java who are accustomed to consuming sweet foods or high glucose levels and foods containing coconut enteral formula which can trigger atherosclerosis and thrombosis, which results in reduced oxygen supply to the brain and can lead to stroke. The outcomes of the different test results for the adequacy of energy intake, carbohydrate intake, and fat intake between the intervention group and the control group showed significant results (p <0.05) but not for fat intake (p> 0.05). The exception is because one of the interventions given to the control group was standard enteral formula from the hospital, where before the 7th day and the patient had gone home, the researchers could not bring the enteral formula to continue therapy at home for one reason or another. Unlike the case with the intervention group before the 7th day; however, the patient had gone home, the researcher could provide the enteral formula to continue therapy at home. It could also be caused by monitoring food consumption before the study was completed. Researchers carry out a recall via cellular media such as WhatsApp because when the study took place coincided with the COVID-19 pandemic, which caused the patient to be unwilling to be visited by the researcher. If the patient during the study was still in the hospital, there was a standard portion for each type of diet. However, it is different from patients who have not returned home before the 7th day. There is also another possibility, such as when researchers do a recall, patients tend to overestimate or underestimate the food they eat. The size of the household measurement also varies, so it can affect recall results. There is also another possibility, such as when researchers do a recall, patients tend to overestimate or underestimate the food they eat. The size of the household measurement also varies, so it can affect recall results. There is also another possibility, such as when researchers do a recall, the patient tends to overestimate or underestimate the food consumed. The size of the household measurement also varies, so it can affect recall results.

The distribution of serum GFAP levels at the start and end of the study found that all patients were at abnormal levels (>0.11 ng/ml).[7] This could result in the presence of high-fat content or other anomalies in the sample that could prompt a matrix effect, which would interfere with detection. The hemolyzed blood sample contains the HRP analog (Horseradish Peroxidase), which can make a false-positive result. The hemolyzed sample releases a substance from the red blood cells, which can be mistaken for HRP. HRP is the critical reagent used for ELISA. Therefore, once the sample is hemolyzed, detection will be affected. This is one of the factors that can influence GFAP.

Other factors that can affect GFAP occur in several cellular processes. The drastic decrease in oxygen in the brain causes the brain to undergo anaerobic metabolism so that the availability of ATP is low. Hypoxic conditions cause neuron cell death in minutes because the brain is a part that is very sensitive to hypoxic states. Hypoxia triggers a series of pathways that lead to decreased energy and the extracellular release of glutamate. Neurons and glial cells can die through various causes such as excitotoxicity, cellular edema, oxidative stress, and inflammation. Hypoxia increases the formation of reactive oxygen species (ROS) which results in oxidative stress on cells. Increased levels of ROS are a major cause of brain tissue damage after hypoxia. ROS damage tissue directly through the modification of cellular proteins, lipids, and DNA. ROS indirectly interferes with cellular signaling and the regulation of gene expression. Astrocytes play an

especially important role in the communication between astrocytes-neurons through the release of several neurotrophic factors to maintain central nervous system homeostasis. Astrocytes also play a role in maintaining homeostasis in the environment around neurons. Recent research has shown that astrocytes are immunocompetent in the brain. If there is a disturbance in the brain that causes damage, the astrocytes will be activated and move quickly to the site of damage (astrogliosis). On excessive activation, it turns out that astrocytes increase the production of several substances, one of which is a glial fibrillary acidic protein (GFAP).[29]

Mean baseline GFAP serum levels in the intervention group 8.37±4.25 and control group 5.4± 1.8 with a value (p = 0.08). The final study serum GFAP levels in the intervention group were 8.30±4.9 and in the control group was 7.5±4. These numbers showed that the intervention given still affects even though the difference is not statistically significant (p = 0.9). In Figure 2, the intervention group declined -0.07±2.26 compared to the control group, which increased 2.45±2.86. This difference was statistically significant (p = 0.03).

The difference is partly because the patients in the study sample were patients who had repeated strokes (had a history of stroke), which according to the study of Ren *et al.* (2016) which examined GFAP levels in 13 subjects with hemorrhagic strokes, 23 subjects with ischemic strokes and 7 subjects with recurrent stroke reported that in patients with ischemic stroke, the mean serum GFAP level was significantly lower in subjects without a history of stroke compared with patients who had a previous stroke (p = 0.004). This increment occurred due to persistent pathophysiological processes including vascular disorders, long-term changes in BBB permeability, and damage to the brain parenchyma itself triggered by an initial stroke.[30] In this study, patients with recurrent strokes found five patients in the control group, three patients in the treated group.

This can then be caused by comorbid patient diseases such as diabetes mellitus. Research conducted by Lotosh *et al.* (2013) on experimental animals by inducing streptozotocin (STZ) showed that autoantibodies against neuron proteins, especially GFAP increased sharply on day five after being induced by streptozotocin. In this study, it can be concluded that STZ produces a toxic effect on astrocytes which causes the release of GFAP protein into the bloodstream and causes the formation of AAb (autoantibody) which is specific to GFAP protein.[31]

The COVID-19 pandemic can also cause another thing during the research, which resulted in a lack of research samples. Where in a study conducted by Zang *et al.* (2019) which examined the relationship between giving citicoline injection to 102 patients with hemorrhagic stroke and GFAP levels showed a significant relationship (p = 0.000).[28]

Another thing is made possible by the dose of enteral formula that is too small, the duration of administration, and the route of administration which can be the cause of the research results. The doses in this study were 45 g protein/day, 384 mg/day phosphatidylcholine, 96 g/day phosphatidylserines, and 9 g/day inulin. In some studies, the minimum duration of intervention given was 14 days and obtained significant results. [26,32–35]

The results obtained by the DM sub-group analysis of GFAP levels in the intervention group showed an increasing trend of 0.4 (-0.3 – 1.3) compared to the control group of 1 (-0.5 – 7.9) and statistically not significantly different (p>0.05). Unlike the case with the non-DM subgroup in the intervention group which showed a decreasing trend of -0.2(-3.5 – 3.4) compared to the control group where there was an increasing trend of 0.8 (-0.5 – 4.9) and not statistically significant (p> 0.05). This occurred because, in patients with hyperglycemia, it can provoke nerve degeneration through oxidative stress. Metabolic and oxidative factors often cause rapid changes in glial cells. A key indicator of this response was an increased synthesis of GFAP, which is an astrocytic marker. The event was evidenced by the study of Baydas *et al.* (2003) who conducted a study on 40 experimental animals divided into three groups. The first group was injected with STZ, the second group was injected with STZ + melatonin, and the last group was the control group. The results showed that the STZ-induced group caused glial reactivity, while the STZ + melatonin-injected group decreased the GFAP recativity.[36]

Research on the effect of giving protein, phosphatidylcholine, phosphatidylserine, and inulin on markers of brain damage such as GFAP is still minimal. In this study, the intervention was administered orally or in the form of powdered enteral formula with protein,

phosphatidylcholine, phosphatidylserine, and inulin. Efficiently, phosphatidylcholine and phosphatidylserine are almost entirely absorbed, which then spreads throughout the body across the blood-brain barrier and into the central nervous system. Inulin is easily dissolved in water and difficult to hydrolyze by enzymes in the digestive tract so that it can reach the large intestine as a whole. Probiotic bacteria will ferment inulin in the intestines and become short-chain fatty acids called SCFA.[10,23,27]

In this study, the intervention gave still affected even though it was not statistically significant (p> 0.05), according to research from Ji *et al.* (2018), a high protein diet given earlier can improve motor coordination. Currently, most of the hospitals have implemented facilities such as flexor and elbow extensor training, physiotherapy, for example, teaching good movement patterns and posture. However, this facility will not be optimal if there are patients with severe disabilities and a lack of cooperation between patients and hospital staff. Increasing SOD expression, restoring iNOS expression, and suppressing MDA expression are among the advantages of the early high protein diet. This suggests that oxidative stress can be inhibited if a high-protein diet is given early on so that free radicals and inflammation in the body can be reduced and can improve post-stroke clinical outcomes.[32]

Supported by research by Aquilani *et al*. (2008) which provided supplementation with hyperproteic nutritional formulas (10% protein = 40 grams of protein) in 20 patients for 21 days, it showed that the National Institutes Stroke Scale (NIHSS) scores of patients with protein administration increased on average 4.4 compared to a control group average of 3.0. However, there was an increase in motor recovery of the paretic arms and legs which were found to be better able to withstand gravity for 10 seconds (with legs outstretched) in the intervention group. The improvement in the performance of the extremities against gravity was less seen in the control group patients.[33] In this study, it has a ratio of 1: 2 related to protein administration with research conducted by Aquilani.

In a study conducted by Tykhomyrov *et al.* (2019) who examined the effect of giving CDP-choline or citicoline injections to 33 ischemic stroke patients with GFAP levels who gave citicoline injection of 1000 mg for 14 days, they decreased GFAP levels by 61% with (p = <0.01). CDP-choline can protect astrocytes and neurons, as well as increase angiogenic capacity through downregulation of angiostatin in post-ischemic patients with atrial fibrillation after acute ischemic stroke. Also, CDP-choline can increase cell proliferation, vasculogenesis, and synaptophysin levels and reduce GFAP levels in the peri-infarct area in ischemic stroke. In this study, enteral nutrition was given with a content of 384 phosphatidylcholine/day.[34]

In a study conducted by Kataoka *et al*. (2010) which examined the effect of Soy-PS administration on 78 elderly people with mild cognitive impairment (50-69 years) were randomly selected to consume Soy-PS (100 mg or 300 mg/day) or placebo for 6 months. Administration of Soy-PS orally for 6 months improved memory function, especially delayed memory in the elderly with memory complaints. Administration of Soy-PS was able to improve memory function, especially delayed memory in the elderly with memory complaints. Increasing the memory score is also one of the benefits of giving Soy-PS. The activation of signaling proteins and receptors that are essential for neuronal survival is influenced by phosphatidylserine. Phosphatidylserine also plays a role in signaling processes such as regulating neurotransmitters. In this study, the intervention was given with enteral nutrition containing 96 phosphatidylserines/day for 7 days.[26]

The administration of inulin in Hofman's study was 8% by injection of inulin in mice which were equivalent to 40 grams of inulin in humans per day. Hoffman's research shows that 8% inulin can increase cecal content, produce more SCFA, increase the number of bacterial enzymes in the cecum, increase systemic metabolism by modulating the gut microbiome, and can reduce the characteristics of brain inflammation in early AD (Alzheimer's Disease) compared to 4% inulin. Hoffman's research also states that 8% is considered the maximum tolerable and beneficial amount for the human organism. In this study, 9 grams of inulin per day were given.[35]

Inulin is a part of prebiotics. Prebiotics is a component of food that cannot be digested by the digestive tract enzymatically, which will then fermented by microbiota in the intestine. Inulin is a substrate that progresses the diversity of beneficial microbiota in the intestine so that the production of SCFAs such as acetate, propionate, and butyrate will increase as a result of anaerobic fermentation. The gut-brain axis is influenced by

SCFA either directly or indirectly. SCFA binds to various receptors to further inhibit histone deacetylation and affect the integrity of the intestinal barrier and intestinal mucosal immunity. The interaction performed by the SCFA on various receptors can induce indirect signaling to the brain by inducing the secretion of intestinal hormones. Systemic inflammation is affected by SCFA by regulating leukin secretion and inducing differentiation of Tregs or T regulatory cells. The blood-brain barrier can be penetrated by the SCFA through the MCT or monocarboxylate transporter and improves the integrity of the blood-brain barrier. SCFA can enhance neurogenesis, function, and homeostasis of nerve cells and improve the morphology and function of glial cells found in the central nervous system. [35,37]

RTPA therapy also appears to contribute to study results. In the study by Davalos *et al.* (2002), 47% of patients were treated with rTPA and given citicoline therapy 1000 mg per 12 hours intravenously for three days, then orally were given 2x500 mg tablets per 12 hours for a total of six weeks. rTPA can rebuild blood flow in the penumbra; therefore it is difficult for citicoline to enhance the performance caused by the effects of thrombolysis. The severity of most stroke patients in Davalos *et al.*'s study was also high so that no penumbra region was susceptible to being saved by citicoline. Consequently, in the subgroup without rTPA, the efficacy of citicoline exhibited better results than the group with rTPA.[24] In this study, 4 patients received rTPA therapy.

Furthermore, it can be caused by patients who have gone home before the 7th day. Although given enteral formula to continue therapy at home, this can cause researchers to be unable to control whether or not the intervention is complete. The drugs received and the presence of comorbidities that concern the improvement of clinical outcome in ischemic stroke patients, because of these factors, a significant reduction in GFAP levels required.

A multicentre study conducted on 4023 ischemic stroke patients who were given a protein-energy supplement of 62.5 g / day for 6-7 months in 125 hospitals and 15 countries showed that routine oral protein energy supplementation of a usual hospital diet did not improve outcomes in patients admitted with recent stroke.[38] A study by Sabin *et al.* (2013) giving citicoline to 347 ischemic stroke subjects (1g / day for 12 months) showed no significant difference (p = 0.186).[39] Supported by Clark's study that gave citicoline to 453 ischemic stroke subjects (1000 mg 2x / day for 12 weeks) showed no significant difference (p>0.05).[40] A study conducted by Manor *et al.* (2013) regarding the administration of phosphatidylserine in 30 elderly with memory complaints (300 mg/day for 12 weeks) showed no significant difference in focused attention, sustained attention, visuospatial learning, spatial short-term memory (p>0.05).[41] The study by Tuncay *et al.* (2018) who performed Enteral Formula with Probiotic Content (EFPC) intervention in 23 neuro-critical care patients for 21 days was associated with more frequent dosing of subjects showing no significant difference.[42]

The results of this study emphasized that enteral formula containing protein, phosphatidylcholine, phosphatidylserine, and inulin had a good effect on GFAP levels, as evidenced by changes (Δ) in serum GFAP levels in the intervention group which decreased -0.07±2.26 compared to the control group, which increased 2.13±2.84 although it was not statistically significant (p>0.05).

In General Linear Model (GLM), it shows that there is no significant difference in (Δ) GFAP levels were controlled by age variable (p>0,05). This may be due to the lack of research samples. During the study, there was a COVID-19 pandemic which made patients tend to hesitate or fear to have themselves examined at Dr. Kariadi. Remembering Dr. Kariadi is a referral hospital in Semarang.

The limitations of this study were the shortage of sample, length of time, and dose of administration. The advantage of this study is to provide enteral nutrition therapy to stroke patients by examining brain damage markers (GFAP) because this study is still very limited, so it can add to the literature for other researchers.

CONCLUSIONS

The addition of 15 g protein, 128 mg phosphatidylcholine, 32 mg phosphatidylserine, and 9 g inulin given for seven days had no significant effect on GFAP levels.

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REFERENCES

1. Liu G, Geng J. Glial fibrillary acidic protein as a prognostic marker of acute ischemic stroke. Hum Exp Toxicol. 2018;37(10):1048–53.
2. Surjawan Y, As'ad S, Ranakusuma TAS, Wijaya A. The different patterns of blood S100b protein and GFAP concentrations in ischemic stroke. Med J Indones. 2013;22(4):215–20.
3. Petzold A. Glial fibrillary acidic protein is a body fluid biomarker for glial pathology in human disease. Brain Res [Internet]. 2015;1600:17–31. Available from: http://dx.doi.org/10.1016/j.brainres.2014.12.027
4. Middeldorp J, Hol EM. GFAP in health and disease. Prog Neurobiol [Internet]. 2011;93(3):421–43. Available from: http://dx.doi.org/10.1016/j.pneurobio.2011.01.005
5. Žurek J, Fedora M. The usefulness of S100B, NSE, GFAP, NF-H, secretagogin and Hsp70 as a predictive biomarker of outcome in children with traumatic brain injury. Acta Neurochir (Wien). 2012;154(1):93–103.
6. Dvorak F, Haberer I, Sitzer M, Foerch C. Characterisation of the diagnostic window of serum glial fibrillary acidic protein for the differentiation of intracerebral hemorrhage and ischaemic stroke. Cerebrovasc Dis. 2009;27(1):37–41.
7. Puspitasari V, Gunawan PY, Wiradarma HD, Hartoyo V. Glial fibrillary acidic protein serum level as a predictor of clinical outcome in ischemic stroke. Open Access Maced J Med Sci. 2019;7(9):1471–4.
8. Raisa N, Sujuti H, Purnomo H, Rahayu M, Dalhar M. the Influence of Sampling Time To Serum Glial Fibrillary Acidic Protein (Gfap) Levels in Acute Stroke. MNJ (Malang Neurol Journal). 2019;5(1):30–3.
9. Presley B. Penatalaksanaan Farmakologi Stroke Iskemik Akut. Bul Rasional [Internet]. 2013;12(1):6–8. Available from: http://repository.ubaya.ac.id/21378/1/Rasional Vol 12 No 1.pdf
10. Glade MJ, Smith K. Phosphatidylserine and the human brain. 2015;31(6):781–6. Available from: http://dx.doi.org/10.1016/j.nut.2014.10.014
11. Overgaard K. The effects of citicoline on acute ischemic stroke: A review. J Stroke Cerebrovasc Dis [Internet]. 2014;23(7):1764–9. Available from: http://dx.doi.org/10.1016/j.jstrokecerebrovasdis.2014.01.020
12. Sahota P, Savitz SI. Investigational Therapies for Ischemic Stroke: Neuroprotection and Neuro recovery. Neurotherapeutics. 2011;8(3):434–51.
13. Kato-Kataoka A, Sakai M, Ebina R, Nonaka C, Asano T, Miyamori T. Soybean-derived phosphatidylserine improves memory function of the elderly Japanese subjects with memory complaints. J Clin Biochem Nutr. 2010;47(3):246–55.
14. Anderson H, Luhovyy B, Akhavan T, Panahi S. Enteral formula proteins in the regulation of body weight, satiety, food intake, and glycemia. Nestle Nutr Work Ser Pediatr Progr. 2011;67:147–59.
15. Lien DTK, Nhung BT, Khan NC, Hop LT, Nga NTQ, Hung NT, et al. Impact of enteral formula consumption on performance and health of primary school children in rural Vietnam. Asia Pac J Clin Nutr. 2009;18(3):326–34.
16. Patel S. Functional food relevance of whey protein: A review of recent findings and scopes ahead. J Funct Foods. 2015;19:308–19.
17. Mitchell CJ, McGregor RA, D'Souza RF, Thorstensen EB, Markworth JF, Fanning AC, et al. Consumption of enteral formula protein or whey protein results in a similar increase in muscle protein synthesis in middle-aged men. Nutrients. 2015;7(10):8685–99.
18. Colonic T, Homeostasis C, Smith PM, Howitt MR, Panikov N, Michaud M, et al. Reports 1000. 2013;(August):569–74.
19. Liu TW, Cephas KD, Holscher HD, Kerr KR, Mangian HF, Tappenden KA, et al. Nondigestible fructans alter gastrointestinal barrier function, gene expression, histomorphology, and the microbiota profiles of diet-induced obese C57BL/6J Mice. J Nutr. 2016;146(5):949–56.
20. Chunchai T, Keawtep P, Arinno A, Saiyasit N, Prus D, Apaijai N, et al. N-

acetyl cysteine, inulin and the two as a combined therapy ameliorate cognitive decline in testosterone-deprived rats. Aging (Albany NY). 2019;11(11):3445–62.
21. Bansal V, Costantini T, Ryu SY, Peterson C, Loomis W, Putnam J, et al. Stimulating the central nervous system to prevent intestinal dysfunction after traumatic brain injury. J Trauma - Inj Infect Crit Care. 2010;68(5):1059–63.
22. BPOM RI. Peraturan Kepala Badan Pengawas Obat dan Makanan Republik Indonesia Nomor 9 Tahun 2016 tentang Acuan Label Gizi. Badan Pengawas Obat dan Makanan Republik Indones. 2016;1–28.
23. Wahyudi R, Hasmono D, Fitrina R, Armal K. Injected Citicoline Improves Impairment and Disability During Acute Phase Treatment in Ischemic Stroke Patients. Folia Medica Indones. 2016;51(4):245.
24. Dávalos A, Castillo J, Álvarez-Sabín J, Secades JJ, Mercadal J, López S, et al. Oral citicoline in acute ischemic stroke: An individual patient data pooling analysis of clinical trials. Stroke. 2002;33(12):2850–7.
25. Wirth R, Smoliner C, Jäger M, Warnecke T, Leischker AH, Dziewas R, et al. Guideline clinical nutrition in patients with stroke. Exp Transl Stroke Med. 2013;5(1):1–11.
26. A. K-K, M. S, R. E, C. N, T. A, T. M. Soybean-derived phosphatidylserine improves memory function of the elderly Japanese subjects with memory complaints. J Clin Biochem Nutr [Internet]. 2010;47(3):246–55. Available from: http://linker.worldcat.org/?rft.institution_id=129635&spage=246&pkgName=UKPMC&issn=0912-0009&linkclass=to_article&jKey=897&issue=3&provider=NLM&date=2010-11&aulast=Kato-Kataoka+A.%3B+Sakai+M.%3B+Ebina+R.%3B+Nonaka+C.%3B+Asano+T.%3B+Miyamori+T.&atitle=Soyb
27. Setiarto RHB, Widhyastuti N, Saskiawan I, Safitri RM. Pengaruh Variasi Konsentrasi Inulin Pada Proses Fermentasi Oleh Lactobacillus acidophilus, Lactobacillus bulgaricus Dan Streptococcus thermophilus. Biopropal Ind. 2016;8(June):1–17.
28. Zang J, Liu AX, Qi L. The cytological mechanism and effects of hypertensive cerebral hemorrhage treatment by citicoline on serum GFAP and copeptin level. Eur J Inflamm. 2019;17.
29. Andriani A, Prijanti AR, Mudjihartini N, Jusman SWA. Dampak Hipoksia Sistemik terhadap Malondialdehida, Glial Fibrillary Acidic Protein dan Aktivitas Asetilkolin Esterase Otak Tikus. eJournal Kedokt Indones. 2016;4(2).
30. Ren C, Kobeissy F, Alawieh A, Li N, Li N, Zibara K, et al. Assessment of serum UCH-L1 and GFAP in acute stroke patients. Sci Rep. 2016;14(6):1–9.
31. Lotosh NG, Savel'Eva EK, Selishcheva AA, Savel'Ev S V. Autoantibodies to neuron-specific proteins S100, GFAP, MBP, and NGF in the serum of rats with streptozotocin-induced diabetes. Bull Exp Biol Med. 2013;155(1):48–51.
32. Ji M, Li S, Dong Q, Hu W. Impact of an early high-protein diet on neurofunctional recovery in rats with ischemic stroke. Med Sci Monit. 2018;24:2235–43.
33. Aquilani R, Scocchi M, Iadarola P, Franciscone P, Verri M, Boschi F, et al. Protein supplementation may enhance the spontaneous recovery of neurological alterations in patients with ischaemic stroke. Clin Rehabil. 2008;22(12):1042–50.
34. Tykhomyrov AA, Kushnir YS, Nedzvetsky VS, Grinenko T V., Kuryata O V. Citicoline affects serum angiostatin and neuro specific protein levels in patients with atrial fibrillation and ischemic stroke. Ukr Biochem J. 2019;91(5):34–45.
35. Hoffman JD, Yanckello LM, Chlipala G, Hammond TC, McCulloch SD, Parikh I, et al. Dietary inulin alters the gut microbiome, enhances systemic metabolism, and reduces neuroinflammation in an APOE4 mouse model. PLoS One. 2019;14(8):1–22.
36. Baydas G, Reiter RJ, Yasar A, Tuzcu M, Akdemir I, Nedzvetskii VS. Melatonin reduces glial reactivity in the hippocampus, cortex, and cerebellum of streptozotocin-induced diabetic rats. Free

Radic Biol Med. 2003;35(7):797–804.
37. Silva YP, Bernardi A, Frozza RL. The Role of Short-Chain Fatty Acids From Gut Microbiota in Gut-Brain Communication. Front Endocrinol (Lausanne). 2020;11(January):1–14.
38. Dennis M. Routine oral nutritional supplementation for stroke patients in hospital (FOOD): A multicentre randomized controlled trial. Lancet. 2005;365(9461):755–63.
39. Alvarez-Sabín J, Ortega G, Jacas C, Santamarina E, Maisterra O, Ribo M, et al. Long-term treatment with citicoline may improve poststroke vascular cognitive impairment. Cerebrovasc Dis. 2013;35(2):146–54.
40. Clark WM, Wechsler LR, Sabounjian LA, Schwiderski UE. A phase III randomized efficacy trial of 2000 mg citicoline in acute ischemic stroke patients. Neurology. 2001;57(9):1595–602.
41. Manor I, Magen A, Keidar D, Rosen S, Tasker H, Cohen T, et al. Safety of phosphatidylserine containing omega3 fatty acids in ADHD children: A double-blind placebo-controlled trial followed by an open-label extension. Eur Psychiatry. 2013;28(6):386–91.
42. Tuncay P, Arpaci F, Doganay M, Erdem D, Sahna A, Ergun H, et al. Use of standard enteral formula versus enteric formula with prebiotic content in nutrition therapy: A randomized controlled study among neuro-critical care patients. Clin Nutr ESPEN [Internet]. 2018;25:26–36. Available from: https://doi.org/10.1016/j.clnesp.2018.03.123

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Collaborative model as a training for increasing village health worker competency about complementary feeding

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ABSTRACT

Background: A Kader refers to a Village Health Worker (VHW), a volunteer, which becomes one of the sources of community reference. Commonly, they sustain a mother's knowledge regarding adequate complementary feeding. However, there are still some VHW who have not possessed a health education background nor been able to be independent.

Objectives: This study aimed to analyze the impact of collaborative models on VHW's competence as in knowledge, self-efficacy, attitudes, and counseling skill on the topic of complementary feeding.

Materials and Methods: The design of this research is a quasi-experiment control group pretest-postest, with the retrieval of subjects using purposive sampling. Subjects were selected based on the location of the posyandu. The population in this research was VHWs in the Lembang district were 40 VHWs in each group. The treatment group was given training intervention for 1 month with a collaborative model, combining several methods into a series. The main topic was counseling and complementary feeding. The control group was given booklets and leaflets. VHW's competence was measured using questionnaires. This research was conducted in January-April 2020.

Results: Statistical test results before the treatment of both groups showed no difference (p>0,05) in each variable. Two months after the intervention, there were significant differensces in the mean score of knowledge (p=0,001), attitude (p=0,001), and VHWs self-efficacy (p=0,000) in both groups. VHW counseling skills (p=0,149) until the first month there was not a significant difference. Unexpectedly, in the second month, the VHW counseling skills could not be observed due to the global COVID-19 pandemic. The mean score of each group increased significantly, but the score of the treatment group was higher than the control group.

Conclusions: A collaborative model is effective when compared to only providing booklets and leaflets in increasing VHW's knowledge, attitudes, and self-efficacy but not effective yet for VHW counseling skills.

Keywords: Collaborative model; Competence; Complementary feeding; VHW

BACKGROUND

The double burden of malnutrition is a health problem that still needs to be seriously addressed. The double burden of malnutrition is indicated by malnutrition problems (stunting and wasting) along with problems with overnutrition (obesity).[1] Based on Basic Health Research (*RISKESDAS or Riset Kesehatan Dasar in Bahasa Indonesia*), the prevalence of malnutrition and malnutrition under five in Indonesia is still high. The progress from 2007 to 2013 has shown improvements, although the process for correcting all forms of malnutrition is still quite slow.[2]

The prevalence of severe malnutrition and malnutrition (weight-for-age) in Indonesia in 2018 was 3.9% and 13.8%. At the provincial level, malnutrition and undernutrition for children under five in West Java were 2.6% and 10.6% respectively. The prevalence of very short and stunted children (weight-for-age) in Indonesia in 2018 is 11.5% and 19.3%. At the provincial level, very short and short children under five in West Java reveals a figure of 11.7%, and 19.4%, which is slightly higher than the national figure. In 2018 the prevalence of very thin and underweight (weight-for-height) in Indonesia was 3.5% and 6.7%. Additionally, very thin and underweight toddlers in West Java expose a figure of 3.2% and 5.2%, which is slightly lower than the national figure.[2] The most serious malnutrition problem in West Java is in West Bandung Regency with a percentage of 22.4%.[3] This high prevalence rate indicates that Indonesia is still

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experiencing serious nutritional problems that need to be prevented and addressed.

One of the factors provoking malnutrition is the inadequate provision of complementary foods and early weaning. The attitudes and understanding of mothers about how to provide complementary foods, how to maintain nutritional intake, and manage food contribute to cases of malnutrition in infants and toddlers.[4,5]

A health *kader* refers to Village Health Worker (VHW) who is a volunteer selected by the community and tasked with developing the community, in this case, VHWs are also summoned as health promoters or promoters.[6] VHWs` contributions to improving access to care and health knowledge, behaviors, and outcomes are well documented, notably for conditions such as asthma, hypertension, diabetes, and HIV/AIDS; for procedures such as cancer screening and immunizations; and for maternal and child health in general.[7–11] One of the duties of a VHW is to provide health education, especially in *Posyandu*. *Posyandu* is Integrated Services Post (SIP) in Indonesian. *Posyandu* is a platform where VHW is linked to the problems currently being faced by society. In other words, VHWs are reference persons who maintain a close relationship with the community.[12,13] The existence of *Posyandu* and VHWs is needed in a promotional and preventive approach, especially concerning improving child nutrition and maternal and child health.[14] In this case, they are expected to gain expertise and proficiency in providing counseling and giving assistance on exclusive breastfeeding and appropriate complementary foods. Based on interviews with several VHWs in Lembang Subdistrict, many VHWs were not skilled in serving and building excellent and pleasant communication. For instance, some VHWs used regional languages that sounded harsh or played high pitch while communicating with the patients.

Other factors that influence the performance of VHWs apart from knowledge include self-efficacy.[15] A person's level of confidence in his ability is expressed as self-efficacy.[16] A person who has high self-efficacy will be able to think quickly and have stable self-confidence in managing his duties when situations requiring high levels of stress.[17] In this manner, a VHW is expected to develop positivity and self-efficacy, one of which can be acquired through the learning process or training.

The local government has provided various sessions of training to VHWs, especially regarding the issues of mothers and toddlers.[18] Based on preceding research, in this Lembang sub-district, training for VHWs had been held. The methods used in the previous training were lectures and workshops, which intended to broaden the knowledge, upgrade skills of VHWs in various matters, as well as giving practical solutions in providing VHWs competence.[19,20] The method used by the Lembang District government was the conventional method. The weakness of the conventional method is that the VHWs get bored quickly, which has made it challenging to absorb the information. Based on the interview, the VHWs wanted training with a modified method because they thought that the substance presented was too heavy to be conveyed by the conventional system.

Learning methods that are considered effective for community-based health education are active and independent learning processes. One of which is the collaborative learning method.[21] Collaborative learning is an educational approach to teaching and learning that involves groups of learners working together to solve a problem, complete a task, or create a product.[22] The learning model is a framework of the learning approach, learning strategies, learning methods, learning media, and learning techniques.[23] Collaborative methods are commonly used in formal education, however, this method is rarely used for community-based health education or training.[24] In this case, this refers to arranging a learning concept that incorporates government and community participation. In the collaborative learning method, implementation with two or more parties involved is preferable.[25]

One research that implemented collaborative training in the cattle breeder community conducted by Bank Indonesia (BI) representatives of West Sumatra, BPTUHPT Padang Mengatas, Faculty of Animal Husbandry, Andalas University, Department of Animal Husbandry and Animal Health, SKPD of Animal Husbandry services in West Pasaman Regency and private business institutions designated that participants encountered changes (improvements) in cognitive aspects of (43.53%).[26]Through a collaborative method of training, VHWs are expected to improve their performance. In regards to the gap of VHW`s proficiency and their main work, it is necessary to research the effect of collaborative learning models on knowledge, self-efficacy, attitudes, and skills of VHWs in counseling regarding complementary feeding.

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MATERIALS AND METHODS

This study employed a quasi-experimental non-randomized control group pretest-postest design by taking the number of subjects using purposive sampling. The population in this study was 161 VHW in Lembang District, West Bandung Regency. Respondents in this study were 80 active VHWs who met the inclusion criteria. All respondents were divided into two groups, namely the group provided with training facilities using collaborative methods which included the provision of booklets and leaflets (the treatment group), and the group that was only given booklets and leaflets as learning facilities (control group). The treatment and control groups were determined by grouping by *Posyandu* area to anticipate interactions between groups during treatment. The subject size was calculated using the formula for the mean of two independent populations. The subjects obtained were 40 VHWs in each group.

The criteria for respondents in this study were taken based on inclusion and exclusion criteria. The inclusion criteria in this study comprise; VHWs are active with 2-5 years of experience, VHW attends the activities, and VHWs are willing to become respondents. VHWs who were unable to attend the event and not willing to be respondents are involved in the exclusion criteria.

This training was conducted in January - April 2020 in Lembang District, West Bandung Regency. The independent variable in this study is the provision of training with a collaborative learning model for VHWs, while the dependent variable is the knowledge, attitudes, self-efficacy, and counseling skills of VHWs. The variables of knowledge, attitudes, and VHW counseling skills were obtained by using a questionnaire that had been tested for validation and reliability testing. The self-efficacy variable uses the General Self-efficacy Scale (GSE) instrument developed by Schwarzel & Jarusalem (1995) in the adaptation of Indonesian which has also been tested for validation and reliability testing.[27]

The pre-test was carried out in the first to the third week of January. Training for the treatment group was carried out from the fourth week of January to the third week of February. Treatment is carried out once a week, on Saturdays for training and during the activities in Posyandu for facilitator assist. The first post-test was carried out in the first to the third week of March and the second post-test was carried out in the first to the third week of April. The training for the treatment group was guided by two facilitators. There were 2 facilitators, consisting of 1 person from the West Bandung District Health Office and 1 lecturer from the Indonesian Education University (Universitas Pendidikan Indonesia). Facilitators are selected based on recommendations from the health office and university. Researchers also discuss and explain the methods used in the training to be implemented. The researcher reiterates whether the facilitator can use an intervention method that combined seminar, role-playing, group discussions, and technical guidance in a series or not. Then decide whom the facilitator will fill in the training. The distribution of booklets and leaflets for VHWs (control group) was carried out at the same time as the first week of training for the treatment group at different locations.

Booklet and leaflet containing discussion of balanced nutrition for infants around 6-24 months. An infant's need for energy and nutrients starts to exceed what is provided by breast milk, and complementary foods. A detailed discussion about what to how recommends that infants start receiving complementary foods. The booklet also contains an explanation of basic communication and counseling for VHWs. Meanwhile, the leaflet only contains infographics regarding complementary feeding, which is made short and easy to understand. This leaflet is given to be a material that VHWs can use in counseling.

Implementation of Training

The training for the treatment group was carried out within 1 month. In the first and second weeks of meetings, training was directed at one of the selected *Posyandu*. In the first week, the VHWs studied a set of materials on complementary foods. The training was carried out with the guidance of two facilitators. At first, VHWs who had been gathered in one class were given a set of booklets and leaflets, succeeded by the presentation by the facilitator. The facilitator presented the materials verbally with slide tools. In presenting the material, the facilitator also explains using props and occasionally explains in role plays.

Subsequently, VHWs were asked to discuss how to form groups of 5-6 people. Each VHW was demanded to make a question. The facilitator exchanged the questions, and later each group was urged to solve the question through discussion. The facilitator went around monitoring the discussion activities and helped out once in a while. Each group presented the results of the discussion, and the other groups presented their responses. The facilitator helped by explaining the remaining unanswered questions.

In the second week, the material presented is about counseling activities. The facilitator also carried out the training activities like the first week. The activities were more or less like the prior week. Two facilitators also conducted the material for the second week. In the third week, the facilitator assisted VHWs during the activities in *Posyandu*. The facilitator reaffirmed the core points to be conveyed to each of the VHWs.

To conduct a statistical analysis of the characteristics of the respondents, we employed descriptive and frequency distribution tables. Analysis of the mean difference in confounding variables used the independent t-test for normally distributed data, the Mann Whitney test for abnormally distributed data, and the Chi-square test for nominal data types. The difference in the mean of each dependent variable in the two groups before and after the intervention was carried out using a paired t-test because the data were normally distributed. To find the mean difference test for each dependent variable before and after the intervention between groups, we utilized the Mann-Whitney test because the data were not normally distributed. The multivariate test conducted was a general linear model, repeated measure against each variable after the intervention in both groups. The statistical test in this study used a 95% confidence level. This research has passed the research ethics review by the Health Research Ethics Commission of the Faculty of Medicine, Diponegoro University.

RESULTS

Respondent Characteristics

Table 1 explicates that most of the age of VHWs in the control and treatment groups were included in the age group 36-55 years. According to the inclusion criteria, the VHWs possessed 2-5 years of experience. In the control group, there were two male VHW members, while in the treatment group, all were women. Most of VHWs had graduated from high school. Most of VHWs had attended previous training and during the research process also received other information regarding research material from other sources. Based on the results of statistical tests (p> 0.05), the characteristics of the control and treatment groups respondents did not have a significant difference or were in the same condition. Equal conditions in the two groups before the treatment was given, are expected to describe the comparison of how far the treatment results in the two groups in this study. later these results can be generalized to a larger subject with the same conditions. Equal conditions also affect the causal relationship between the independent variable and the dependent variable.[28,29]

Table 1. Characteristics of VHW

Characteristics of VHW	Control			Treatment			*p*
	Mean ± SD	Min	Max	Mean ± SD	Min	Max	
Age (years)	43.40 ± 9.48	22	71	43.23 ± 9.37	23	59	0.934[1]
Length of time as a VHW (years)	3.48 ± 1.15	2	5	3.52 ± 1.11	2	5	0.842[2]
Characteristics of VHW		**n**	**%**		**n**	**%**	
Gender							0.247[3]
Male		2	5		0	0	
Female		38	95		40	100	
Education							0.213[2]
SD		8	20		5	12.5	
Junior High		11	27.5		10	25	
High school		19	47.5		20	50	
D3 / S1		2	5		5	12.5	
Training							0.500[3]
Yes		36	90		35	87.5	
Not		4	10		5	12.5	
Other sources of information							0.644[3]
Yes		36	90		36	90	
Not		4	10		4	10	

[1] Independent sample T-Test
[2] Mann-Whitney
[3] Chi-square
* Significant

VHW Competence before Treatment

Before VHWs were given training and facilities to learn, VHWs were first given a pre-test to see the initial scores in the two groups. The results of this study indicated that the knowledge, attitudes, self-efficacy, and VHW skills of counseling before being treated statistically have no differences between groups (p> 0.05). The absence of this significant difference indicates that the two groups possessed identical initial conditions.

VHW Competence 1 Month after Treatment

The data collection for the post-test 1 was carried out one month following the training completion, namely in the 4th week of February to the 3rd week of March 2020. Based on the results in Table 2, the statistical test showed a difference in the mean score of each variable (p <0.05) between the control group and the treatment group after 1 month of training, except for the variable of counseling skills. The difference in mean scores stated that the collaborative model training carried out for the treatment group exhibited a higher and significant increase in scores compared to the control group which was only given booklets and leaflets as learning facilities. However, in the counseling skills variable of VHWs, there were no significant differences between groups, but the change in scores in the treatment group was greater.

VHW Competence 2 Months after Treatment

Data collection for post-test 2 was carried out two months after the training was completed, namely in the 4th week of March to the 3rd week of April 2020. For the variable of VHWs counseling skills in post-test 2, observations could not be made when the VHWs conducted counseling during the *Posyandu* activities. Observations could not be executed because of the pandemic, which resulted in the elimination of *Posyandu* activities in April.

Based on the results in Table 2, the statistical test showed a difference in the mean score of each variable (p <0.05) between the control group and the treatment group after two months of training, except for the variable counseling skills. In post-test 2, the mean score for each variable also showed that the treatment group had a significantly higher score. Just like in post-test 1, this attested that training with collaborative methods provides more optimal results when compared to only providing booklets and leaflets as learning materials for VHWs. The optimal result shows that the collaborative method/learning process is one of the suitable methods for VHW learning. Collaborative learning is an educational approach to teaching and learning that involves groups of learners working together to solve a problem, complete a task, or create a product. The main characteristics of collaborative learning are a common task or activity in small group learning, cooperative behavior, interdependence, and individual responsibility and accountability.[30,31]

Table 2. **Average Competency Score of VHWs in Each Group**

VHW **Competence**	**Pretest**						
	Control			**Treatment**			
	Mean ± SD	**Min**	**Max**	**Mean ± SD**	**Min**	**Max**	***p***
Knowledge	68.38±10.7	45	85	68.0±10.73	40	85	0.876[1]
Attitude	71.95±4.94	62	82	71.87±4.76	62	82	0.945[1]
Self-efficacy	34.08±2.53	29	39	34.23±2.57	29	39	0.793[1]
Counselling Skills	70.38±12.31	45	90	70.13±11.12	50	90	0.924[1]
	Post Test 1						
Knowledge	71.88±11.41	50	95	79.87±11.17	60	100	0.002[1*]
Attitude	73.15±4.45	66	82	76.45±4.19	68	85	0.001[1*]
Self-efficacy	34.35±2.77	30	40	36.53±3.64	31	43	0.004[1*]
Counselling Skills	72.63±11.98	45	90	76.25±10.17	50	95	0.149[1]
	Post Test 2						
Knowledge	72.25±10.56	50	95	80.50±11.08	60	100	0.001[1*]
Attitude	73.45±4.26	66	82	76.78±4.1	69	85	0.001[1*]
Self-efficacy	34.65±2.87	30	41	37.30±3.52	31	44	<0.001[1*]
Counselling Skills	-	-	-	-	-	-	-

[1] Independent sample T-Test
*significant

Change in VHW Competence

Changes in the competence of VHWs in the control group and the treatment group were seen by comparing the mean values of pre-test, post-test 1, and post-test 2 in each group. The difference between each pair of data collection time (pre-test - post test1, post test1 - post-test 2, and pre-test - post-test 2) will also be seen in the comparison between the control group and the treatment group.

VHW Knowledge

The comparison of the mean scores of VHWs knowledge in pre-test - post-test 1 and pre-test - post-test 2 showed that there was a significant difference ($p < 0.05$) in the two groups, but there was no significant change ($p = 0.323$ and $p = 0.200$) in comparison of the mean score of post-test 1 to post-test 2. Likewise, based on the comparison of the delta score, the knowledge value of VHWs between groups ($p = 0.633$). The results of the different mean score test are in line with previous research which also showed a significant difference from pre-test to post-test 1 and from pre-test to post-test 2.[32]

Although both groups had a statistically significant mean score of knowledge, the mean final score in the treatment group was more notable than the control group. This result is also in line with other studies, although there were very significant differences in the two groups, the changes that occurred in the treatment group were much greater than in the control group.[33]

Attitude of VHWs

The comparison of the mean score of the attitude of the VHWs in the pre-test - post-test 1, post-test 1 - post-test 2, and pre-test – post-test 2 showed that there were significant differences in the two groups. Nonetheless, if you look at the mean score of the attitude of the VHWs in the post-test 1 and 2, the treatment group has a higher mean score than the control group. This event is in line with the results of previous research that the provision of nutrition education interventions for mothers of toddlers and *Posyandu* VHWs increased attitudes, the average nutritional attitude of VHWs was higher in the intervention group than in the control group.[34]

On the change in the mean score of the attitude of the VHWs, the comparison of the time pairs of data taking post-test 1– post-test 2 had insignificant results ($p = 0.662$). When viewed from the changes or deltas of each pair of data collection time, it can be seen that the most massive increment in value is in the difference between the pre-test and post-test 2 values in each group, especially the treatment group. This result is in line with other studies. From the pre-test and post-test, it can be seen that there is an increase in the mean attitude of *VHW*s in exclusive breastfeeding efforts in both groups.[33]

Table 3. Changes in Mean Score before and after Treatment

Change (delta)	Control	*p*	Treatment	*p*	*p*
			VHW Knowledge		
Pre – Post1	3.5 ± 3.6	<0.001[1*]	11.87 ± 5.15	<0.001[1*]	<0.001[3*]
Post1 – Post2	0.38 ± 2.37	0.323[1]	0.62 ± 3.04	0.200[1]	0.633[3]
Pre – Post2	3.88 ± 3.49	<0.001[1*]	12.5 ± 5.31	<0.001[1*]	<0.001[3*]
Pre – Post1 – Post2		<0.001[2*]		<0.001[2*]	
			Attitude of VHWs		
Pre – Post1	1.2 ± 1.36	<0.001[1*]	4.58 ± 2.01	<0.001[1*]	<0.001[3*]
Post1 – Post2	0.3 ± 0.61	0.003[1*]	0.33 ± 0.62	0.002[1*]	0.662[3]
Pre – Post2	1.5 ± 1.5	<0.001[1*]	4.9 ± 2.16	<0.001[1*]	<0.001[3*]
Pre – Post1 – Post2		<0.001[2*]		<0.001[2*]	
			VHW Self Efficacy		
Pre – Post1	0.28 ± 0.75	<0.001[1*]	2.3 ± 1.45	<0.001[1*]	<0.001[3*]
Post1 – Post2	0.3 ± 0.61	0.003[1*]	0.78 ± 0.89	<0.001[1*]	0.006[3*]
Pre – Post2	0.58 ± 0.98	0.001[1*]	3.08 ± 1.69	<0.001[1*]	<0.001[3*]
Pre – Post1 – Post2		<0.001[2*]		<0.001[2*]	
			Counselling Skills		
Pre – Post1	2.25 ± 2.52	<0.001[1*]	6.13 ± 3.84	<0.001[1*]	<0.001[3*]
Post1 – Post2	-	-	-	-	-
Pre – Post2	-	-	-	-	-
Pre – Post1 – Post2		-		-	

[1] paired samples t-test
[2] repeated measures
[3] Mann-Whitney
*significant

VHW Self Efficacy

The comparison of the mean scores of the VHWs self-efficacy in the pre-test - post-test 1. post-test 1 - post-test 2. and pre-test - post-test 2 explicated that there were significant differences in the two groups. Based on the results of different tests. the mean score of the VHW's self-efficacy in the two groups had a significant change. These results are in line with other studies that show a significant difference in self-efficacy scores after attending the training. However. if you look at the mean scores of the VHWs self-efficacy in the post-test 1 and 2, the treatment group had a higher mean score than the control group.[35]

Counseling Skills

The results of the comparison of the mean score of VHWs counseling skills in the pre-test and post-test 1 showed that there were significant differences in the two groups. The comparison of the mean score of VHWs' counseling skills at the pre-test or post-test 1 with the post-test 2 in the two groups was not calculated because the variables were not taken at that time.

Based on the different test results, the mean score of counseling skills of VHWs in the two groups had a significant change. If we see that the mean score of the post-test 1 treatment group has a higher mean score than the control group. But if you look back at table 1, the comparison of the mean score of the counseling skills of VHW between each group on the post-test 1 was not significant.

DISCUSSION

Collaborative Model and VHW Competence

VHW Knowledge

VHWs in the treatment group or those who received training using the collaborative method coupled with the provision of booklets and leaflets experienced a higher increase in knowledge compared to the control group who were only given booklets and leaflets. Based on the compared mean pre-post1-post2 score with statistical tests, it can be seen that there is a significant difference ($p < 0.001$) in the increase in the mean score of VHW knowledge of the two groups over time. Other research on collaborative models also shows that the condition of knowledge and understanding of learning citizens after participating in the application of collaborative learning models shows a significant increase. This condition is an indication that shows that the application of the collaborative learning model is quite powerful in increasing the knowledge and understanding of citizens in achieving optimal learning outcomes. The name social constructivism flows from the belief that learners construct their networks of knowledge by collaborating with others as they connect new information to their present knowledge and interests. Because each person is different, students come away from the same activity or lesson with different individual representations of the ideas studied.[36] Active learning like the collaborative model, providing students with opportunities to interact with people from a wide range of social, cultural, and ethnic backgrounds. It is often directed towards priority health needs and the redistribution of resources to specific populations and requires a synthesis of clinical skills, knowledge, capabilities, and attitudes.[21]

As stated in theory, health education in the short term can result in changes and increases in knowledge of individuals, groups, and society.[15] Likewise, the results of other studies stated that the lecture method training accompanied by discussions, simulations, and practices increased student knowledge in weighing toddlers at *Posyandu*.

Attitude of VHWs

Based on the compared mean pre-post1-post2 score with statistical tests, it can be seen that there is a significant difference ($p < 0.001$) in the increase in the mean score of an attitude of the VHW of the two groups over time. However, the increase in the mean value of the treatment group was greater than the control group. The significant increase in scores from pre-test to post-test 2 in the treatment group indicated that training using collaborative methods was optimal in improving VHWs' attitudes.

The increase in the attitude of VHWs in the treatment group was allegedly due to a stimulus in the form of support from the training facilitators (the University and West Bandung District Health Office), and village officials because of the cooperation. This statement is following the theory which asserts that attitude formation is determined by several elements including personal experience, the influence of others who are considered essential, cultural influences, mass media, educational institutions, and religious institutions as well as emotional influences.[37] External factors that are deliberately given can change human attitudes, so that consciously or not, the individual concerned will adopt a certain attitude.[38]

Learning methods that are considered effective for community-based health education are active and independent learning processes.[18] Collaborative learning is an educational approach to

teaching and learning that involves groups of learners working together to solve a problem, complete a task, or create a product.[22] This research also combines discussion methods, role modeling, and technical guidance in the field during the training process as a stimulus to get an energetic atmosphere when VHWs receive information unlike the case with the control group, which only received material in the form of booklets and leaflets.

VHW's Self Efficacy

Based on the results of statistical tests, the mean score of the VHWs' self-efficacy in the two groups had a significant change after treatment. Also, based on the compared mean pre-post1-post2 score, it can be seen that there is a significant difference ($p < 0.001$) in the increase in the mean score of VHWs' self-efficacy of the two groups over time. This is in line with the results of previous studies, that there was a change in scores in the two groups that were significantly different. The significant results up to post-test 2 indicate that the training effect can still be felt by the experimental group when the measurement is carried out.[35]

However, if you look at the mean score of the VHWs' self-efficacy in post-test 1 and 2, the treatment group has a higher mean score than the control group. A study affirmed that training and experience have a direct effect on knowledge, self-efficacy, and skills.[39] This statement is consistent with that given to the treatment group. During the training, the facilitator provided an overview of examples of counseling in *Posyandu* activities; it presented experience for VHWs and has made it easy for them to understand and retain information.

In this case, optimal learning suggests that the ability of the learning community after they receive the learning experience changes. Learning to realize that knowledge is not something fixed or finished, but always requires further elaboration and depending on the perspective taken. This condition means that there had been an increase from the previous one or what is usually occurred, due to the innovative learning model.[40] This point can be reached by developing a learning model that can increase the learning outcomes of citizens, namely through collaborative learning models.

Counseling Skills

Based on the compared mean score of counseling skills of the control group (72.63 ± 11.98) and the treatment group (76.25 ± 10.17) in Table 1, it is found that there is no significant difference ($p = 0.149$) between these groups. Thus, although the mean score of VHW skills in the treatment group was greater, this collaborative model was not yet effective in improving VHWs' counseling skills until the first month. Although based on the compared mean pretest - post-test 1 between the control and the treatment group there was a significant difference ($p < 0.001$). This significant increase in the delta is not sufficient to describe a significant increase in VHW skills in the final results of the comparison between groups. Skills are procedural matters that require repeated practice so that they can become new reflex habits that are not easily lost.[41] The direct practice method is one method that can be used to implement skills.[42]

CONCLUSION

Collaborative methods by providing booklets and leaflets to the treatment group increased the knowledge, attitudes, and self-efficacy of VHWs more than the control group, which was only given booklets and leaflets. Collaborative methods are more effective in increasing VHWs competence so that collaborative methods can be applied as an alternative training model for VHWs. However, collaborative methods have not been able to significantly improve the counseling skills of VHWs until the first month.

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REFERENCES

1. WHO. The double burden of malnutrition: Policy Brief. WHO. 2016;
2. Kementerian Kesehatan RI. Riset Kesehatan Dasar: Riskesdas 2013. Kementerian Kesehatan RI. 2013;
3. Dinas Kesehatan Provinsi Jawa Barat. Profil Kesehatan di Jawa Barat Tahun 2016. Dinas Kesehatan Jawa Barat. 2017;
4. Hijra H, Fatimah-Muis S, Kartasurya MI. Inappropriate complementary feeding practice increases the risk of stunting in children aged 12-24 months. Universa Medicina. 2016;

5. Pokharel P, Adhikari A, Lamsal P, Adhikari R. Effect of Complementary Feeding Practices and Nutritional Status of Children (6-23 months) in Tamang Community, Ambhanjyang VDC of Makwanpur. Janaki Medical College Journal of Medical Science. 2017;
6. Ismawati C. Posyandu dan Desa Siaga. Yogyakarta: Nuha Medika; 2010.
7. Rosenthal EL, de Heer H, Rush CH, Holderby LR. Focus on the future: a community health worker research agenda by and for the field. Progress in community health partnerships : research, education, and action. 2008;
8. Swider SM. Outcome effectiveness of community health workers: An integrative literature review. Public Health Nursing. 2002.
9. Brownstein JN, Chowdhury FM, Norris SL, Horsley T, Jack L, Zhang X, et al. Effectiveness of Community Health Workers in the Care of People with Hypertension. American Journal of Preventive Medicine. 2007.
10. Brownstein JN, Bone LR, Dennison CR, Hill MN, Kim MT, Levine DM. Community health workers as interventionists in the prevention and control of heart disease and stroke. American Journal of Preventive Medicine. 2005.
11. Lewin S, Babigumira S, Bosch-Capblanch X, Aja G, van Wyk B, Glenton C, et al. Lay health workers in primary and community health care: A systematic review of trials. Lay Health Workers. 2006;
12. Pradana D. Pengaruh Pendidikan Kesehatan Oleh Kader Terhadap Praktek Ibu Dalam Pemberian Mp-Asi Di Wilayah Kerja Puskesmas Ledokombo Kabupaten Jember. Universitas Jember; 2012.
13. Vaughan JP, Walt G. Village health workers, and primary health care. Tropical Doctor. 1983;
14. Yumianti. Faktor-Faktor Yang Mempengaruhi Keaktifan Ibu Balita Berkunjung Ke Posyandu Delima Mekar 03 Di Desa Karangdadap Kec. Kalibagor, Kab. Banyumas. Universitas Muhammadiyah Purwokerto; 2014.
15. Notoatmodjo S. Pendidikan Dan Perilaku Kesehatan. Jakarta: Rineka Cipta; 2003.
16. Bandura A, Caprara GV, Barbaranelli C, Pastorelli C, Regalia C. Sociocognitive self-regulatory mechanisms governing transgressive behavior. Journal of Personality and Social Psychology. 2001;
17. Baranowski T, Cheryl P, Guy SP. How individual. environment and health behavior interact, social cognitive theory in Text Book Health Behavior and Health Education. 2nd ed. California: Jossey-Bass Inc; 1997.
18. Hariyanti T, Prabandari YS. Efektivitas Metode Kolaboratif Learning dan Kooperatif dalam Pendidikan Kesehatan Berbasis Komunitas. Jurnal Pendidikan Kedokteran Indonesia. 2014;3(1):9–17.
19. Rusmana A, F Boesoeri S, Khadijah ULS. Pendidikan Pola Hidup Sehat Melalui Literasi Kesehatan Di Kecamatan Lembang Kabupaten Bandung Barat. Dharmakarya: Jurnal Aplikasi Ipteks untuk Masyarakat. 2017;6(3):153–5.
20. Tanbrin NNI. Program Pelatihan Intervensi Dini Bersumberdaya Keluarga Bagi Kader Rehabilitasi Berbasis Masyarakat Di Desa Lembang Kabupaten Bandung Barat. Universitas Pendidikan Indonesia; 2020.
21. Mennin S, Petroni-Mennin R. Community-based medical education. Clinical Teacher. 2006;
22. Laal M, Laal M. Collaborative learning: What is it? In: Procedia - Social and Behavioral Sciences. 2012.
23. Risman Z. Pengaruh Model Pembelajaran Kolaboratif Terhadap Hasil Belajar IPA Peserta Didik Kelas V MIS DDI Bosalia KAB. Jeneponto. Universitas Islam Negeri (UIN) Alauddin Makassar; 2017.
24. Jones KA, Jones JL. Making Cooperative Learning Work in the College Classroom : An Application of the 'Five Pillars' of Cooperative Learning to Post-Secondary Instruction. 2008;8(2):61–76.
25. Beard C, John P. Experiential Learning: A Best Practice Handbook for Educators and Trainer. Kogan Page Publishers; 2006.
26. Madarisa F. Pelatihan Kolaboratif Untuk Perbaikan Kompetensi Peternak Sapi di Kabupaten Pasaman Barat. Universitas Andalas; 2016.
27. Schwarzer R, Jerusalem M. Generalized self-efficacy scale. Measures in health psychology: A user's portfolio. 1995;
28. Plutchik R. The nature of emotions: Human emotions have deep evolutionary roots, a fact

that may explain their complexity and provide tools for clinical practice. American Scientist. 2001;
29. Mumford MD, Hester KS, Robledo IC, Peterson DR, Day EA, Hougen DF, et al. Mental Models, and Creative Problem-Solving: The Relationship of Objective and Subjective Model Attributes. Creativity Research Journal. 2012;
30. Chandra R. Collaborative Learning for Educational Achievement. IOSR Journal of Research & Method in Education (IOSR-JRME). 2015;
31. Panitz T. Collaborative versus Cooperative Learning: A Comparison of the Two Concepts Which Will Help Us Understand the Underlying Nature of Interactive Learning. Cooperative Learning and College Teaching. 1997;
32. Sukiarko E. Pengaruh Pelatihan Dengan Metode Belajar Berdasarkan Masalah Terhadap Pengetahuan Dan Keterampilan Kader Gizi Dalam Kegiatan Posyandu Studi Di Kecamatan Tempuran Kabupaten Magelang. Universitas Diponegoro; 2007.
33. Jumiyati, Nugrahaeni S, Margawati A. Pengaruh Modul Terhadap Peningkatan Pengetahuan, Sikap Dan Praktek Kader Dalam Upaya Pemberian Asi Eksklusif. Gizi Indonesia. 2014;37(1):19–28.
34. Nikmawati E, C KM, Khomsan A, Sukandar D, Atmawikarta A. Intervensi pendidikan gizi bagi ibu balita dan kader Posyandu untuk meningkatkan PSK (Pengetahuan Sikap dan Keterampilan) serta status gizi balita. Jurnal Pendidikan Teknologi Kejuruan. 2009;5(15):407–17.
35. Ardiyanti D, Alsa A. Pelatihan " PLANS " untuk Meningkatkan Efikasi Diri dalam Pengambilan Keputusan Karir. Gadjah Mada Journal Of Propfesional Psychology. 2015;1(1).
36. Jacobs GM. Collaborative Learning or Cooperative Learning? Beyond Words. 2015;3(1).
37. Strachan DL, Källander K, Ten Asbroek AHA, Kirkwood B, Meek SR, Benton L, et al. Interventions to improve motivation and retention of community health workers delivering integrated community case management (iCCM): stakeholder perceptions and priorities. American Journal of Tropical Medicine and Hygiene. 2012;
38. Gisore P, Rono B, Marete I, Nekesa-Mangeni J, Tenge C, Shipala E, et al. Commonly cited incentives in the community implementation of the emergency maternal and newborn care study in western Kenya. African Health Sciences. 2013;
39. Partiprajak S, Thongpo P. Retention of basic life support knowledge, self-efficacy and chest compression performance in Thai undergraduate nursing students. Nurse Education in Practice. 2016;
40. Elbers E, Streefland L. Collaborative learning and the construction of common knowledge. European Journal of Psychology of Education. 2000;
41. Sugiarto I. Mengoptimalkan Daya Kerja Otak dengan Berpikir Holistik dan Kreatif. Jakarta: Gramedia Pustaka Utama; 2011.
42. Herijulianti E, Indriani T, Artini S. Pendidikan Kesehatan Gigi. Jakarta: EGC; 2001.

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The acute supplementation of combination juice of yellow watermelon *(citrullus lanatus thunb.)* - plantain (*musa paradisiacal* var. *Sapientum l.*) suppress post-exercise blood lactic acid production in rats

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ABSTRACT

Background: Yellow watermelon contains citrulline, which can suppress lactic acid production, while plantains contain potassium which is important for muscle performance. The yellow watermelon and plantain combination juice potential to be a natural sports drink that delays muscle fatigue by suppressing lactic acid production after exercise.
Objectives: To determine the effect of yellow watermelon-plantain juice on lactic acid in rats after swimming test.
Materials and Methods: This true experimental study used a post-test-only with controlled group design. Thirty Sprague Dawley rats, eight-week-old, male, were divided into five groups, namely positive control (C+), negative control (C-), dose 1 (P1), dose 2 (P2), and dose 3 (P3). The C (+) group received no juice and was not tested swimming, the C (-) group received no juice but was tested swimming, P1 received combined juice up to 1.8 g and tested swimming, P2 received combined juice up to 3, 6 g and tested swimming, P3 received combined juice up to 1.8 g with the addition of 0.27 g granulated sugar and tested swimming. The juice is given 30 minutes before the test. The swim test was performed for three minutes; after that, the blood was taken to test the lactic acid levels. The data were analyzed using the one-way ANOVA and the advanced post-hoc with the least significant difference test.
Results: The lactic acid levels in C (+), in C (-), P1, P2, and P3 groups after swimming test were 1.38 mMol / L; 7.14 mMol / L; 3.74 mMol / L; 1.66 mMol; and 2.91 mMol/L. There were differences in levels of lactic acid ($p < 0.05$) in each group after the combination juice intervention was given.
Conclusion: Combination juice of yellow watermelon-plantain has an effect on lactic acid levels after swimming test. Dose 2 (3.6 g) was the best because it produces the lowest lactic acid after the swimming test.

Keywords: Yellow watermelon; Plantain; Lactic acid; Swimming test

BACKGROUND

Energy metabolism during anaerobic exercise is accentuated exclusively from muscular strength with high explosive power.[1] This process begins with the glucose breakdown process (glycolysis) as well as the glycogen breakdown process (glycogenolysis) and is independent of oxygen availability during ATP (Adenosine Tri Phosphate) formation.[2] Thus, it is important to acknowledge glucose availability in the muscle since its metabolic advantage is providing speed and strength in a short period.

Low performance in exercise can be caused by lactic acid accumulation which faster than it should be. However, exercising has an unavoidable side effect that is the production of lactic acid, which might induce muscle fatigue. L-citrulline is one of the most popular types of nutritional supplements that are legal ergogenic aids. L-citrulline has beneficial functions such as accelerating metabolite waste removal like lactic

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acid, improving endurance performance, and promoting faster recovery after exercise.[3] L-citrulline also acts as a source of energy during exercise.[4] A findings by Pérez-Guisado and Jakeman reported that supplementation of 8 g of citrulline malate can reduce muscle soreness at 24 and 48 hours following anaerobic exercise.[5] Supplementation with 8 g of citrulline 1 hour before exercise also can increase the number of reps lifting weights.[6]

Previous studies related to nutritional ergogenic have investigated that some fruits potentially be a source of nutritional ergogenic. Watermelon is one type of fruit that naturally rich in amino acids including L-citrulline. Watermelon also contains carbohydrates, potassium, phytonutrients such as carotenoids (lycopene and beta carotene), polyphenolics, and vitamins.[7] The rind watermelon juice contains 45.02 mg/g L-citrulline and flesh watermelon juice contains 43.81 mg/g.[8] Rind watermelon juice contains a higher L-citrulline content compared to flesh watermelon, and L-citrulline in yellow watermelons is higher than red watermelons.[9] Nevertheless, the findings may suggest that both flesh and rind watermelon juices potentially offer similar benefits to pure L-citrulline for improving exercise performance.[10] A study by Ridwan, *et al*., showed that supplementation with 100% flesh watermelon juice improves endurance in swimming performance in rats.[8]

Another type of fruit that has a beneficial effect as nutritional ergogenics is plantain. Plantain is one type of banana fruit, contains about 31.15 g of carbohydrates and 564 mg potassium.[11] Potassium is an electrolyte that acts as a body fluid balancer, to deliver nerve impulses and muscle contractions. Potassium plays role in muscle relaxation and also promoting muscle fatigue delay.[12] A previous study by Ustafia, *et al*., showed that bananas were more effective than banana milkshakes in removing fatigue after exercise.[13] A findings by Faturochman also showed that banana was effective to prevent muscle fatigue in an anaerobic sprint.[14] The carbohydrates both in watermelon and plantain are a good source of energy during exercise, so they can be promoting fatigue delay. Increasing the amount of glycogen storage by 25-100% can be done by consuming the carbohydrates before the exercise. It can delay fatigue during exercise up to 20%.[15] Many evidence suggest that L-citrulline give a beneficial effect for improving exercise performance. However, there is little evidence to support that watermelon juices could provide such improvements. Cutrufello, *et al*., reported that acute watermelon juice supplementation appears to be ineffective in improving exercise performance.[16] Ridwan *et al*., who reported that supplementation with 100% flesh watermelon juice improves endurance in swimming performance did the experiment in the longer term to avert such drawbacks.[8] Nevertheless, this study design used acute term supplementation by considering that the nutritional ergogenics from both the fruits will give the effect in a short period. This study used rats as subjects not in athletes directly, because the formula with the best effect has not been found. This study needs many subjects to investigate which the best formula with optimal effect.

MATERIALS AND METHODS

Design, location, and time

This study was a true experimental with a post-test-only randomized controlled group design. The experiment was conducted in Food and Nutrition Research Centre Inter-Laboratory Universitas Gadjah Mada (UGM) in May 2019. Ethical research for this study was approved by the research ethics committee with reference number 2190/KEPK/V/2019.

Materials and juices preparation

The yellow watermelon was classified as Black Orange type (*Citrullus lanatus 'Black Orange'*), which was obtained from yellow watermelon plantation, Nusawungu, Cilacap, Central Java. with approximately weighing between 4.0 – 5.0 kg. The plantain banana used in this study was classified as *Musa paradisiaca Linn,* which was obtained from plantain plantation, Kalimanah, Purbalingga, Central Java. The plantain was characterized as yellow-colored, weighing 150 – 200 grams each fruit. Total glucose and potassium content from yellow watermelon and plantain in this study were measured in Food and Nutrition Research Centre

Inter-Laboratory UGM with analysis certificate number PS/157/V/2019. The total glucose contents for yellow watermelon and plantain were 5.19% and 15.40%, and the potassium was 111.921 mg/kg and 438.910 mg/kg respectively. The watermelons and plantains peeled to obtain the yellow flesh and white rind watermelons and the flesh of plantains. The 100 g watermelons (flesh and rind) and 100 g flesh plantains processed using a commercial blender to obtained combination juice. The juice is prepared freshly.

Animal models and treatment doses

Thirty-five, healthy, eight-week-old male Sprague-Dawley rats were obtained from the Food and Nutrition Research Centre Inter-Laboratory UGM. The animals were acclimatized for three days with a normal pellet diet and filtered tap water *ad libitum*. Rats were assigned into 5 groups of 7 rats: negative control (C-) group (not treated with combination juice before exercise), positive control (C+) group (no combination juice administration and no exercise), single-dose (P1) group (treated with single-dose of combination juice, 1.8 grams of solid forms diluted with water, before exercise), double dose (P2) group (treated with double-dose of combination juice, 3.6 grams of solid form diluted with water, before exercise), and single-dose with sugar (P3) group (treated with single-dose of combination juice 1.8 grams of solid form diluted with water and added with 0.27 grams of granulated sugar, before exercise). Determination of the combination juice doses administered to the rats was based on the calculation of mean fruit consumption servings number in humans. In humans, these fruits can be consumed separately as a fruit or as a juice with the addition of water. The details for determining the dose of combination juice in this study are shown in Table 1 below.

Table 1. Determination of The Combination Juice Doses

Groups	Treatment Dose	Explanation
Treatment 1 (P1)	1.8 g combination juice	In humans, equal with consumption of 50 g yellow watermelon + 50 g plantain
Treatment 2 (P2)	3.6 g combination juice	In humans, equal with consumption of 100 g yellow watermelon + 100 g plantain
Treatment 3 (P3)	1.8 g combination juice + 0.27 g granulated sugar	In humans, equal with consumption of 50 g yellow watermelon + 50 g plantain + 13 g granulated sugar

Exercise treatment

The rats were fed with combination juice as the source of energy, L-citrulline, and potassium before exercising. Thirty minutes after the combination juice was administered, the exercise was conducted. The anaerobic exercise was set as a swimming test, where the rats were drowned in a water pool and were left to swim for three minutes. Three minutes is the maximum duration of a lactic acid system in anaerobic exercise.[17] After three minutes of swimming, the rats were pulled up from the pool, and the post-exercise blood sample was taken.

Serum Analysis

The blood sample was collected immediately after exercise. Blood serum was attained through rats' orbital sinus to obtain lactic acid levels. Blood serum analysis was required to analyze lactic acid level after exercise resulted from a different dose of yellow watermelon-plantain juice administration specifically in treatment groups, and compared between treatment groups and control groups.

Data Analysis

Data obtained were analyzed using SPSS 20 for Windows (SPSS Inc., Chicago, IL). The data were presented as mean ± standard deviation (SD). One-way analysis of variance (ANOVA) followed with post-hoc LSD test were applied to identify statistical differences between groups. Statistically, a significant result was considered at a p-value < 0.05.

RESULTS

The mean body weight of rats in this study was 209 g. Since the study design only used a post-test, data from the positive control group were used as a comparison standard. Rats are considered to be due to factors that can affect lactic acid production, such as training volume or physical activity, as homogeneous. Figure 1 shows the mean lactic acid levels in the blood of rats after swimming for three minutes. As a normal standard, the results of the C (+) group lactic acid test were used when the mice received no treatment, and their blood lactic acid levels were monitored simultaneously with the other treated groups. The normal blood lactic acid levels in this study were 1.38 mmol / L, which corresponds to the normal blood lactic acid concentrations (at rest) in humans which are < 2 mmol / L.

The highest lactic acid levels were in the C (-) group, which received no juice but did a swim test. The blood lactic acid level of group C (-) was five times higher than C (+). Meanwhile, in the treated group, the highest lactic acid levels were produced by the P1 group, almost three times higher than normal levels. The P2 group scored was the lowest of the treatment groups, almost equal to the normal group. The P3 group, with the same juice dose as P1 with added sugar, produced lower lactic acid levels than P1. The addition of sugar affects lactic acid production after swimming. In general, however, the results of statistical tests showed that there were significant differences between groups of dose variations $p < 0.05$.

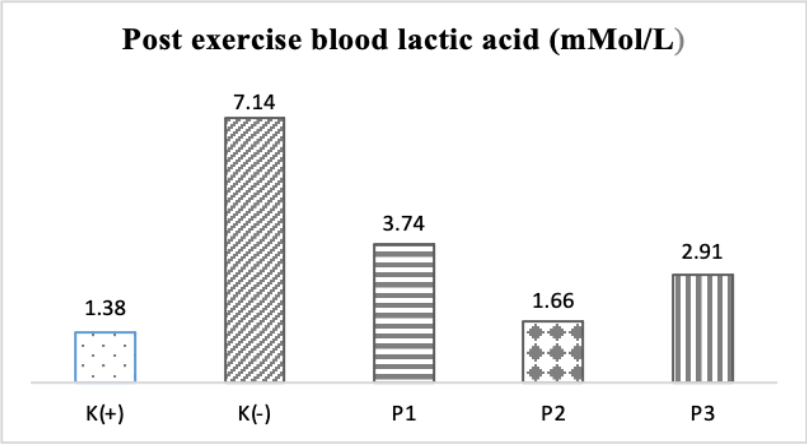

Figure 1. Comparison of mean lactic acid levels

Table 2. Mean difference lactic acid levels

Groups	Mean Difference (mMol/L)	IK 95%		*p**
		Minimum (mMol/L)	Maximum (mMol/L)	
C (+) vs C (-)	-5.76	-6.03	-5.49	0.00
C (+) vs P1	-2.35	-2.62	-2.08	0.00
C (+) vs P2	-0.28	-0.54	-0.01	0.04
C (+) vs P3	-1.53	-1.80	-1.26	0.00
C (-) vs P1	3.40	3.13	3.67	0.00
C (-) vs P2	5.48	5.21	5.75	0.00
C (-) vs P3	4.23	3.96	4.50	0.00
P1 vs P2	2.08	1.80	2.34	0.00
P1 vs P3	0.83	0.55	1.09	0.00
P2 vs P3	-1.25	-1.52	-0.98	0.00

* *Post hoc LSD test* (p<0,05)

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The largest mean difference between the groups was between C (+) and C (-), with both standard comparisons being under opposite conditions, resting and receiving a swim test. The largest inter-group difference between treatment groups was between P1 and P2. The P2 juice dose was twice the juice dose on P1 and the result was posted swimming lactic acid production was suppressed by more than 50%. The smallest difference was between groups P1 and P3. With the addition of 0.27 g of sugar, the P3 group produced less lactic acid than P1 after swimming. The addition of 0.27 g of granulated sugar has a protective effect on lactic acid production after swimming, as the addition of sugar increases the availability of glucose for energy. However, the effect was not as good as a double dose of juice.

DISCUSSION

Muscle fatigue occurs more quickly in high-intensity and short-duration sports that require rapid energy over a short period, while oxygen uptake is very limited. This condition causes the metabolism to produce a byproduct called lactic acid. Lactic acid, which accumulates, inhibits muscle contraction and causes pain. This condition is known as muscle fatigue.[18]

The results of this study show that the administration of a combination juice at different doses to Sprague Dawley rats caused a significant difference in the mean blood lactic acid levels. The main difference was in C (+) and C (-) groups, both of which were the same control group that did not receive juice, but C (-) got three minutes of swimming activity while C (+) rested. The C (+) groups as the normal inactivity standard had a blood lactic acid level of 1.38 mmol / L. This value corresponds to the average lactic acid level of normal human blood under inactivity conditions of 1-2 mmol / L.[19] Meanwhile, C (-) group as the standard for full activity, which has the three-minute swim test without prior consumption, had blood lactic acid levels of 7.14 mmol / L.

In the treatment group, the P2 group with a juice dose twice the dose of P1 had the lowest blood lactic acid levels after exercise of all treatment groups. The lactic acid levels in the P2 group were even close to normal lactic acid levels such as C (+) under resting conditions and when the person during physical inactivity. This mean was not met by P1 and P3 groups, which means that the other two treatment groups have standard blood lactic acid levels as if they were active. Thus, it can be said that the dose of P2 has a very good protective effect in suppressing the production of lactic acid, a byproduct in the formation of energy in exercise with high intensity and short time where O2 supply for the metabolism is very limited. The P2 which administered with a combined juice of up to 3.6 g / 200 g body weight of rats had higher citrulline values than the P1 dose, which only administered of 1.8 g / 200 g body weight of rats or P3 dose that the combination juice was given 1.8 g / 200 g body weight rats with the addition 0.27 g sugar.

Citrulline is a type of amino acid that plays a role in improving athlete performance by suppressing lactic acid production. Citrulline is classified as an ergogenic non-essential amino acid involved in three metabolic pathways: detoxification of ammonia in the urea cycle, synthesis of glutamine to arginine in the gut and kidneys, and synthesis of nitric oxide.[20] During the formation of ATP for energy production in intense exercise, dehydrates AMP (adenosine monophosphate) produces ammonia. Ammonia activates phosphofructokinase and facilitates the production of lactic acid.[21] Citrulline detoxifies ammonia so that lactic acid production can be controlled.[22] Citrulline accelerates the process of elimination of ammonium and lactate in plasma and other muscle metabolites.[23] Watermelon is known to have a fairly high content of citrulline, especially in the mesocarp. Seedless yellow watermelon varieties have a higher citrulline content than the other varieties. Administering 2 g of yellow watermelon mesocarp extract can reduce lactic acid production.[24]

In P1 and P3 groups, citrulline contained in the combined juice was relatively equal, as the groups which received a juice dose of 1.8 g per 200 g of rat body weight respectively. The difference was 0.27 g of sugar was added to the P3 group. The results of blood lactic acid levels of both groups after exercise showed that P3 was better than P1. It because 0.27 g of sugar was

added to the P3 group, which means that sugar absorption was increased. The addition of a simple sugar intake increases the energy source that is ready to be used in the body for activity. The availability of more ready-to-use energy sources increases the ability to perform longer activities so that muscle fatigue does not occur in a short period.[25] Previous studies have also shown a significant difference in the lactic acid levels in the blood of football players before and after given brown sugar. The treatment group showed a decrease in blood lactic acid level before and after administration of brown sugar from 11.5 mmol / L to 7 mmol / L. In the control group, there was an increase in blood lactic acid levels from 9 mmol / L to 10.8. mmol / L.[26]

This study used a combination of yellow watermelon and plantain. Juicing with a combination of two types of fruits was intended to increase nutritional value. The role of plantains in contributing to carbohydrates and high potassium. While the role of the yellow watermelon is to contribute carbohydrates and citrulline. The function of potassium as an electrolyte regulates the pH balance, the cofactor of the enzyme pyruvate kinase, Na + K + -ATPase which plays a role in energy production, synthesis of glycogen. In addition, potassium can also help train muscles and prevent muscle spasms and cramps.[12] Giving bananas before exercise greatly prevents muscle fatigue in the anaerobic phase.[27] Meanwhile, citrulline have beneficial functions accelerating metabolite waste removals such as ammonium and lactate from the plasma and the other results of muscle metabolism so the muscle fatigue does not occur quickly.[3]

The dose of combination juice was supplemented varied in each group. The doses were given according to watermelon and banana household standards consumption, 100 g each fruit. The dose was converted into mice with a conversion number of 0.018. In this study, the P2 group which received the juice dose of 3.6 g / 200 g body weight showed the lowest blood lactic acid production. It showed that the dose of combination juice made from 100 g of yellow watermelon and 100 g of plantain is most effective in suppressing lactic acid production in the blood during high-intensity short-term activities.

CONCLUSION

The combination juice of yellow watermelon-plantain affected suppressing the production of post-exercise blood lactic acid. The best dose was 3.6 g per 200 g rats body weight. It produced optimal suppression of post-exercise lactic acid production with the lowest blood lactic acid levels as if there was no activity.

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REFERENCES

1. Kemenkes RI. Pedoman gizi olahraga prestasi. Kementrian Kesehatan RI: Jakarta. 2014.
2. Fink HH, Mikesky AE. Practical applications in sports nutrition. Jones & Bartlett Learning; 2017.
3. Meneguello MO, Mendonca JR, Lancha Jr AH, Costa Rosa L. Effect of arginine, ornithine and citrulline supplementation upon performance and metabolism of trained rats. Cell Biochem Funct. 2003;21(1):85–91.
4. Curis E, Nicolis I, Moinard C, Osowska S, Zerrouk N, Bénazeth S, et al. Almost all about citrulline in mammals. Amino Acids. 2005;29(3):177.
5. Pérez-Guisado J, Jakeman PM. Citrulline malate enhances athletic anaerobic performance and relieves muscle soreness. J Strength Cond Res. 2010;24(5):1215–22.
6. Glenn JM, Gray M, Wethington LN, Stone MS, Stewart RW, Moyen NE. Acute citrulline malate supplementation improves upper-and lower-body submaximal weightlifting exercise performance in resistance-trained females. Eur J Nutr. 2017;56(2):775–84.
7. Jayaprakasha GK, Patil BS. A metabolomics approach to identify and quantify the phytochemicals in

watermelons by quantitative 1HNMR. Talanta. 2016;153:268–77.
8. Ridwan R, Abdul Razak HR, Adenan MI, Md Saad WM. Supplementation of 100% flesh watermelon [Citrullus lanatus (Thunb.) mats. And nakai] juice improves swimming performance in rats. Prev Nutr Food Sci. 2019;24(1):41–8.
9. Rimando AM, Perkins-Veazie PM. Determination of citrulline in watermelon rind. J Chromatogr A. 2005;1078(1–2):196–200.
10. Tarazona-Díaz MP, Alacid F, Carrasco M, Martínez I, Aguayo E. Watermelon juice: potential functional drink for sore muscle relief in athletes. J Agric Food Chem. 2013;61(31):7522–8.
11. Kemenkes RI. Daftar Komposisi Bahan Makanan. 2008.
12. McKenna MJ, Bangsbo J, Renaud J-M. Muscle K+, Na+, and Cl− disturbances and Na+-K+ pump inactivation: implications for fatigue. J Appl Physiol. 2008;104(1):288–95.
13. Ustafia L, Tursilowati S, Noviardhi A. Pemberian Milkshake Pisang Sebelum Latihan terhadap Kelelahan dan Tekanan Darah Atlet Sepak Bola. J Ris Gizi. 2017;5(2):41–7.
14. Faturochman, Said Junaidi AS. Journal of Sport Sciences and Fitness DI SEMARANG. 2020;5(2):104.
15. Irawan MA. Nutrisi, Energi & Performa Olahraga. Polt Sport Sci Perform Lab. 2007;1:1–13.
16. Cutrufello PT, Gadomski SJ, Zavorsky GS. The effect of l-citrulline and watermelon juice supplementation on anaerobic and aerobic exercise performance. J Sports Sci. 2015;33(14):1459–66.
17. Mooren F, Völker K. Molecular and cellular exercise physiology. Human Kinetics Publishers; 2005.
18. Williams MH. Nutrition for health, fitness, and sport. WCB/McGraw-Hill; 1999.
19. Janssen Peter GJM. Training lactate pulse rate. Oule Finl Polar ElectroOy. 1989;
20. Rabier D, Kamoun P. Metabolism of citrulline in man. Amino Acids. 1995;9(4):299–316.
21. Tomlinson C, Rafii M, Ball RO, Pencharz P. Arginine synthesis from enteral glutamine in healthy adults in the fed state. Am J Physiol Metab. 2011;301(2):E267–73.
22. Takeda K, Machida M, Kohara A, Omi N, Takemasa T. Effects of citrulline supplementation on fatigue and exercise performance in mice. J Nutr Sci Vitaminol (Tokyo). 2011;57(3):246–50.
23. Aranta Galuh Fridintya AGF. Perbedaan Efektivitas Pemberian Jus Pisang Ambon dan Jus Pisang Raja dalam Mengatasi Kelelahan Otot pada Tikus Wistar. Diponegoro University; 2011.
24. Adnyana IK, Arlinda ND, Safitri D. EFEK ANTILELAH EKSTRAK AIR MESOKARP SEMANGKA KUNING (Citrullus lanatus Thunb.) TANPA BIJI. Kartika J Ilm Farm. 2014;2(2):1–6.
25. Hairy J. Fisiologi Olahraga. Jakarta: Dirjen Dikti; 1989.
26. Abdurahman MN, Ray HRD, Ruhayati Y. Potensi Gula Merah dalam Meningkatkan Penampilan Olahraga. J Terap Ilmu Keolahragaan. 2018;3(1):24–9.
27. Kumairoh S, Syauqy A. Pengaruh pemberian pisang (Musa paradisiaca) terhadap kelelahan otot anaerob pada atlet sepak takraw. Diponegoro University; 2014.

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Effect of tomato and red guava juice on blood glucose level in overweight woman

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ABSTRACT

Background: Based on the results of Riskesdas 2018 that the prevalence of diabetes mellitus in Indonesia showed an increase from 6.9% to 8.5%. Overweight is closely related to impaired blood glucose, insulin resistance, and decreased insulin secretion. Preventive efforts that have been made using non-pharmacological treatments, such as increasing the intake of fiber and lycopene from fruits.
Objectives: This study aimed to determine the effect of tomato juice and guava juice on blood glucose levels in overweight women.
Materials and Methods: The study design is a true experiment conducted in 11subjects in treatment group and 11 subjects in control group. The subject of this study were overweight adult women 45-55 years. Blood sampling was taken in the morning, then glucose levels were measured using the GOD-PAP method (Glucose Para Amino Phenazone). We gave 600 ml of tomato juice and red guava every day for 21 days. To find the effect of juice on fasting blood glucose levels controlled by nutrient intake and physical exercise using the Repeated Measure ANOVA test.
Results: In the treatment group, there was a decrease of 3.24 mg/dl in blood glucose levels; while, in the control group, the decrease in blood glucose levels was only 0.26 mg/dl. However, we found no statistically significant differences in both groups.
Conclusions: Consumption of tomato juice and red guava reduced fasting blood glucose.

Keyword: Blood glucose; Tomato; Red guava; Women; Overweight

BACKGROUND

At the age of 45, people experience an increased risk of developing diabetes and glucose intolerance due to decreased ability of beta cells to produce insulin whose function is to metabolize glucose[1]. Females are more prone to diabetes due to estrogen. As estrogen decreases, insulin resistance begins to arise which causes an increase in blood glucose[2]. Overweight is closely related to several conditions, including impaired blood glucose, disrupted balance of glucose and insulin, and decreased insulin metabolism. Obesity causes an increase in fatty acids or Free Fatty Acid (FFA) in cells and this could lead to insulin resistance[3].

People suffering from DM will depend on drugs throughout their life. Anti-hyperglycemia drugs cause side effects such as nausea, weight gain, diarrhea, and bloated stomach which lead to non-adherence medication[4]. Therefore, we need an alternative that does not have side effects, which is by utilizing fruit with a low glycemic index. Fruit is a safe alternative because it is part of the diet.

Tomato contains an active substance called lycopene. Lycopene is an antioxidant whose ability is to resist free radicals. Lycopene can lower blood glucose by reducing insulin resistance, leading to cell tolerance to glucose increases, and excess blood sugar levels can be overcome[2].

Red guava is a fruit with high vitamin C content and pectin-type fiber. Pectin is hypoglycemic which can reduce blood glucose levels as pectin plays a role in the formation of gels in the gastrointestinal tract. Vitamin C plays a role in reducing glucose toxicity, thereby preventing a decrease in β cell mass and insulin level.

In this research, 300 grams of tomatoes and 300 grams of red guavas were processed into juice. This combination is expected to reduce blood glucose levels in overweight females aged 45 - 55 years.

MATERIALS AND METHODS

This research implemented a true experimental design with randomized pre-test and

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post-test control group design. It was conducted in RW 02 Pedurungan Tengah, Semarang. The research subjects were grouped randomly. The sample size was calculated using the hypothesis test formula on two independent groups, obtaining 11 subjects in the treatment group and 11 subjects in the control group. The subjects involved were females aged 45-55 years with a BMI ≥ of 23 kg/m^2, not taking medication, and had never been diagnosed by a doctor with diabetes mellitus. This study has been approved by Health Research Ethics Committee of Health Polytechnic of Semarang Number 113/EA/KEPK2019.

The subjects in the treatment group had their blood glucose levels measured before and after giving tomato and red guava juices. While the subjects in the control group had their blood glucose levels measured, but not receiving tomato and red guava juices. Blood sampling was carried out in the morning after the subjects were asked to fast for 8 hours. Measurement of fasting blood glucose levels was done using the GOD-PAP method (Glucose Oxidase Para Amino Phenazone). The research variables included the independent variable of tomato and red guava juice provision. Tomatoes and red guavas, each at 300 grams, were juiced together by adding 150 ml of water. The treatment group was then given the juice to consume 2 times a day, in the morning and evening, with each administration of 300 ml for 21 days. The dependent variable was fasting blood glucose.

The instruments used consisted of sample identity form, data collection form to record food intakes, product acceptability test form, juice consumption adherence form, and physical exercise form. To measure food intakes, the 2 x 24-hour food recall method was used to determine energy and carbohydrate intakes. Meanwhile, a food frequency questionnaire (FFQ) was used to determine fiber, vitamin C, and lycopene intakes. Juice consumption adherence was measured by asking the subjects how much juice was consumed. Microtoice was used for height measurement and digital weighing was used for weight measurement. To determine the effect of tomato and guava juice on fasting blood glucose levels before and after the research, and analysis was performed using the Repeated Measure ANOVA test, with $\alpha = 0.05$.

RESULTS

Table 1 shows that the research subjects were aged between 45 - 55 years. The average BMI in the treatment group was 28.26 kg/m^2 and the control group was 29.2 kg/m^2. The average fasting blood glucose levels in the treatment and control groups before treatments were 84.77 mg/dl and 85.66 mg/dl, respectively. The average fasting blood glucose levels after treatments in the treatment and control groups were 81.53 mg/dl and 85.4 mg/dl, respectively. The average energy adequacy levels in the treatment and control groups were 69.12% and 52.81%, carbohydrate intakes were 54.53% and 42.2%, fiber intakes were 10.55 g and 8.08 g, lycopene intakes were 2.42 μg and 1.21 μg, and vitamin C intakes were 57.92 mg and 45.98 mg.

Table 1. Research Subject Characteristics

Variable	*Treatment*			Control			
	Mean±SD	*Min*	*Max*	*Mean±SD*	*Min*	*Max*	*P-value*
Age	49.67±3.24	45	55	50.67±3.96	45	55	0.29
BMI	28.26±3.76	23.1	34.7	29.22±4.3	23.1	35.3	0.57
Initial GDP	84.77±13.11	63.6	103.5	85.66±13.67	66.3	114.0	0.86
Final GDP	81.53±11.46	63.6	104.0	85.4±6.66	72.5	92.6	0.31
Energy (%)	69.12±25.69	26.6	117.8	52.81±19.1	28.5	91.2	0.47
Carbohydrate (%)	54.53±20.32	25.2	90.2	42.2±13.1	28.5	91.2	0.41
Fiber (g)	10.55±2.98	4.8	14.1	8.08±2.25	4.8	11.8	0.47
Lycopene (μg)	2.42±1.04	0.8	4.2	1.21±0.51	0.8	2.5	0.10
Vit C (mg)	57.92±15.4	34.8	78.9	45.98±9.55	34.7	60.9	0.10

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Table 2. Differences in Blood Glucose Levels between Treatment and Control Groups

	Between Treatment *Mean ±SD*	After Treatment *Mean ±SD*	Difference	P-value
Control group	85.66 ± 13.67	85.40 ± 6.66	-0.26	0.533
Treatment group	84.77 ± 13.11	81.53 ± 11.46	-3.24 ± 6.85	0.438

Based on the results of analysis presented in table 2, there was a decrease in fasting blood glucose levels for subjects in the treatment group by 3.24 mg/dl and in the control group by 0.26 mg/dl. The statistical test on the difference in decreased fasting blood glucose levels obtained a p-value of 0.438 in the treatment group and a p-value of 0.533 in the control group. Therefore, it can be concluded that there is no difference in fasting blood glucose levels between the treatment and control groups before and after treatments. It is in line with research conducted by Widyasulistya (2018) where there was no significant difference between blood glucose levels before and after treatments of giving guava juice for 3 days to football athletes[5].

Table 3. Repeated Measure ANOVA Multivariate Test

Variable	P-value
Kec_Energy	0.299
Kec_Carbohydrate	0.472
Kec_Fiber	0.370
Kec_VitC	0.293
Groups	0.521

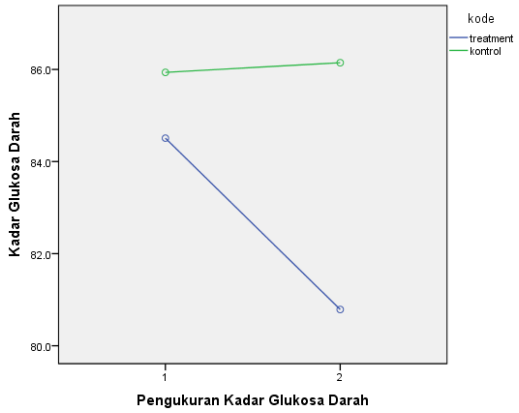

Figure 1. The difference in Fasting Blood Glucose Levels between Treatment and Control Groups

This test's results showed that there was no significant difference (p = 0.521) in blood glucose levels between the treatment group and the control group, although clinically there was a decrease in blood glucose levels in the treatment group by 3.24 mg/dl. Meanwhile, the level of adequate energy, carbohydrate, fiber, and vitamin C did not affect changes in blood glucose levels. As shown in Figure 1, there was a decrease in the treatment group after being given tomato and guava juice.

DISCUSSION

Tomato and red guava juices were given 2 times a day, in the morning and evening, as much as 300 ml per administration. This mixed juice contains soluble fiber such as pectin, lycopene, and vitamin C, with each contribution of 96% of lycopene, 78% of fiber, and 121% of vitamin C. The analysis showed that there was no effect of giving tomato and red guava juices on blood glucose levels in overweight adult females aged 45-55 years (p = 0.521). The level of energy, carbohydrate, fiber, and vitamin C intakes did not

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statistically have a significant effect on changes in blood glucose levels. The results of this research are in line with that of Ayuhapsari et al (2018) where they found that energy, carbohydrate, and fiber intakes did not affect changes in fasting blood glucose[6].

A factor that resulted in the non-significant effect of treatments in this research was the age of the subjects over 45 years. At that age, there will be an increase in blood glucose due to changes in pancreatic beta cells that produce insulin, resulting in a decreased insulin production. The decrease in insulin production results in a reduced amount of glucose that enters cells, so glucose remains in the blood vessels and causes increased blood glucose level[2].

The nutritional status of research subjects could cause less effective effects of treatments, with the research subjects' average BMI of 28.26 kg/m^2 included in the category of obesity level I. Obesity is related to conditions of impaired blood glucose, disrupted balance in glucose and insulin, and decreased insulin metabolism[3].

As seen in Table 1, the average fasting blood glucose levels before treatments in the treatment group were 84.77 mg/dl, and after treatments were 81.53 mg/dl. It showed a decrease of 3.24 mg/dl, but the decrease was still in the category of normal blood glucose level. Whereas in the control group the average fasting blood glucose levels were 85.66 mg/dl and went down to 85.40 mg/dl. There was only a decrease of 0.26 mg/dl. This showed that consumption of 600 ml of tomato and red guava juice for 21 days could reduce blood glucose levels by 3.24 mg/dl and/or maintain blood glucose levels in the normal category.

Changes in fasting blood glucose levels between the treatment and the control groups could be caused by the consumption of tomato and red guava juice. Tomato contains lycopene whose function is anti-diabetic by increasing the concentration of insulin[2]. Lycopene can lower blood glucose levels by inhibiting glucose absorption in the intestine, increasing glucose transport in the blood, stimulating glycogen synthesis, and inhibiting glucose synthesis. Lycopene prevents degenerative diseases such as diabetes through an oxidative mechanism. Lycopene binds to reactive oxygen and increases antioxidant potency, thereby reducing oxidative damage to lipids. Meanwhile, the non-oxidative mechanism of lycopene is done through gene function which improves gap-junction communication, hormone modulation, and immune response, all of which can reduce degenerative diseases such as DM[7].

Research conducted by Astuti and R (2018) revealed that provision of tomato juice to blood glucose levels in pre-diabetes as much as 200 ml for 21 days showed a decrease in blood glucose by 9.00 mg/dl[2]. Another research conducted by Yusni (2015) showed that the administration of tomato extract and mangosteen extract to diabetic mice each 50 mg/kg bb/day for seven days showed a decrease in blood glucose levels by 56.67%[7]. Research by Chairunnisa (2012) was conducted by giving tomato paste to diabetic mice as much as 62 mg for 7 days reduced blood glucose levels by 262 mg/dl with a decreasing percentage reaching 75.60%[8]. Another research conducted by Wuryaningrum (2016) on giving 250 ml of rainbow smoothies to type 2 DM patients for 10 days showed statistically significant results and decreased blood glucose levels by 43.75 mg/dl[9].

Changes in fasting blood glucose levels could be caused because of guava consumption. Guava contains water-soluble fiber, such as pectin, and vitamin C. Pectin can reduce blood glucose levels because it is hypoglycemic and its physiological function is to increase glucose tolerance in people suffering from DM[10]. Pectin plays a role in gel formation because there is a reaction of fiber with water in the gastrointestinal tract. This gel will slow down gastric emptying and the movement of food through the upper gastrointestinal tract and inhibit the mixing of the gastrointestinal contents with digestive enzymes so that there is a reduction in the absorption of nutrients in the proximal part. This inhibition affects the slow absorption of glucose, causing a decrease in blood glucose[11,12]. Pectin can envelop carbohydrate molecules so that they inhibit carbohydrate absorption and will be released slowly so that the amount of sugar that enters the blood is reduced and an increase in excess blood glucose levels can be avoided[13].

Vitamin C contained in red guava is higher than in orange, which is at 49 mg per 100 grams[14]. Vitamin C can increase insulin sensitivity and lower blood glucose levels by reducing glucose toxicity, preventing decreased beta-cell mass, and increasing the amount of insulin. These three mechanisms of vitamin C protect against organ damage in diabetes: vitamin C as an antioxidant, vitamin C inhibits intracellular sorbitol

accumulation, vitamin C reduces protein glycosylation[15].

Vitamin C plays a role in modulating insulin action in people suffering from diabetes and is associated with lowering blood glucose levels. Vitamin C can inhibit the accumulation of sorbitol caused by hyperglycemia through the polyol-sorbitol pathway. Vitamin C acts as an inhibitor of the aldolase reductase enzyme which converts glucose in cells into sorbitol, so it can prevent sorbitol buildup and reduce oxidative stress and improve endothelial function. The buildup of sorbitol in cells or tissues can cause damage or change in functions[16,17]. Research conducted by Santi (2013) revealed that the provision of red guava juice as much as 2 gr/head/day for 10 days to hyperglycemic mice resulted in decreased blood glucose levels by 164.5 gr/dl[13].

CONCLUSIONS

Consumption of tomato and red guava juice did not significantly reduce blood glucose levels, with a p-value > 0.05. Consumption of tomato and guava juice can be used as part of a daily diet to control blood sugar within the normal range.

REFERENCES

1. Betteng R, Pangemanan D, Mayulu N. Analisis Faktor Resiko Penyebab Terjadinya Diabetes Melitus Tipe 2 pada Wanita Usia Produktif Dipuskesmas Wawonasa. J e-Biomedik. 2014;Volume 2,(2):9.
2. Astuti YD, Murwani H. Pengaruh Pemberian Jus Tomat Terhadap Kadar Glukosa Darah Pada Prediabetes. J Nutr Coll. 2013;2:111–7.
3. Nabila R, Widyastuti A, Murbawani EA. Hubungan Lingkar Pergelangan Tangan Dengan Kadar Glukosa Darah Wanita Obesitas Usia 40-55 Tahun. J Nutr Coll [Internet]. 2018;7(2):92–9. Available from: http://ejournal3.undip.ac.id/index.php/jnc/
4. Eliana F. Penatalaksanaan DM Sesuai Konsensus Perkeni 2015. Satelit Simp. 2015;1–7.
5. Widyasulistya R, Rahmawati AY, Aristiati K. Pengaruh Pemberian Jus Buah Jambu Biji (Psidium Guajava L) Terhadap Kelelahan Otot Anaero. J Ris GIZI. 2018;40–7.
6. Ayuhapsari N, Wijaningsih W, Jaelani M. Efektifitas Pemberian Buah Apel Manalagi dengan Dosis Bervariasi terhadap Penurunan Kadar Glukosa Darah pada Penderita Diabetes Mellitus tipe II di Kelompok PROLANIS Puskesmas Genuk Kota Semarang. J Ris GIZI. 2018;13–8.
7. Yusni, Akbar IB, Rezania, Fahlevi R. Penurunan Kadar Gula Darah Akibat Pemberian Ekstrak Manggis (Garcinia mangostana) dan Tomat (Lycopersicum esculentum Mill) pada Tikus Diabetes Blood Sugar Levels Reduction by Mangosteen (Garcinia mangostana) and Tomato (Lycopersicum esculentum Mill. J Glob Med Heal Commun. 2017;5(1):57–63.
8. Chairunnisa R. Pengaruh Jumlah Pasta Tomat Terhadap Penurunan Kadar Gula Darah Pada Mencit Diabetes. J Teknol Ind Pertan. 2012;1–12.
9. Wuryaningrum E, Jaelani M, Sunarto. Efektifitas Rainbow Smoothies Terhadap Penurunan Kadar Gula Darah Pasien DM Tipe 2 Di Klub Senam Prolanis Bina Sehat Mandiri Cepu. J Ris GIZI. 2016;1–6.
10. Sunarti. Serat Pangan Dalam Penanganan Sindrom Metabolik. Yogyakarta: Gajah Mada University Press; 2018.
11. Lianah, Tyas DA, Armanda DT, Setyawati SM. Aplikasi Umbi Suweg (Amorphophallus campanulatus) sebagai Alternatif Penurun Gula Darah pada Penderita Diabetes Mellitus. J Biol Appl Biol. 2018;1(1):1–12.
12. Saputro PS, Estiasih T. Pengaruh Polisakarida Larut Air (PLA) Dan Serat Pangan Umbi-Umbian Terhadap Glukosa Darah. J Pangan dan Agroindustri. 2015;3(2):756–62.
13. Santi DA. Efek Jus Buah Jambu Biji Terhadap Gangguan Toleransi Glukosa Pada Tikus Putih Jantan Akibat Efek Samping Deksametason. J Ilm Mhs Univ Surabaya [Internet]. 2013;2(1):1–19. Available from:

http://webhosting.ubaya.ac.id/~journalubayaac/index.php/jimus/article/view/169

14. Astuti EP, Ismawati, Suwarlim T. Uji Efek Penurunan Glukosa Darah Air Perasan Buah Jambu Biji (Psidium guajava L) pada Mencit (Mus musculus) yang diberikan Glukosa per Oral. J Ilm Kesehat. 2009;3(2):107–13.

15. Santosh HN, David CM. Role of ascorbic acid in diabetes mellitus: A comprehensive review. J Med Radiol Pathol Surg [Internet]. 2017;4(1):2–3. Available from: http://www.jmrps.net/eJournals/ShowText.aspx?ID=79&Type=FREE&TYP=TOP&IN=_eJournals/images/JPLOGO.gif&IID=13&Value=1&isPDF=YES

16. Dini CY, Sabila M, Habibie IY, Nugroho FA. Asupan Vitamin C Dan E Tidak Mempengaruhi Kadar Gula Darah Puasa Pasien DM Tipe 2. Indones J Hum Nutr. 2017;4(2):65–78.

17. Ermawati T. Hubungan kadar glukosa darah dengan Pertumbuhan A. actinomycetemcomitans Pada Tikus Diabetes Melitus. J Kedokt GIGI. 2012;9(1):19–22.

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The implementation of nutrition improvement programs for underweight children, wasting and stunting in the Department of Health, Central Buton district, Southeast Sulawesi

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ABSTRACT

Background: Health status can affect the Human Development Index (HDI) of a country. To improve the quality of human resources, the Indonesian Government has applied various policies, such as the nutrition improvement program, since there are still lots of toddlers suffering from underweight, wasting dan stunting. Even one out of three toddlers in Indonesia were detected stunting.
Objectives: This study described the implementation of a nutrition improvement program for toddlers in the Central Buton District Health Office.
Materials and Method: This was a qualitative study that involved ten informants. Three of those informants are nutritionists in the District Health Office, Head of Public Health Department, Head of NutritionDepartement. Also, two nutrition workers in primary healthcare centers and two mothers of malnourished toddlers.
Results: These findings showed that in terms of input, trained human resources on nutrition were lacking, and the budget for the nutrition improvement program was inadequate. While, in the process, all implementors had done very well, although they still had no collaboration across sectors. In the output aspect, the health status of underweight, wasting, and stunting toddlers improved. Also, monitoring and evaluation were conducted on toddlers registered at the integrated service posts in 2018.
Conclusions: There were still constraints on the input, process, and output aspects, even though there was an increase in the nutritional status of children under five, but nutrition problems for children under five in Central Buton Regency were still high.

Keywords: Nutritional improvement program; Toddlers; Implementation

BACKGROUND

Nutritional problem is a problem in the life cycle, starting from pregnancy, infants, toddlers, teens, to the elderly. The nutritional problem can occur in all age groups, even nutritional problems in a certain age group will affect nutritional status in the next life cycle (intergenerational impact).[1]

Indonesia nowadays is still faces nutritional problems such as underweight, wasting, and stunting, this can affect the quality of human resources, because growth constraints during toddlers have the potential to experience non-communicable diseases in adulthood.[2] According to the President of the Republic of Indonesia, the fulfillment of nutrition is one of the best long-term investments that can be made, apart from requiring skilled human resources, of course, Indonesia also needs healthy human resources. The health status of a nation can affect the human development index (HDI), therefore the government is committed to making efforts to overcome various nutritional problems by issuing various policies on nutrition improvement.[3]

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Toddler age is a period where the process of growth and development occurs very rapidly, with high activity and learning changes. At this time the child's brain develops rapidly up to 80%, so it requires sufficient nutrient intake in both quantity and quality, if the nutritional intake is not fulfilled, the physical and intellectual growth of toddlers will experience disruption, as a result, toddlers become lost generation, and can adversely affect the country due to the lack of quality human resources.[4]

In 2017, the number of children under five in Indonesia is around 22.4 million every year, there are at least 5.2 million women in Indonesia who are pregnant, so the average number of baby-born every year is 4.9 million, and baby with short birth conditions have the potential to the increase in the number of children under five with stunting, whereas in 2018 there were 17.8% of children under five suffering from malnutrition, 12.7% among under-five children with malnutrition, 12.7% of whom were short, for that intervention to reduce stunting would reduce under-nutrition Malnutrition).[5]

Stunting is a chronic nutritional problem caused by a lack of nutritional intake for a long time, characterized by a lack of height from the standard children in general, this occurs since the child is still in the womb until the age of 2 years or the first 1000 days of life and the child is stunted after 2 years of age cannot be changed anymore so what can be done is to maximize the potential for brain development. In contrast to stunting, malnutrition takes place in a shorter time, that is, when the child experiences normal growth up to a certain age, then there is a change in diet which causes the toddler to no longer getting enough intake, as a result, the child loses weight in a short time but the recovery also fast. Apart from stunting and malnutrition, There is also malnutrition, namely children under five who are underweight children of their age. Another nutritional problem is obesity or excess nutrition. Nutritional status is an important indicator for the health of children under five whose physical impact is measured anthropometry and categorized based on WHO standard with index weight/age, height/age, and body weight/height.[6]

The problem of child nutrition in Indonesia is very worrying because 1 in 3 Indonesian children is stunted, nationally the proportion of stunting among toddlers is 37.2% in 2013 to 30.8%, in 2018, the prevalence of under-nutrition is 19.6% years 2013 to 13.8% in 2018, the prevalence of malnourished toddlers is 5.7% in 2013 to 3.9% in 2018, the prevalence of underweight children is 12.1% in 2013 to 10.2% in 2018 while obese toddlers are 11, 9% in 2013 to 8.0% in 2018, this is what then causes Indonesia to face a double burden. Although nationally from 2013 to 2018 there has been a decrease in the prevalence of nutritional problems in children under five, there are still gaps between provinces, especially the problem of stunting.[7]

The prevalence number of stunting at the provincial level is still very high, where 2 provinces have a stunting prevalence >40%, 18 provinces have a stunting prevalence of 30-40% and 23 provinces including Southeast Sulawesi have a stunting prevalence of 20-30%, and only DKI Jakarta has a stunting prevalence < 20%. Based on the results of monitoring the national nutritional status of Southeast Sulawesi in 2017, the highest stunting problem was in Central Buton Regency, namely 48.8% under five, 25.9% underweight, and 13.3% underweight. Also, the frequency of visits to children under five to the posyandu is decreasing along with the increasing age of the children, from 12,060 the number of children under five, with the coverage of under-fives weighed as much as 8,786 (73%) and 3,247 (27.14%) under-fives whose body weight is not monitored.[8]

Toddlers as the assets of our future (nation) must receive optimal nutritional care and attention as a series of individual and community nutritional needs through prevention, improvement, healing, and recovery efforts carried out in the community and health service facilities, including involving related sectors.[9] The central government gives authority to the provincial government which then the provincial government delegates to the city/regency government to be responsible for implementing efforts to improve nutrition. Based on the results of the field study, Central Buton is one of the districts in Southeast Sulawesi which is responsible for implementing the nutrition improvement program. The programs that have been implemented include monitoring the growth of toddlers at the health center, providing additional food, and tracking cases of toddler nutrition, this is done based on the regulation by the Indonesian Minister of Health no. 23 of 2014, to improve food consumption patterns following balanced nutrition improving behavior awareness of nutrition, physical activity, and health.[10,11] Based on this situation, the authors are interested in describing the implementation of the toddler nutrition improvement program which consists of early detection of the status of children under five years old in the

Central Buton District through system elements consisting of Input, Process, and Output.

MATERIALS AND METHODS

This research is an observational study with a qualitative descriptive design, which was conducted in September 2018 - August 2019. The subjects of this study were nutrition staff and mothers of malnutrition children under five years old in the work area of the Health Office of Central Buton District, Southeast Sulawesi Province. The selection of informants was carried out by using the purposive sampling technique by observing. Data collection methods were obtained through in-depth interviews, observation, and documentation of 10 informants consisting of 3 main nutrition staff informants (IU1, IU2, IU3) of the Health Office, and data validity was carried out on 7 triangulation informants consisting of the Head of the Department Health (IT1), Head of Public Health (IT2), Head of the Family Health and Nutrition Section (IT3), 2 Nutrition Implementers of Puskesmas (IT4, and IT5), and 2 mothers under five years old with malnutrition (IT6, and IT7). The data collected includes system elements in a program policy consisting of input elements which include human resources, infrastructure, and funds used in implementing the nutrition improvement program for children under five years old, especially monitoring the nutritional status of toddlers, next is the process element which includes planning, organizing, implementing, and monitoring evaluation. In addition to the two elements of input and process, there is also a third element, namely output as a reference to determine the success of the nutrition improvement program for children under five at the Health Office of Central Buton District, Southeast Sulawesi Province, where the nutritional status of children is assessed based on the Body Weight (BW) / Age (A) index which is categorized as undernutrition (Underweight), good nutrition and more nutrition. Body Height (BH) / Age (A) children were categorized as very short, short, and normal. Furthermore, the weight/height of children is categorized as very thin, thin, normal, and obese. This is based on the z-score according to the standard deviation value of growth according to the World Health Organization (WHO). Data analysis techniques in this study were carried out with data collection, data reduction, data display, and conclusion.

RESULTS

Central Buton Regency is one of the regencies in Southeast Sulawesi which consists of 7 districts. The Health Office is one of the elements implementing government affairs that is tasked with helping the regent carry out government affairs, especially in the health sector, this is stated in the Central Buton Regent Regulation No.13 of 2018 concerning the Position of the Organizational Structure, Duties and Functions and Work Procedure of the Health Service of Central Buton Regency. The Health Office consists of several fields, one of which is the public health sector, including a section of nutrition and family health which is responsible for running community nutrition improvement programs and carrying out coaching and cooperation with health centers in implementing community nutrition programs. maternal and child health services as well as other tasks related to family health and community nutrition. In this study, the authors focus on discussion with several aspects by a systems approach to be able to get an overview of the implementation of early detection programs and monitoring of the nutritional status of toddlers because a program will run well if it meets the target input, process and output indicators.

Input

1. Human Resources

The results of the interview on the implementation of the nutrition improvement program for children under five years old at the Health Office of Central Buton District based on the input element seen from the perspective of human resources (HR), that those involved in the nutrition improvement program were the Head of the Public Health Sector as the person in charge of the program, the head of the Family Health and Community Nutrition section. A program manager, and nutrition staff serving as report compilers and field technical advisors, and health center nutrition implementing staff (TPG) as executors of program activities. Human resources are people who are responsible for and coordinate the implementation of a program. Following are the results of interviews about human resources:

Based on the results, it was found that the nutrition staff of the Health Office was 3 people, 2 were civil servants (PNS), but one of them had never attended training on nutrition because only 6 months had worked as a nutrition staff at the health office and 1 person was still as an intern, the head of the family health and nutrition section that has an educational background (Diploma/DIII) in Midwifery with a long term of 4 years in the health department. The head of the public health sector has an educational background (Diploma/DI) Nutrition department, has served for 2

years, and the Head of the Health Office with a 2 years term. Nutrition staff at the Health Office can be said to be still lacking because at one time the nutrition staff who were still as an intern to become the person in charge of the nutrition improvement program, and the nutrition staff could get over-work. Besides, there is nutrition staff who have never attended training, especially training on nutrition during their time as nutrition staff at the Health Office.

IU1: the staff is still lacking, because we are only three of us, 1 is still an apprentice, but has worked 4 years and has attended training on nutrition, and 2 people are civil servants (PNS), but one of them has only worked for 6 months and has never attended any training while working at the health office, especially regarding nutrition, so sometimes we feel overwhelmed if suddenly there are reports of cases of malnutrition at the same time from various sub-districts, and the three of us and have to do tracking, some are doing intervention, field technical guidance and making nutrition reports to be accountable to during the evaluation meeting.

IT3: actually still lacking, because usually the person holding and responsible for one of the programs is an apprentice staff, as well as providing field technical guidance.

So it can be concluded that the nutrition staff at the Central Buton District Health Office in terms of quality and quantity is still not under the Minister of Health Regulation No. 26/2013 concerning the Implementation of Work and Practices for Nutritionists in Service Centers in section 17 states that nutrition workers in implementing nutrition services in health service facilities have the authority to participate in education, training, research and development of nutrition services.[12]

2. Facilities and infrastructure

Based on the results of interviews and observations of facilities and infrastructure, there were no obstacles in the provision of infrastructure because, in terms of the procurement of all supporting facilities, they were well maintained and available at the Health Office and the Puskesmas. The tools available consist of anthropometric tools, toddler weight scales, writing instruments, digital weighing devices, and height measurement tools, KMS (public health) books, recording and reporting forms, technical instructions for program implementation, additional food and medicines (vitamin A), which was available at the posyandu. Also, the District Health Office validates health service facilities based on a predetermined operational permit. Validation is done by comparing the suitability of the condition of the medical equipment facilities and infrastructure needed in the field and those available in health service facilities. Following are the results of interviews with key informants and triangulation.

IU2: for the too-provision by the government (Dinas) then distributed to each puskesmas, however, there must be a report if the puskesmas needs/changes equipment, and if food for toddlers is malnutrition or lacking, usually the Health Office provides instant food such as biscuits, while the puskesmas works together Posyandu cadres provide local food, porridge, eggs and so on, and there are no obstacles in the provision of facilities and infrastructure at the Health Office.

IT2: anthropometric equipment is usually the agency that provides according to the needs of the puskesmas, and so far there have been no obstacles in the procurement of sarpras because there is always coordination and validation of medical devices in health services between the office and the puskesmas.

3. Fund

One of the components of the resources needed in organizing a health program is health financing or funds. Based on the results of interviews from the three nutrition staff at the Health Office, it was stated that the nutrition improvement program had been budgeted for in the Central Buton Regency (APBD), but because of the reduction or rationalization of the APBD, not all activities from the nutrition improvement program received a budget, such as socialization on nutrition problems for children under five years old such as stunting. the stakeholders who have an important role in the success of the program cannot be done because the funds have not been programmed and are not sufficient. Following are the results of interviews with key informants and triangulations.

Process

1. Planning

Planning is the basis of the process of implementing a program, so it must be formulated and conveyed so that the results are following the desired objectives.

The planning process for the nutrition improvement program for children under five at the Health Office of

Central Buton District consists of human resource planning, implementation plans, budget plans, and monitoring and evaluation plans. Following are the results of interviews with informants.

Based on the results of the interview and data from budget planning book-keeping that there is a harmony between the statements of the main informant and the triangulation informant, it can be said that planning on the implementation of the nutrition improvement program for children under five years old has been carried out by the Health Office of Central Buton District so that program implementation can run well and achieve maximum results.

2. Organizing

Organizing is the process of preparing an organizational structure under the goals of the organization, its resources, and the environment. Organizing is very necessary to facilitate the implementation of programs that have been previously formulated in planning. The organization of the nutrition improvement program for children under five years old at the Health Office of Central Buton District is described in several stages, namely the division of tasks, the appointment of a coordinator, and the activities to be carried out. Following are the results of interviews with key informants and triangulation

IU3: for the organizational structure, the implementation of the nutrition improvement program at the health office is in accordance with the organizational structure of the head of the service, in charge of the community health sector, coordinating the family health and nutrition section and nutrition staff as executors and monitoring whether or not the program is running at the puskesmas level because it is the party fully implementing it puskesmas, assisted by posyandu cadres because the target is toddlers with nutritional problems. The activities are monitoring the growth of children under five, providing additional food, and nutrition counseling. Most of the activities are carried out at the posyandu, besides that, the puskesmas collaborates with villages/sub-districts to provide additional food so that nutritional problems such as malnutrition, malnutrition and stunting are reduced.

IT4: because the service as a monitor is not the implementer, so we always involve the puskesmas in its implementation, starting with the posyandu cadres, who report a problem to the puskesmas, then the puskesmas reports the problem to the health office, the office makes an activity plan citing the problem, then then reported to the government, in order to get support from both material and non-material.

3. Implementation

An implementation is an act of striving for all members involved in the program to achieve predetermined goals by their respective main tasks in planning. In the program for the implementation of nutrition improvement activities in the working area of the Health Office of Central Buton District, monitoring the growth of children under five at the posyandu, providing additional feeding for toddlers, and tracking cases. Following are the results of interviews with key informants and triangulation

IU1: It is still lacking because the nutrition improvement program has a variety of activities, so we are smart about using it, if the priority is usually we still try to do it with these funds, but we cannot do socialization so that we cannot work together with other sectors. outside of health to solve nutritional problems under five years old.

IT2: It is enough but there is still more supervision for funding program implementation, because there is a reduction or rationalization of the APBD Budget.

IU1: this nutrition improvement activity is carried out to reduce nutritional problems and improve nutritional status, so it must involve many people such as the head of the public health, head of health and nutrition, nutrition staff, health center nutrition officers, medical personnel, cross-program medics and posyandu cadres, all sources the power to work together to improve the nutritional status of toddlers, such as pmt in posyandu, toddler nutrition screening or toddler nutrition tracking, For our own budget plans, for example for next year's budget, we must input this year's report, even then the funds we include in the activity report, are usually not suitable with the funds we get, then again we determine priority activities then the budget is put in dpa, while monitoring is carried out every quarter.

IT1: us and the puskesmas, and the community are trying to coordinate so that the implementation of nutrition improvement programs and other programs runs well according to the desired results

IT3: nutrition staff as the person in charge of the program in the department, nutrition implementing staff at the health center in charge of the puskesmas level, cadres, medical personnel, environmental health, for budget planning, the range I forget to look at later in the bookkeeping, because it is the nutrition staff who compile the budget. We plan to evaluate the evaluation once a year, because we also do monitoring every three months, so from monitoring we can correct any mistakes in implementation.

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From the interview result with the nutrition staff as the main informant (IU1) and health center nutrition executives as triangulation informants (IT4), it was stated that the implementation of monitoring of the nutritional status of children under five was carried out guided by posyandu cadres under the auspices of the puskesmas. In addition, the next activity is the selection of cases of malnutrition, along with the results of interviews with nutrition staff at the health office (IU2), health center nutrition staff (IT4), and mothers of malnourished children under five years old (IT7). In addition to monitoring at the posyandu, there was also a screening to find out malnourished toddlers at the posyandu, puskesmas, and even home visits intending to find cases of malnutrition under five years old by measuring body weight according to age (BW/A) and weight according to height (BW/BH), as well as the presence of clinical signs such as abdominal disease. Also, the program for the improvement of nutrition for children under five is providing additional food for children under five. Following are the results of interviews with nutrition staff from the health office (IU3), TPG nutrition at the puskesmas (IT5), and mothers of malnourished children under five (IT7).

IU1: For nutritional status monitoring, it is done at the posyandu, if the office is just waiting for a report from the puskesmas if cases of malnutrition are found, so we only monitor what the puskesmas do.

IT4: We do monitoring the nutritional status of toddlers with cadres, like weighing them, because from weighing we can know whether there is a nutritional problem or not, but for malnutrition, we monitor it once a week by making home visits for 3 months.

4. Evaluation Monitoring

Monitoring and evaluation are carried out to determine performance in program implementation and to find out how the achievements of activities that have been implemented. The evaluation referred to in this paper is the evaluation and monitoring by the health office of the puskesmas as the field implementer in the nutrition improvement program for children under five at the Health Office of Central Buton District. Following are the results of interviews with key informants and triangulation informants.

From the statement of the nutrition staff as the main informant (IU1) and the head of the health department as the triangulation informant (IT1), that monitoring is carried out to determine the course of an activity, to prevent and minimize problems in the field, apart from monitoring the Health Office also evaluates the implementation of nutrition improvement programs. Following are the results of interviews with key informants and triangulation informants.

IU2: usually also done during the inspection at the posyandu, at the puskesmas, and even at home visits to find cases of malnutrition under five years old, usually due to lack of nutritious food intake and congenital diseases.

IT4: We conduct home visits if there are reports of cases of malnutrition, from posyandu cadres, for the counseling to be conducted every two months, even nutrition cases can also be obtained from the puskesmas, when a child being examined for health at the puskesmas is suspected of suffering from malnutrition.

IT7: So every time you weigh it at the clinic, your body weight does not increase, sometimes it goes down from the previous scale, you were also sick and then you were taken to the health center.

IU3-DKK: pmt is given on the guidance by cadres who work closely with the health center nutrition officers, but the materials are from the office such as biscuits, then the service distributes them to each puskesmas, later the puskesmas will be distributed at the posyandu, but usually for the posyandu we always recommend pmt in the form of local food.

IT5-PKM: after giving the pmt, we usually conduct counseling first to mothers of toddlers with nutritional status problems, to always provide food that contains energy and protein, as well as for the pmt that we provide if possible are sufficient for children who suffer from nutritional problems every day, and do not eat it by his brother or other people. Because pmt is only given to children who have nutritional status problems such as malnutrition, malnutrition, and stunting. But for toddlers with malnutrition cases once a week we check and monitor the child's weight at home for 90 days, besides that we are very helpful because the village is also involved in holding pmt.

IT6-IB: Every week the health worker visits the house, the child's weight is continuously measured, and given food, biscuits, packaged baby porridge, milk.

From the interview's result with nutrition staff at the Health Office as the main informant (IU3) and the head of the family health and nutrition section as triangulation informants (IT3), it was stated that the evaluation at the Health Office of Central Buton District was carried out to determine the performance achievements of activities.

DISCUSSION

1. Human Resources

Human resources (HR) is an important aspect of implementing a program. Based on the results of the research, in carrying out the nutrition improvement program for children under five, it is carried out by the public health sector, especially the family health and nutrition section of the Health Office of Central Buton District, but in its implementation, it involves the puskesmas and cadres, who are tasked with carrying outweighing activities for children under five at posyandu, providing additional food, tracking the nutrition of children under five, as well as other activities related to nutrition problems. Also, the results of the research show that the Health Office has limited human resources in the nutrition improvement program, both in terms of numbers, where there are only 3 nutrition staff, namely 2 civil servants (PNS), and 1 apprentice staff. If this is not done yet, it can become an obstacle in implementing the nutrition improvement program for children under five at the Health Office of Central Buton District.[12] Based on previous research, there was a lack of health personnel resources at the Health Office of Central Buton District, especially nutrition staff, and there were still health centers that did not have nutrition workers, as well as inadequate access to roads so that they encountered obstacles in both the networking and tracking malnutrition activities.[13]

IU1: for field monitoring we usually do it by monitoring the results of reports from the puskesmas, from here we can see the progress of activities in the field, and if there are obstacles, while for monitoring at the service level we usually hold a meeting every quarter to find out about program progress and solve problems. which is a barrier to implementation

IT1: supervision is carried out in stages, from the head of the department, head of the field, head of sections and programmers, so that each of them reports the course of activities and the results obtained, and is reported at the periodic meeting every quarter.

IU-3: evaluation is carried out once a year, namely at the end of the year, if there are things that have not been achieved then we will go to the field to see, what are the obstacles, such as yesterday's coverage of under-fives weighing is still below the national target, so we are working with the health center to carry out a sweeping of under-five weighing each village / kelurahan in their respective puskesmas area, by mobilizing cadres to take notes and toddlers who do not participate in posyandu, so that at the next posyandu they can attend the posyandu

IT3: usually at the end of each year we hold an evaluation meeting to find out the results of the implementation of the program that has been running for almost a year, and compare whether the results obtained are on target, and take corrective action if the results are not on target, so that the following year can reach the target .

2. Facilities and infrastructure

Apart from competent human resources, supporting facilities and infrastructure are also needed in the implementation of nutrition improvement program activities for children under five. Based on the results obtained, the facilities and infrastructure for the nutrition improvement program for children under five in the working area of the Health Office of Central Buton District are complete, both at the puskesmas and posyandu, such as anthropometric tools, tables for posyandu implementation activities, besides that the posyandu also involves the role of the village in providing materials local food for supplementary feeding (PMT) in the form of eggs and green bean porridge, this is very influential in the success of the implementation of the nutrition improvement program for children under five. Although the number of active posyandu in Central Buton District decreased from 91 posyandu in 2017 to 85 (62%) active posyandu in 2018, However, the level of awareness of mothers to invite their children to posyandu increased from 73% in 2017 to 85.7% in 2018, this can be a picture of the maximum implementation of several priority posyandu programs, especially nutrition improvement programs. In line with previous research that posyandu is expected to be able to organize five priority programs, namely maternal and child health, family planning, nutrition improvement, immunization, and diarrhea prevention. Availability of adequate facilities and infrastructure, optimal management, and utilization can help achieve the success of a program. In line with previous research that posyandu is expected to be able to organize five priority programs, namely maternal and child health, family planning, nutrition improvement, immunization,

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3. Fund

After competent human resources and complete infrastructure are needed, funds are also needed to support the implementation of a policy or program, because it will become complicated when realizing policy objectives when the funds needed are not sufficient to finance the entire series of activities of a program. In the implementation of the program to improve malnutrition, malnutrition, and stunting at the Health Office of Central Buton District, it is known that some program implementers have stated that the funds are not fully sufficient to carry out innovative activities such as cross-sector socialization, making posters and campaigns on improving nutrition, especially stunting toddlers. Based on secondary data, funds for the nutrition improvement program for children under five at the central Buton District Health office are around Rp. 10.100,000.00 in 2018, the lack of funds could be one of the causes for not maximizing the achievement of health development targets in the Central Buton District. Following previous research, although the availability of health service Facilities continues to increase, if this availability is not supported by the quality of health service in terms of human resources and financial support, it can result in the implementation of policies/programs being hampered in achieving goals because insufficient funds will affect the quality implementation of programs in the community.[15]

4. Planning

Based on the previous research, planning must be clear, both in terms of budget, human resources, and implementation targets according to the target, such as the activity plan for the empowerment team for family welfare is always synergized with the work plan and budget for regional work units.[16] The Health Office of Central Buton District has established a concept to minimize the problem of nutritional status in toddlers, especially stunting, by determining the structure of the program implementation where the person in charge of the program is the nutrition staff at the health office, and the person in charge at the puskesmas level is the puskesmas nutrition officer. In this case, the health office nutrition staff is tasked with providing technical field guidance and making reports to be accountable to the section head, while the puskesmas is in charge of executing activities and goes directly to the community, together with health or community cadres who have been empowered and become partners for the puskesmas as a community mobilizer around them to care about the health and take part in the utilization of the health services provided, Also, other cross-programs are also involved, such as medical personnel, paramedics, environmental health and community health centers, but this has not been involved in other sectors. The target in implementing the program is toddlers with nutritional problems, as for the target plan, which is to reduce underweight toddlers to 5.3%, wasting toddlers 2.7%, and stunting toddlers 14% in 2019.

For budget planning is prepared a year before the implementation of the program carried out by the nutrition staff of the health office then the budget activity plan (RKA) is submitted to the government, usually the budget that comes out is less than the planned budget, after which the budget is reported in the form of a budget utilization document (DPA).) by the nutrition staff of the Health Office of Central Buton District. The Monitoring Plan will be carried out every quarter and evaluation is carried out once a year, namely at the end of the activity or towards the end of the year. To get maximum activity results, planning must be formulated clearly so that the process runs systematically, in other words, managerial planning will provide a comprehensive perspective on the activities to be carried out, both resources, time, or implamentation.[17]

5. Organizing

Based on the results of the research on the organization of the nutrition improvement program for children under five at the Health Office of Central Buton District, it is described in several stages, namely the division of tasks, the determination of the coordinator, and the activities to be carried out. The nutrition improvement program for children under five at the Health Office of Central Buton District is carried out following the existing organizational structure at

the Health Office and in its implementation, it involves various parties, both from the scope of the health office and from the scope of the village / kelurahan. The nutrition improvement program that is implemented as the policyholder is the Head of the Health Office of Central Buton District, then the authority for its implementation is handed over to the public health sector which houses 3 sections, namely, the family health and nutrition section, the health promotion section, and the environmental health section.

Then regarding the division of tasks and responsibilities in implementation, it is entirely up to the puskesmas because there are already nutrition officers or regional supervisors whose job is to monitor the development of nutrition in their area. Based on the policy of the Health Office of Central Buton District government that all health workers at the health center, whether midwives, nutrition workers, or other health workers must cooperate in efforts to treat cases of nutrition in children under five, both in the form of screening and tracking, but the Health Office has not been able to cooperate with other people. other sectors/stakeholders, due to the lack of funds for socialization and advocacy.[18]

6. Implementation

The implementation of the program is one indicator of the success of a program because the success of the program will be obtained if the planned program can be implemented properly. The implementation of the nutrition improvement intervention program for malnourished children under five, malnutrition and stunting at the Central Buton district health office is by monitoring the nutritional status of children under five at the posyandu, screening for cases of malnourished children under five, and providing additional food for malnourished toddlers. From the results of the study, it was found that growth monitoring at the posyandu consisted of weighing children under five, measuring the height of toddlers which was carried out every month by posyandu cadres together with puskesmas officers, consisting of midwives, nutrition implementing officers, and nurses, giving vitamins and medicines, supplementary feeding for toddlers, as well as monitoring the growth of children under five with stunting, wasting, underweight to prevent and improve the nutritional status of children under five. Growth monitoring is a follow-up to policies and programs at the community level so that people have the power and efforts to address health problems so that nutritional problems can be resolved properly. Apart from that, the selection is also carried out to trace cases of malnourished toddlers, so that all mothers of toddlers routinely take their children to the posyandu. The implementation of screening is carried out in two stages, the first is that the screening is carried out every two months at the posyandu, secondly, the screening is carried out at the health center when the toddler visits the health center to have his disease checked and the health worker knows that the toddler is suffering from malnutrition

To fulfill and improve the nutritional status of children under five, nutrition officers together with posyandu cadres provide additional food for toddlers. The categories for under-nutrition and stunting children were given additional food in the form of biscuits which were given by cadres at the posyandu, while toddlers with cases of malnutrition were given factory food, biscuits, milk, eggs, and local food if there was assistance from the village / kelurahan. as well as educating mothers of toddlers on providing nutritious foods both at the community center at their home. Apart from that, nutrition workers at the health center, as well as from the local government office, also conduct home visits to malnourished toddlers every week for 3 months to find out the progress and monitor the growth of toddlers by measuring BH, BW, Lila, and head circumference of toddlers, as well as monitoring the environment where toddlers live. [19]

7. Evaluation monitoring

Monitoring evaluation is very necessary so that the implementation stage can run well and on target according to the program plan. Based on the results of the study, it was found that in the nutrition improvement program for children under five, the health department also carried out a monitoring stage which was carried out every quarter, monitoring was carried out by the health office based on the activity report of the puskesmas because from these reports the health department nutrition staff could find out and supervise the program activities and prevent any deviations in the field. If irregularities are found during supervision, the service office can make corrections as soon as possible so that activities can return to the predetermined path. The monitoring stages are also leveled from the posyandu level, the puskesmas level, and the health department level. Monitoring by the Health Office can be carried out in 2 stages, namely direct visits or by observing the results of the activity report. Monitoring is the process of observing activities on an ongoing basis to minimize any deviations because

if there are deviations, modifications or changes are needed so that implementation remains on the planned path.[20,21]

In addition to monitoring, the Health Office also conducts an evaluation every once a year, the evaluation is carried out at the end of the year, but if there is an extraordinary event (KLB) in an area, we usually do an evaluation as well. Evaluation is carried out in meetings and meetings at the Health Office, where at the meeting each program holder describes the results of the achievement of the implementation of activities as well as obstacles during program implementation. The evaluation of the results obtained from the implementation of the nutrition improvement program for children under five at the Health Office of Central Buton District consisted of the results of monitoring the status of toddlers at the center, the results of the screening of cases of malnutrition, the results of the provision of additional food for under-nutrition, malnutrition, and stunting in the district. Central Buton, If there are indicators that have not met the target, an evaluation meeting is held at the health department level, to find out the obstacles and how to overcome them so that the target year can be achieved in the next implementation. The health office as the holder of the authority for the nutrition improvement program plays a full role in observing all existing activities in the field by continuing to coordinate with the puskesmas and cadres so that problems that occur can be minimized properly.[22] Evaluation is an orderly and systematic process of comparing the results achieved with the benchmarks set criteria, then drawing conclusions based on suggestions and input from each program implementer, because it will be difficult to know the extent to which the objectives of the planning are achieved if no evaluation is carried out.[23]

Output

Nutrition status monitoring activities are actively carried out by nutrition officers at the health center through the weighing month for under-fives which is carried out twice a year. The state of poor nutrition will reduce the body's resistance which will cause children to get sick easily, resulting in death. Prevention of cases of malnutrition, deficiency, and stunting is carried out by providing additional food (PMT) which is funded through the Central Buton Regency APBD and Southeast Sulawesi Provincial APBD, PMT which is given in the form of milk, MP-ASI biscuits, and milk porridge. Almost all of the cases assisted had gained weight. Prevention of malnutrition under five who require treatment is carried out at the health center, but if there is a congenital disease, the toddler is referred to the Central Buton Hospital.

Nutritional status	2017	2018
Underweight	25.9%	17%
Stunting	48.8%	28%
Wasting	13.3%	9.5%

Based on the results of research and primary data, it is revealed that all toddlers with nutritional problems have received 100% treatment, so there is an increase in the nutritional status of toddlers in 2018, which is marked by a very significant decrease in the problem of nutritional status of children from 2017-1018. In addition, there was an increase in the coverage of under-fives weighing, where the frequency of under-five visits to posyandu in 2017, from 12,060 the number of toddlers, with the coverage of under-five weighed as much as 8,786 (73%) and 3,247 (27.14%) under-fives whose body weight was not monitored, while in 2018 The number of toddlers weighed was 10,870 children with a community participation level of 85.7% of the results obtained, exceeding the national target that has been set, namely 80%, and exceeding the scope of achievement in 2017.[10]

CONCLUSIONS

The nutrition improvement program for children under five in Central Buton District still has several unfulfilled aspects, such as a lack of human resources in quantity and quality, a lack of budget for nutrition improvement programs, which is only around Rp. 10,100,000.00 in 2018, lack of socialization, especially stunting across other sectors, as a result, the problem of stunting in Central Buton Regency is the highest in the Southeast Sulawesi province in 2018, for that the local government needs to make even better efforts.

There is still a need for training for nutrition staff at the Health Office and Nutrition Implementers at health centers, need to increase health funds from the APBD for health development programs, especially community nutrition, need cross-sector and cross-program cooperation in reducing nutrition problems in children under five in Central Buton district, and increasing utilization and socialization of cadres to mothers of children under five so that local communities have higher motivation in efforts to

improve health status both within the family and in the community.

REFERENCES

1. Mitra. Permasalahan Anak Pendek (Stunting) dan Intervensi untuk Mencegah Terjadinya Stunting (Suatu Kajian Kepustakaan). J Kesehat Komunitas. 2015;2(5):254–61.
2. Renyoet BS, Nai HME. Estimasi potensi kerugian ekonomi akibat wasting pada balita di indonesia. J Gizi Indones. 2019;7(2):127.
3. Kementerian Kesehatan Republik Indonesia. Bersama Cegah Stunting. 2018;(Warta Kesmas Tahun 2018).
4. Khoeroh H, Indriyanti D. Evaluasi Penatalaksanaan Gizi Balita Stunting. Unnes J Public Heal. 2017;6(3).
5. Kementerian Kesehatan Republik Indonesia. Gizi, Investasi Masa Depan Bangsa. 2017;(Warta Kesmas Tahun 2017).
6. Zilda O, Sudiarti T. Faktor Risiko Stunting pada Balita (24-59 Bulan) di Sumatera. J Gizi dan Pangan. 2013;8(3):175–80.
7. Badan Penelitian dan Pengembangan Kesehatan. Kementerian Kesehatan. Has Utama Riskesdas 2018. 2018;
8. Gizi Masyarakat D, Jenderal KM D, Kesehatan RI K. Hasil Pemantauan Status Gizi (PSG) Tahun 2017. Buku saku pemantauan Status Gizi. 2017;
9. Rosha CH B, Sari K, Yunita SP I, Amaliah N, Utami N. Peran Intervensi Gizi Spesifik dan Sensitif dalam Perbaikan Masalah Gizi Balita di Kota Bogor. Bul Penelit Kesehat. 2016;44:127–38.
10. Dinas Kesehatan Kabupaten Buton Tengah. Profil Kesehatan Kabupaten Buton Tengah Tahun 2018. 2019;
11. Menteri Kesehatan Republik Indonesia. Peraturan Menteri Kesehatan Republik Indonesia Nomor 23 Tahun 2014. 2014;
12. Peraturan Menteri Kesehatan RI No 26 tahun 2003. Penyelenggaraan Pekerjaan dan Praktik Tenaga Gizi. 2013.
13. Sugiarto A, Retnaningsih C, Indrayati Y. Pendayagunaan Bidan Sebagai Tenaga Kesehatan Di Puskesmas dalam pelaksanaan kebijakan upaya perbaikan gizi pada balita di Kabupaten Buton Tengah Provinsi Sulawesi Tenggara. Soepra J Huk Kesehat. 2018;4(1):1–17.
14. Boli EB, Baliwati YF, Sukandar D. Komitmen Politik dan Peluang Pengembangan Kebijakan Gizi Pemerintah Daerah Provinsi Nusa Tenggara Timur Political Commitment and Opportunity to Advance Nutrition Policy in East Nusa Tenggara Province. J MKMI. 2018;14(4):351–9.
15. Subarsono. Analisis Kebijakan Publik. Pelajar P, editor. Yogyakarta; 2012.
16. Tomi S, Dian H, Soemarno. Evaluasi Program Perbaikan Gizi Masyarakat (Penanggulangan Gangguan Akibat Kekurangan Iodium) di Kota Malang. J Pembang Dan Alam Lestari. 2016;7(1):44–51.
17. Azwar A. Pengantar Administrasi Kesehatan. Bina Rupa Aksara, editor. jakarta; 2012.
18. Sutisna. Manajemen Kesehatan. UNS, editor. surakarta; 2009.
19. Febry F. Jurnal Ilmu Kesehatan Masyarakat. 2014;3(November 2012).
20. Sulaeman S E. Manajemen Kesehatan Teori dan Praktik di Puskesmas Yogyakarta. Yogyakarta: Gadja Mada University Press; 2011.
21. Wijayanti T, Niken P. Studi Kualitatif Proses Asuhan Gizi Terstandar Di Ruang Rawat Inap Rs St. Elisabeth Semarang. J Nutr Coll. 2013;2(1):170–83.
22. Sawitri N. Pelaksanaan Program Perbaikan Gizi Masyarakat Oleh Dinas Kesehatan Kota Pekanbaru. J JOM Fisip. 2018;Vol. 5 No.(1):Hal : 1-15.
23. Sarwono J. Metode Penelitian Kuantitatif dan Kualitatif. In: Graha Ilmu. Yogyakarta; 2006.

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Profile of nutritional status, energy availability, haemoglobin levels and bone density in *santriwati* (Islamic female student) with chronic energy deficiency risk

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ABSTRACT

Background: Santriwati (Islamic female student), women of reproductive age, were susceptible to experienced Chronic Energi Deficiency (CED). CED reflects the low energy availability of someone who can risk reducing bone density. **Objectives**: This study aimed to analyze the differences in body mass index, body fat percentage, hemoglobin levels, energy availability, and bone mineral density of female students who experienced CED risk and not experienced CED risk.
Materials and Methods: The research design was a cross-sectional study, with 101 female students as subjects who were selected by random sampling. The research was conducted from February to March 2019 at the Kyai Galang Sewu Islamic Boarding School, Semarang. CED risk data was taken using the upper arm circumference measurement. Percent body fat and BMI data were taken using BIA. Energy availability data is obtained from the difference between energy intake (energy intake) and energy output (energy expenditure through physical activity) divided by Fat-Free Mass (FFM). Energy intake data was taken using the SQ-FFQ questionnaire, and energy expenditure was calculated using the 24-hour activity record form. Anemia data were collected using strip hemoglobin measurements. Bone density data were taken using the Osteosys Sonost 3000 densitometer. Bivariate analysis used the Independent T-Test.
Results: A total of 57.2% of subjects experienced anemia. Subjects who had underweight nutritional status were 20.8%. Santriwati experienced osteopenia as much as 13.9%. There was no difference in bone density and hemoglobin levels between female students who were at risk of CED and not CED risk ($p > 0.05$), but there were differences in energy availability, body fat percentage, BMI between those at risk of CED and not CED risk ($p < 0.05$)
Conclusion: subjects at risk of CED (Lila <23.5 cm) had lower energy availability, body fat, and BMI than subjects who were not at risk of CED.

Keywords: Anemia; Body fat; BMI; Bone Density; CED risk; Santriwati

BACKGROUND

The number of women of reproductive age (WRA) in Indonesia has the largest number in Southeast Asia with 65 million.[1] Nutritional problem in WRA in Indonesia is still high. One of the nutritional problems that often occur in WRA is Chronic Energy Deficiency (CED). CED is a condition when a person suffers an imbalanced nutrient intake (energy and protein) that lasts for a long time.[2] The prevalence of CED risk on non-pregnant Indonesian women in 2018 based on the Mid-Upper Arm Circumference (MUAC) indicator is 14.5%.[3] The prevalence of CED risk on WRA, 15-19 years old, has increased from 30.9% to 36.3%, and the risk on WRA 20-24 years old also increased from 28.2% to 23.3% in 2007-2018.[3]

One of the WRA groups in Indonesia that tends to have nutritional problems is *santriwati*, female students in Islamic schools. A study conducted at the Al-Hidayah Islamic Boarding School in Grobogan Regency showed that 51.1% of the students were malnourished caused by insufficient nutritional intake.[4] Other research at Slafiyah Kauman Pemalang Islamic Boarding School also showed that 38% of santriwati were malnourished.[5] Body Mass Index (BMI) is used as the indicator to assess the nutritional status, where the body weight (kg) is divided by the square of height (m^2). BMI is

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a valid predictor and is widely used around the world.[6]

Another nutrition issue that always occurs in santriwati is anemia. Studies conducted in Islamic Boarding Schools in Indonesia showed that some Santriwati have low hemoglobin. For example, is in Husada Islamic Boarding School, 83.90% of santriwati experience anemia.[7] In Darul Ulum Peterongan Jombang Islamic Boarding School showed that 57.5% of santriwati had anaemia.[8] The malnutrition and low hemoglobin status of santriwati were usually caused by low food intake both in quantity and quality, for example, is the research that was conducted in Kepung Kediri observed a difference between teenage girls who lived in boarding schools than at home. The study showed that more teenage girls who lived in boarding schools experienced anemia due to the lack of knowledge, parental control and the ability to provide healthy food.[9]

The low energy reserves and nutrients can cause CED in WRA during adolescence may continue to pregnancy and breastfeeding.[10] Energy availability represents the remaining energy in the body or energy reserves for metabolic processes so that it can carry out the physiological function properly.

Energy availability is the difference between energy intake and energy expended during activity (relative to fat-free mass).[11] The energy availability value of 30 kcal/kg FFM /day is the threshold of energy availability required for physiological body functions.[12] Low energy availability usually occurs in athletes. They have a high energy expenditure and are often not compensated with adequate food intake to maintain their body weight.[13]

Aside from the abovementioned problems, a health problem that often arises in women of reproductive age is low bone density. Women were more likely to have a four-time risk of developing osteoporosis than men.[14] Research conducted by Ria Stella on bone density in young adult women showed that nutritional status and body weight had a significant association with bone density. Low bone density was associated with a lower percentage of body fat in subjects, where the body fat mass produces estrogen for the formation of bone mass.[15]

Based on these backgrounds, the researchers were interested in examining the differences between body mass index, percentage of body fat, hemoglobin levels, energy availability, and bone density in female students who are at risk for CED and those who are not at risk for CED.

MATERIALS AND METHODS

This study was conducted at Kyai Galang Sewu Islamic Boarding School, Semarang, in April 2019. This is an observational study with a cross-sectional design. The random sampling technique was used to select a total of 101 participants who fulfill the inclusion criteria. The inclusion criteria include, santriwati who were active at Kyai Galang Sewu Islamic Boarding School, did not suffer from infectious diseases (typhoid, chronic diarrhea, upper respiratory tract infections (URTI), TB, hepatitis, malaria, and dengue fever), did not consume any supplement or drug which may affect body composition, and willing to participate in this study.

For this study, the dependent variables are bone density, hemoglobin level, energy availability (EA), BMI, and body fat percentage. In comparison, the independent variable is chronic energy deficiency (CED) status, which is classified into at risk (CED group) and not at risk (normal group) of CED. The CED status was determined by measuring the participants' mid-upper arm circumference (MUAC) using MUAC tape. The measurement was performed on the left arm. For left-handed women, their right arm was measured. Wearing clothes as thin as possible, and if possible, rolling up the sleeves until the acromion is visible. Mark the acromion and the olecranon process, then mark the mid-point between these marks. Wrap a MUAC tape around the mid-point mark. The participants were categorized as at risk of CED if the mid-upper arm circumference is less than 23.5 cm.[3]

The bone density was measured using a densitometer from Osteosys Sonost 3000 with a quantitative ultrasound method. The bone density was normal if the T-score $-1 \leq SD < 2.5$, the participants were categorized as osteopenia if the T-score is $-2.5 \leq SD < -1$ and osteoporosis if the T-score was < -2.5.[16]

The hemoglobin level was determined by the hemoglobin test strip. The assessment was carried out by drawing blood from the participants to stain the strip before putting the stained strip into a hemoglobin meter from the Hemocue brand. The normal hemoglobin for women is 12 g/dL; anemia is defined by the hemoglobin of less than 12 g/dL.[7]

The BMI is calculated as weight divided by the square of the height in meters. The participants' body height was measured using a microtoise or stature meter to an accuracy of 0.01 cm. the participants stood barefooted and bare of any accessory on their heads.[17] The body weight was measured using a BIA Tanita DC 360 scale. The participants are categorized as underweight if the BMI was <18.5 kg/m^2, normal if the BMI was 18.5-24.9 kg/m^2, and overweight if the BMI was 25-29.9 kg/m^2; and obese if the BMI was ≥30 kg/m^2.

To calculate the body fat percentage, a body composition measurement using BIA Tanita DC 360 was conducted. Previously, the result of the height measurement from the stature meter was put into BIA Tanita DC 360. Twenty-four hours before the data collection, participants were recommended to drink at least eight glasses of water in a day and avoid caffeine and alcohol to achieve good hydration status.[18] The body fat percentage was classified into athletic (10-15%), good (16-19%), normal or acceptable (20-25%), overfat (26-29%), and obese (>30%).[19]

The energy availability was calculated from the difference between energy intake and energy expenditure divided by fat-free mass (FFM). The energy availability status was classified into low EA (<30 kcal/kg FFM/day), good (30-45 kcal/kg FFM/day), and high (>45 kcal/kg FFM/day).[20] The energy intake was obtained by analyzing 24 hour-food recall 6x24 hours using Nutrisurvey 2007 application. During physical activities within 24 hours, the energy expenditure was calculated using the following formula: 3.5 x bodyweight x Basal Metabolic Rate x time.[21] The fat-free mass was obtained from body composition measurement using BIA Tanita DC 360.

The univariate analysis was performed to describe each variable. The bivariate analysis using an independent t-test was performed to analyze the differences in bone density, hemoglobin level, BMI, body fat percentage, and energy availability among santriwati in the CED and normal groups. This study has been approved by the Health Research Ethics Committee (KEPK) Faculty of Medicine, Universitas Diponegoro and Central Public Hospital dr. Kariadi number 162/EC/KEPK/FK-UNDIP/V/2019.

RESULTS

Participants Characteristics

Table 1. Characteristics of participants

Variable	Median	Minimum	Maximum
Age (year)	20	18	24
Weight (kg)	46.7	32.9	67.5
Height (cm)	152.5	139	169.5
Body Mass Index (kg/m^2)	20	14	32
Mid-upper arm circumference (cm)	23.5	19	30.5
Body Fat Percentage (%)	27.3	16	44.9
Haemoglobin (g/dL)	11.6	6.3	15.1
Bone Density (T-Score)	-0.5	-1.6	1.8
Energy intake (kcal)	1310028.5	815 815	1601 0596
Energy availability (kcal/kgFFM/day)	1.9	1	9,6

Table 1 shows the distribution of the participants' characteristics, namely BMI, body fat percentage, mid-upper arm circumference (MUAC), bone density, and hemoglobin level. Some participants recorded a BMI of 14, while others recorded 32 kg/m^2. By the same token, the median hemoglobin level was 11.6 g/dL, but one of the participants recorded only 6.3 g/dL. The bone density among santriwati was relatively normal, although some recorded negative T-score, it was still within the normal range.

Table 2. Characteristics of participants based on nutritional status, anemia status, Chronic Energy Deficiency, and bone density

Category	n	%
Anemia Status		
Anemia	58	57.4
Normal	43	42.6
Chronic Energy Deficiency		
Risk	45	44.6
Normal	56	55.4
Body Fat Percentage		
Normal	68	67.3
Overfat	16	15.8
Obese	17	16.8
Body Mass Index for Age (BMI/U)		
Underweight	21	20.8
Normal	60	59.4
Overweight	11	10.9
Obese	9	8.9
Bone Density		
Normal	87	86.2
Osteopenia	14	13.9

Table 2 further displays the participants' characteristics. Based on hemoglobin assessment, 57.4% of the participants (n=58) were anemic. The nutritional status assessment using MUAC measurement reported 44.6% (n=45) participants were at risk of CED; meanwhile, 68.3% (n=68) had normal body fat percentage. Nutritional status based on BMI showed that 20.8% (n=21) of the participants were underweight. Most of the participants had a normal bone density, but 13.9% (n=14) had osteopenia.

Table 3. Difference of bone density, haemoglobin levels, Chronic Energy availability, body mass index and body fat percentage between CED risk and normal groups

Variable	CED risk (n=45) Mean ±DS	Normal (n=56) Mean ±DS	*P value*
Bone Density (T-Score)	-0.48±0.5	-0.4±0.56	0.458 [a]
Haemoglobin Levels (g/dL)	11.29±1.75	11.48±1.95	0.61 [a]
Energy Availibility (kcal/kgFFM/day)	1.77±0.78	3.98±2.74	<0.001[a*]
Body Mass Index (kg/m^2)	18.68±1.47	22.16±2.83	<0.001[a*]
Body Fat Percentage (%)	24.89±3.76	31.56±5.08	<0.001[a*]

[a]=Independent t-test *significant (p<0.05)

Based on table 3, there was a difference in energy availability between CED risk and normal groups (p<0.001). The mean of energy availability in the CED risk group was lower (1.77 kcal/kgFFM/day ±0.78) than the normal group (3.98 kcal/kgFFM/day ±2.74). Additionally, there was a difference in BMI between the normal and CED groups (p<0.001). The mean BMI in the CED group was lower (18.68 kg/m^2±1.47) than the normal group (22.16 kg/m^2±2.83). There was a difference in body fat percentage between the normal and CED groups (p<0.001). The mean body fat percentage in the CED group was lower (24.89 (%±3.76) than the normal group (31.56 (%±5.08). However, there were no differences in bone density and hemoglobin

levels among the CED and normal groups (p=0.458; p=0.61)

DISCUSSION

The santriwati at Kyai Galang Sewu Islamic Boarding School also receive normal education where most of the santriwati were college students. Based on the analysis, 44.6% of the participants were at risk of CED, determined by the MUAC measurement. MUAC is considered practical to use in the field to assess the risk of CED.[22] The study conducted at AL-Munawwir Islamic Boarding School Yogyakarta reported 26.7% of santriwati were at risk of CED.[23] Furthermore, most of the santriwati were anemic. Anemia among women of reproductive age may increase the risk of maternal death, low birth weight, prone to infection, miscarriage, and increase the risk of premature labor.[24] A report conducted at Salafiyah Syafi'iyah Islamic Boarding School Sukorejo, Situbondo, stated that 79% of the participants were anemic based on their hemoglobin level.[25] Nutritional status measurements based on BMI also indicated that 20.8% of participants in this study were underweight. Another study conducted at Al-Hidayah Islamic Boarding School, Grobogan District, reported that 51.1% of santri had low nutritional status.[4]

The Difference of BMI between Santriwati With and Without Risk of CED

There was a difference in BMI between the normal group and the CED group. It was indicated by the mean BMI in the CED group, which was lower compared to the normal group. Another study by Brito et al. supported this result with their report, which showed a significant association between MUAC and BMI. The lower the MUAC is, the lower BMI would be.[26] Another study in India registered a positive relation between MUAC and BMI among pregnant women.[6] BMI measurement is the most common measurement of nutritional status. Yet, due to the needs of several tools and skills, other means of measurement, such as MUAC, may be used to determine women's nutritional status. This measurement is considered practical and gives an overview of women's nutritional status well.[6] More articles showed an association between MUAC and BMI, which also explains the difference in BMI between santriwati with and without CED risk. The BMI of the CED group was lower than the normal group.

The Difference of Body Fat Percentage between Santriwati With and Without Risk of CED

Based on the analysis, there was a difference in body fat percentage between the CED and normal groups. This finding is supported by another study in India that reported a significant association between BMI and body fat percentage. The lower the BMI score is, the lower the body fat percentage would be.[27] BMI also gives an overview of CED in women.[28] It may explain the difference in the mean body fat percentage between the CED and normal groups. The body fat percentage of the CED group was lower than the normal group.

The Difference of Energy Availability between Santriwati With and Without Risk of CED

Based on the analysis, there was a difference in energy availability between the CED and normal groups. The mean score of energy availability in the CED group was lower compared to the normal group. The energy status assessment, such as energy availability measurement, is an accurate and objective measurement that can also reflect CED's compensation compared to using only nutritional status assessment.[29] The energy availability refers to the amount of energy leftover and available for body function after the energy expended for physical activities is subtracted from the energy consumed.[20] Low energy availability among santriwati was mostly caused by low energy consumption. Low energy reserves and nutrients may increase the risk of CED.[11] Loucks stated that low energy availability might cause loss of muscle mass or fat mass in the body. If it continues for a prolonged time, then physiological function may be compromised.[30] CED's risk increases if the depletion of muscle mass develops continually without adequate energy intake.[31]

The Difference of Haemoglobin Level between Santriwati With and Without Risk of CED

Based on the statistical analysis, there was no significant difference in CED and normal groups' hemoglobin levels. It contrasts with Pramodya et al. at Kediri District's findings, which reported a significant difference in hemoglobin levels between students with and without CED.[32] However, a report

by Dea Intartanti showed an insignificant association between nutritional status and anemia among female adolescents.[33] This may be since macronutrients (carbohydrate, protein, fat) intake is not the only affecting factor hemoglobin level. Micronutrient intake also affects the hemoglobin factor, it was reported that the micronutrient intake is relatively good in the said study.[33] Energy intake deficiency may cause anemia as the protein breakdown is no longer aimed at erythropoiesis (red blood cell production) but instead to produces energy or glucose.[32] Another study by Wiraprasidi et al. at Lolak Public Health Centre also reported similar findings; there was an insignificant association between hemoglobin level and MUAC.[34]

The Difference of Bone Density between Santriwati With and Without Risk of CED

A significant difference in bone density between CED and normal groups was not found based on the analysis. This finding supports a previous study by Ana Yuliah Rahmawati, which stated no significant association between BMI and bone density among participants.[35] Likewise, Shera Mutiara reported no significant association between nutritional status and bone density among female adolescents.[19] Many factors influence bone density; one of them being protein intake. Adequate protein intake is necessary to maintain estrogen production, which regulates bone synthesis.[15] However, the protein intake in both the CED and normal groups was relatively similar; this may justify bone density similarity in both groups. Another influencing factor is body fat percentage. A significant association between bone density and body fat percentage was reported in a previous study conducted in Bandung. The higher the body fat percentage, the lower risk of osteoporosis is. However, maintaining a body fat percentage in the normal range is recommended to prevent degenerative diseases.[36]

CONCLUSIONS

Based on the hemoglobin level, 57.4% of santriwati had anemia. Nutritional status assessment based on MUAC showed that 44.6% of participants were at risk of CED. On the other hand, 67.3% of the participants had a normal body fat percentage. Santriwati with osteopenia accounted for 13.9% of the total participants. There were significant differences in energy availability, BMI, body fat percentage between CED and normal groups. The mean scores in the CED group were lower than the normal group. Yet, there were no significant differences in bone density and hemoglobin levels between CED and normal groups.

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REFERENCES

1. Mujiati I, Budijanto D, Khairani. Situasi Keluarga Berencana di Indonesia. Indonesia: Kementerian Kesehatan Republik Indonesia; 2013.
2. Muliawati S. Faktor Penyebab Ibu Hamil Kurang Energi Kronis di Puskesmas Sambi Kecamatan Sambi Kabupaten Boyolali tahun 2012. 1. 2013;3(3):40–62.
3. Kementerian Kesehatan RI. Riset Kesehatan Dasar 2018 (RISKESDAS 2018). Indonesia: Kementerian Kesehatan Republik Indonesia; 2018.
4. Peni Catur Inayati. Hubungan antara Status Gizi dan menstruasi dengan Kejadian Anemia pada Santri Putri Pondok Pesantren Al-Hidayah Kecamatan Karangrayung Kabupaten Grobogan tahun 2009 [Internet] [other]. Universitas Negeri Semarang; 2009 [cited 2020 Sep 21]. Available from: https://lib.unnes.ac.id/1384/
5. Hidayah N, Rahfiludin MZ, Aruben R. Hubungan Status Gizi, Asupan Zat Gizi, dan Aktivitas Fisik dengan Siklus Menstruasi Remaja Putri Pondok Pesantren Salafiyah Kauman Kabupaten Pemalang Tahun 2016. Jurnal Kesehatan Masyarakat (e-Journal). 2016 Nov 2;4(4):537–444.
6. Kumar P, Sinha R, Patil N, Kumar V. Relationship between mid-upper arm circumference and BMI for identifying maternal wasting and severe wasting: a cross-sectional assessment. Public Health Nutr. 2019;22(14):2548–52.
7. Priyanto LD. The Relationship of Age, Educational Background, and Physical Activity

on Female Students with Anemia. JBE. 2018 Aug 30;6(2):139–46.
8. Sya`Bani IRN, Sumarmi S. Hubungan Status Gizi dengan Kejadian Anemia pada Santriwati di Pondok Pesantren Darul Ulum Peterongan Jombang. Jurnal Keperawatan Muhammadiyah. 2017 Mar 28;1(2):7–15.
9. Purwandari ES. Comparison Anemia In Adolescent Girls Who Stay At Pondok Pesantren And At Home In Madrasah Aliyah Pondok Pesantren Darussalam Kepung Kediri. J I K E B. 2018 Sep 25;4(2):114–9.
10. Ariyani DE, Achadi EL, Irawati A. Validity Mid-Upper Arm Circumference to Detect Chronic Energy Malnutrition Risk of Indonesia Women. Kesmas: National Public Health Journal. 2012 Sep 1;7(2):83.
11. Ong JL, Brownlee IA. Energy Expenditure, Availability, and Dietary Intake Assessment in Competitive Female Dragon Boat Athletes. Sports (Basel). 2017 Jun 21;5(2):45.
12. Papageorgiou M, Dolan E, Elliott-Sale KJ, Sale C. Reduced energy availability: implications for bone health in physically active populations. Eur J Nutr. 2018;57(3):847–59.
13. Sygo J, Coates AM, Sesbreno E, Mountjoy ML, Burr JF. Prevalence of Indicators of Low Energy Availability in Elite Female Sprinters. Int J Sport Nutr Exerc Metab. 2018 Sep 1;28(5):490–6.
14. Noprisanti N, Masrul M, Defrin D. Hubungan Asupan Protein, Kalsium, Phosfor, Dan Magnesium Dengan Kepadatan Tulang Pada Remaja Putri di SMP Negeri 5 Padang. Jurnal Kesehatan Andalas. 2018 Oct 17;7:29–36.
15. Septriani RS, Fitranti DY. Hubungan Asupan Protein dan Kafein dengan Kepadatan Tulang pada Wanita Dewasa Muda. J NutriColl. 2013 Oct 3;2(4):483–90.
16. Schtscherbyna A, Ribeiro BG, Farias MLF de. Bone Health, Bone Mineral Density, and Sports Performance. In: Nutrition and Enhanced Sports Performances. San Diego: Elsevier; 2013. p. 75–80.
17. Zhang Y, Wang Z, Chu Z, Zhao J. Profiles of body mass index and the nutritional status among children and adolescents categorized by waist-to-height ratio cut-offs. International Journal of Cardiology. 2016 Nov;223:529–33.
18. Lee RD, Nierman DC. Anthropometry. In: Nutritional Assessment. 3rd ed. New York: McGraw-Hill Education; 2003. p. 184–203.
19. Rahmani SM, Probosari E. Hubungan Indikator Obesitas dengan Kepadatan Tulang pada Wanita Dewasa Awal. Journal of Nutrition College. 2016;5(4):444–51.
20. Márquez S, Molinero O. Energy availability, menstrual dysfunction and bone health in sports; an overview of the female athlete triad. Nutr Hosp. 2013 Aug;28(4):1010–7.
21. Setyarsih L, Ardiaria M, Fitranti DY. Hubungan Densitas Energi dan Asupan Cairan dengan Berat Jenis Urin pada Remaja. Journal of Nutrition College. 2017;6(4):326–32.
22. Sandjaja. Risiko Kurang Energi Kronis (KEK) pada Ibu Hamil di Indonesia. Gizindo. 2014 Sep 1;32(2):128–38.
23. Alifah HN, Anita DC. Hubungan Status Gizi dengan Kadar Hemoglobin pada Santriwati di Pondok Pesantren Al-Munawwir Krapyak Bantul Yogyakarta [Internet] [s1_sarjana]. [Yogyakarta]: Universitas 'Aisyiyah Yogyakarta; 2017 [cited 2020 Sep 21]. Available from: http://lib.unisayogya.ac.id/
24. Sudikno S, Sandjaja S. Prevalensi Dan Faktor Risiko Anemia Pada Wanita Usia Subur Di Rumah Tangga Miskin Di Kabupaten Tasikmalaya Dan Ciamis, Provinsi Jawa Barat. Kespro. 2016;7(2):71–82.
25. Hamidiyah A, Rohmani L, Zahro NA. Faktor Determinan Anemia Santri Putri. Oksitosin : Jurnal Ilmiah Kebidanan. 2019 Feb 1;6(1):64–72.
26. Benítez Brito N, Suárez Llanos JP, Fuentes Ferrer M, Oliva García JG, Delgado Brito I, Pereyra-García Castro F, et al. Relationship between Mid-Upper Arm Circumference and Body Mass Index in Inpatients. PLoS ONE. 2016;11(8):e0160480.
27. Misra P, Singh AK, Archana S, Lohiya A, Kant S. Relationship between body mass index and percentage of body fat, estimated by bio-electrical impedance among adult females in a rural community of North India: A cross-sectional study. J Postgrad Med. 2019 Sep;65(3):134–40.
28. Shetty PS, James WP. Body mass index. A measure of chronic energy deficiency in adults. FAO Food Nutr Pap. 1994;56:1–57.

29. Williams NI, Statuta SM, Austin A. Female Athlete Triad: Future Directions for Energy Availability and Eating Disorder Research and Practice. Clin Sports Med. 2017 Oct;36(4):671–86.
30. Holtzman B, Ackerman KE. Measurement, Determinants, and Implications of Energy Intake in Athletes. Nutrients. 2019 Mar;11(3):665.
31. Petrika Y, Hadi H, Nurdiati DS. Tingkat asupan energi dan ketersediaan pangan berhubungan dengan risiko kekurangan energi kronik (KEK) pada ibu hamil. Jurnal Gizi dan Dietetik Indonesia (Indonesian Journal of Nutrition and Dietetics). 2016 Aug 30;2(3):140–9.
32. Wardhani JP, Rahfiludin MZ, Pradigdo SF. Perbedaan Aktivitas Fisik, Kadar Hb, dan Kesegaran Jasmani (Studi pada Siswi KEK dan Tidak KEK di SMAN 1 Grogol Kabupaten Kediri). Jurnal Kesehatan Masyarakat (e-Journal). 2017 Dec 13;3(3):205–12.
33. Indartanti D, Kartini A. Hubungan Status Gizi dengan Kejadian Anemia pada Remaja Putri. Journal of Nutrition College. 2014;3(2):310–6.
34. Wiraprasidi IPA, Kawengian SE, Mayulu N. Faktor-Faktor yang Berhubungan dengan Kadar Hemoglobin pada Ibu Hamil di Puskesmas Lolak. eBiomedik [Internet]. 2017 [cited 2020 Sep 21];5(2). Available from: https://ejournal.unsrat.ac.id/index.php/ebiomedik/article/view/18602
35. Rahmawati AY. Hubungan antara Indeks Massa Tubuh (IMT), Asupan Zat Gizi dan Riwayat Reproduksi dengan Kepadatan Mineral Tulang pada Wanita Pre-Menopause. Jurnal Riset Kesehatan. 2016;5(2):83–91.
36. Widyanti LRE, Kusumastuty I, Arfiani EP. Hubungan Komposisi Tubuh dengan Kepadatan Tulang Wanita Usia Subur di Kota Bandung. Indonesian Journal of Human Nutrition. 2017 Jun 25;4(1):23–33.

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Ganyong-kelor snack bar's glycemic index as a diet for diabetics

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ABSTRACT

Background: Lack of insulin or the inability of cells to respond to insulin causes high blood glucose levels or hyperglycemia, a hallmark of diabetes. Consumption of foods with a low glycemic index and high fiber has been shown to provide the same benefits as pharmacological therapy in the control of postprandial hyperglycemia and can prevent the incidence of hypoglycemia in people with diabetes. Ganyong (Canna edulis) is a food source of carbohydrates and fiber. Kelor (Moringa oliefera) contains protein and some phytochemical compounds which have a hypoglycemic effect.
Objectives: The objective of the study was to analyze the glycemic index of ganyong-kelor snack bars as a diet for diabetics.
Materials and Methods: Ten respondents fasted for 10 hours and checked their fasting blood glucose levels, then consumed 105 grams of bread as the reference food. Every 30 minutes after eating, the blood glucose levels were checked. In the following week, after fasted, all respondents consumed 157 grams of a ganyong-kelor snack bar and checked their blood glucose levels every 30 minutes.
Results: Every 100 grams of ganyong-kelor snack bar contains 230.13 kcal, 31.97 grams of carbohydrates, 9.25 grams of fat, and 4.75 grams of protein. In this study, bread was used as a reference food. If bread was corrected with glucose as a reference food, the glycemic index of the ganyong-kelor snack bar was 38.08. The calculation of the glycemic load used the converted-glycemic index and the total carbohydrates contained in 100 grams of the food. Ganyong-kelor snack bar had a glycemic load value of 12.10.
Conclusions: Ganyong-kelor snack bar had good nutritional content and was categorized as food with a low glycemic index. The hypoglycemic effect of the ganyong-kelor snack bar came from its high fiber content. Ganyong-kelor snack bar can be consumed as a healthy snack for diabetic people.

Keywords: Ganyong; Canna; Kelor; Moringa; Diabetes

BACKGROUND

Diabetes mellitus is a chronic condition that occurs as a result of increased levels of glucose in the blood caused by the body's unable to produce sufficient amounts of insulin or the body is unable to use the insulin produced effectively.[1] Lack of insulin or the inability of cells to respond to insulin causes high blood glucose levels, or hyperglycemia, which is a hallmark of diabetes.[2] In 2017 the prevalence of diabetes mellitus globally is 8.8% or around 425 million adults with 85-95% of these cases are type 2 diabetes mellitus and is expected to increase to 9.9% in 2045.[2-4] The prevalence of diabetes mellitus in Indonesia increases rapidly from 5.7% in 2007 to 6.9 % in 2013 to 8.5% in 2018.[5] Diabetes mellitus has a significant economic impact on countries, health systems, and diabetics or families through direct medical costs and job losses and reduced wages.[5]

Modifiable risk factors for type 2 diabetes mellitus include poor diet and nutrition, excess adiposity, low physical activity, prediabetes or impaired glucose tolerance (IGT), smoking habits, and a history of fetal exposure to high blood glucose during pregnancy.[4] Recent evidence suggests an association between regular exposure to a high glycemic load diet and causing postprandial hyperglycemia.[6-11] Consumption of foods with low glycemic index and high fiber has been shown to provide the same benefits as pharmacological therapy in the control of postprandial hyperglycemia in the medium term and can prevent the incidence of hypoglycemia in people with diabetes.[11-13] This type of diet can improve insulin sensitivity and fat metabolism.[14] This can provide a good choice for diabetics and can reduce health care costs.

Ganyong (*Canna edulis*) is a food source of carbohydrates with a low glycemic index (20.8) and high fiber (8.59%). The starch content of ganyong consists of 25% amylose.[15.16] The protein content of kelor (*Moringa oliefera*) leaves is 35% of the dry weight. Kelor leaves contain some phytochemical compounds such as alkaloids, flavonoids, glycosides, tannins, and steroids, which have a hypoglycemic effect.[17,18] Diabetics must maintain their diet, type, and calorie content in food, especially in those who use insulin secretion-enhancing drug therapy or

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insulin injection therapy.[19] G*anyong-kelor* snack bar is expected to have low index glycemic and can be consumed as a snack for diabetics without causing hyperglycemia.

MATERIALS AND METHODS

This research was an experimental study. *Ganyong-kelor* snack bar is made using ingredients such as ganyong/ canna obtained from Giwangan Market, canna flour obtained from the Center for Food and Nutrition Studies Universitas Gadjah Mada, kelor/ moringa leaves obtained from Purworejo, non-calorie sweetener, skim milk, margarine, eggs, and essence chocolate. This research has been approved and received ethical permission from *Komite Etik Penelitian Kesehatan Politeknik Kesehatan* (KEPK) Yogyakarta (LB.01.01/KE-01/XXXIV/731/2018). Testing of nutritional content includes analysis of protein (the Kjeldahl method), fat (the Soxhlet method), carbohydrates (the difference method), crude fiber (the gravimetric method), and amylose content (the IRRI method) carried out at the Food Technology Laboratory, Universitas Gadjah Mada.

The glycemic index test was carried out on 10 subjects who had fasted for 10 hours then ate the reference food in the form of white bread with 50 grams of carbohydrates. Every 30 minutes (for 2 hours) a 20 μL blood sample was taken to measure the postprandial blood glucose level. One week later, the same procedure is carried out by providing a snack bar containing 50 grams of carbohydrates. The glycemic index was obtained by the incremental area under the blood glucose response curve (IAUC) method by comparing the area under the blood glucose response curve to a snack bar with a reference food.

RESULTS

Ten selected respondents who had signed an informed consent have normal nutritional status measured by body mass index (BMI) and fasting blood sugar 70-100 mg/dL. Respondents fasted for 10 hours and checked their blood sugar twice with an interval of 7 days. The average result of the first measurement of fasting blood sugar for all respondents was 84.90 mg/dL while the second measurement was 84.30 mg/dL. The mean value of the two measurements is within the normal fasting blood glucose range (70-100 mg/dL).

After the respondents fasted and checked their fasting blood glucose levels, all respondents consumed 105 grams (50 gr carbohydrates) of bread as the reference food. Every 30 minutes after eating, the blood glucose levels were checked for each respondent. In the following week, after the respondents fasted, all respondents consumed 157 grams (50 gr carbohydrates) of the *ganyong-kelor* snack bar and checked their blood glucose levels every 30 minutes.

Table 1. Nutrient Content per 100g G*anyong-Kelor* Snackbar

Nutrient	100 g G*anyong-Kelor* Snackbar
Energy	230.13 kkal
Water	49.79 %
Carbohydrate	31.97 %
Amilosa	10.04 %
Fat	9.25 %
Protein	4.75 %
Ash	4.23 %
Crude fiber	2.64 %

The blood glucose response of each respondent after consuming the reference food and *ganyong-kelor* snack bar is seen in Figure 1. There was an increase in blood glucose levels (51.11%) from 84.9 mg/dL to 128.3 mg/dL within 30 minutes after consuming bread. This blood glucose level becomes the peak of the increase in blood glucose levels. In the next 30 minutes, there was a decrease in blood glucose levels to 121.7 mg/dL or 5.14% of the peak value and continued to decrease to 24.70% at 2 hours after consuming bread.

The peak increase in blood glucose levels after snack bar consumption reached 114.7 mg/dL or 36.06% from fasting blood glucose (84.3 mg/dL). Measurement of blood glucose levels in the next 30 minutes to 2 hours showed a decrease in blood glucose levels of 14.55%, 16.73%, and 20.48% respectively from the highest levels. Compared to bread, consumption of the *ganyong-kelor* snack bar caused a slower increase in blood glucose levels and closer to normal blood glucose levels after 2 hours of snack bar consumption. The glycemic index is calculated from the increase in blood glucose with fasting glucose levels as the initial value. The glycemic index is obtained by calculating the broad curve of the increase in blood glucose levels after

consuming bread (A) and consuming ganyong-kelor snack bar (B) in Figure 2. The B/A ratio multiplied by 100 will reveal the glycemic index of the ganyong-kelor snack bar.The ganyong-kelor snack bar has a glycemic index value of 55.19. In this study, bread was used as a reference food. If this study uses glucose as a reference (glycemic index 100 and glycemic index of bread 69) and corrected bread as a reference food, it will make the glycemic index value of the ganyong-kelor snack bar to 38.08. The calculation of the glycemic load uses the converted-glycemic index value and the total carbohydrates contained in 100 grams of the food. Ganyong-kelor snack bar has a glycemic load value of 12.10.

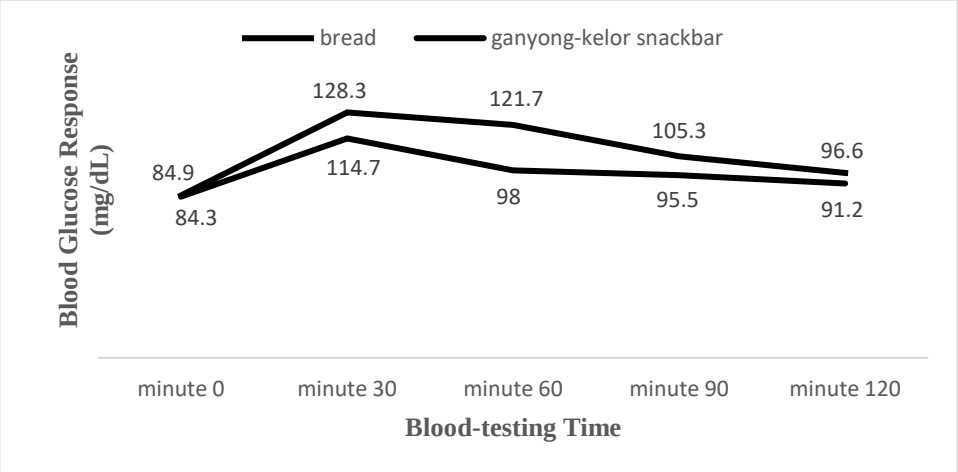

Figure 1. Blood Glucose Response After Consuming Bread and *Ganyong-Kelor* Snackbar

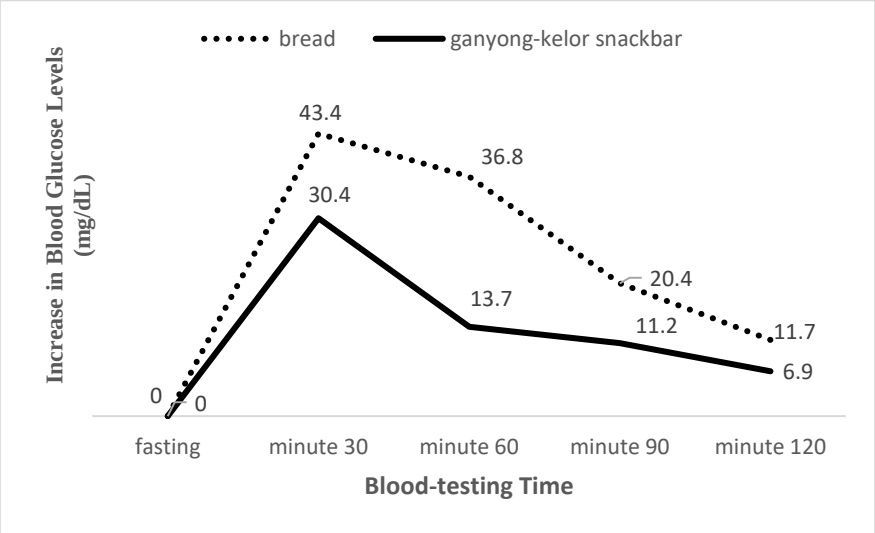

Figure 2. Increase in Blood Glucose Levels After Consuming Bread and G*anyong-Kelor* Snackbar

Table 2. Glycemic Index of G*anyong-Kelor Snackbar* and Bread

Food	Glycemic Index (with bread as reference)	Glycemic Index (with glucose as reference)	Glycemic Load
Bread	100	69	32.85
ganyong-kelor snackbar	55.19	38.08	12.10

DISCUSSION

Diabetics must maintain their diet. type, and calorie content in food, especially in those who use insulin secretion-enhancing drug therapy or insulin

injection therapy.[19] A snack is needed to accompany the main meal in meeting nutritional needs and to help control diabetics' blood glucose levels. Snack bars are a food product that has begun to be developed as a snack for diabetics that can help prevent hyperglycemia by using foods high in fiber and low on the glycemic index. It is recommended that diabetics have a snack that contains 10-15% of their daily energy needs and can be consumed 2-3 times a day.[19]

Adequate energy to do daily activities and maintain ideal body weight is met by consuming a *ganyong-kelor* snack bar with the appropriate portion and calorie content. Consumption of foods with excess calories can lead to obesity which is a risk factor for type 2 diabetes.[21] In every 100 grams of *ganyong-kelor* snack bar contains 230.13 kcal. Diabetics can consume 43.45 grams of the *ganyong-kelor* snack bar to get 100 kcal of calorie intake. This portion contains 13.89 grams of carbohydrates, 4.01 grams of fat, and 2.06 grams of protein. *Ganyong-kelor* snack bar has a glycemic index of 55.19. This study using bread as a reference food. If using glucose as a reference, makes the glycemic index of the g*anyong-kelor* snack bar to 38.08. Based on the classification of the glycemic index, after correcting the *ganyong-kelor snack bar* is included in a low glycemic index food (> 55).

The ingredients of the *ganyong-kelor snack bar* that are thought to have an impact on the low glycemic index are ganyong/ canna and kelor/ moringa leaves. In these two food ingredients, fiber content is the part that greatly affects the glycemic index. The total dietary fiber in 100 grams of canna starch ranges from 81.9 to 85.9% dry weight. The high amylose and fiber content in canna affects the low glycemic index of the *ganyong-kelor snack bar*. As a high-carbohydrate food, ganyong has a lower glycemic index (20.8) when compared to rice (72.8).[15] Consumption of foods with a low glycemic index can improve insulin sensitivity and decrease the rate of glucose absorption which improves glycemic control in diabetics.[11-13, 22]

Food with a low glycemic index will be digested and converted into glucose gradually and slowly resulting in a slower release of glucose into the bloodstream so that the increase in blood glucose levels will be lower and the fluctuation of the increase in blood glucose is relatively shorter. This will affect increasing insulin secretion and glucose consumption by liver cells which will result in reduced blood glucose levels. Consumption of foods with a low glycemic index can improve insulin sensitivity and reduce the rate of glucose absorption. This can improve glycemic control in people with diabetes mellitus.[11-13,22] Some of the factors that influence the glycemic index are carbohydrate composition, tertiary structure, and enzymatic digestion.[22] Clinical studies show that consumption of foods high in water-soluble fiber such as guar gum, pectin, β-glucans found in nuts, vegetables, fruit, and oat cereals can control insulin levels and lower postprandial blood glucose levels.[22] The hypoglycemic effect of a diet high in soluble fiber can be achieved through slowing gastric emptying and enzymatic absorption, shorter intestinal transit times, and the formation of a physical barrier on carbohydrates that slows down glucose uptake.[22,23]

The glycemic load is defined as the glycemic index of a food multiplied by the carbohydrate content in the product. The calculation of the glycemic load is intended to provide information about the effect of food consumption on increasing blood glucose levels associated with the glycemic index. The lower the carbohydrate content, the lower the glycemic load. Then the food consumed will trigger a lower increase in blood glucose levels compared to the reference food. *Ganyong-kelor snack bar* has a glycemic load value of 12.10. This food is included in the category of moderate glycemic load foods (range of 11 to 19).

This study has many limitations. The material used as the standard for measuring the glycemic index is bread. The author suggests further research using glucose as a reference. However, the analysis of the results of this study can provide important evidence regarding the use of canna and moringa leaves as materials for snack bars, introducing *ganyong-kelor snack bar* as an alternative snack for diabetics, and can be the starting point for further research.

CONCLUSIONS

Ganyong-kelor snack bar has good nutritional content and is categorized as food with a low glycemic index. Ganyong-kelor snack bar can be consumed as a healthy snack for diabetes without causing hyperglycemia. Every 100 grams of *ganyong-kelor snack bar* contains calories of 230.13 kcal, 31.97 grams of carbohydrates, 9.25 grams of fat, and 4.75 grams of protein. The glycemic index of the *ganyong-kelor snack bar was* 38.08 (low category) with a glycemic load of 12.10 (moderate category). The hypoglycemic effect of snack bars comes from their high fiber content. The hypoglycemic effect of a diet high in fiber can be achieved through slowing gastric emptying and enzymatic absorption, shorter intestinal transit times, and the formation of a physical barrier on carbohydrates that slows down glucose uptake. Knowing the glycemic load on food will help us to choose food and determine food

portions wisely so that it will limit carbohydrate intake too high which can cause an increase in blood glucose levels. This is especially important for diabetics or people who want to limit their calorie intake to control their weight.

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REFERENCES

1. World Health Organization. Definition, Diagnosis, and Classification of Diabetes Mellitus and its Complications. Part 1: Diagnosis and Classification of Diabetes Mellitus (WHO/NCD/NCS/99.2). Geneva: World Health Organization; 1999. *http://apps.who.int/iris/handle/10665/66040*

2. The International Diabetes Federation. Diabetes Atlas 8th Edition. Brussels: The International Diabetes Federation; 2017. *https://www.idf.org/e-library/epidemiology-research/diabetes-atlas.html*

3. World Health Organization. Global report on Diabetes. Geneva: World Health Organization; 2016. *http://apps.who.int/iris/bitstream/10665/204871/1/9789241565257_eng.pdf*

4. NCD Risk Factor Collaboration. Worldwide trends in diabetes since 1980: a pooled analysis of 751 population-based studies with 4.4 million participants. Lancet. April 2016; 387: 1513-1530. DOI: *http://dx.doi.org/10.1016/S0140-6736(16)00618-8*

5. Badan Penelitian dan Pengembangan Kesehatan. Laporan Nasional Riskesdas 2018. Jakarta: Lembaga Penerbit Badan Penelitian dan Pengembangan Kesehatan; 2019.

6. Castro-Acosta ML, Stone SG, Mok JE, Mhajan RK, Fu CI, Lenihan-Geels GN, et al. Apple and blackcurrant polyphenol-rich drinks decrease postprandial glucose, insulin, and incretin response to a high-carbohydrate meal in healthy men and women. Journal of Nutritional Biochemistry. 2017; 49:53–62. DOI: *http://dx.doi.org/10.1016/j.jnutbio.2017.07.013*

7. Schulze MB, Manson JE, Ludwig DS, Colditz GA, Stampfer MJ, Willett WC, et al. Sugar-sweetened beverages, weight gain, and incidence of type 2 diabetes in young and middle-aged women. JAMA. 2004; 292(8): 927–34. DOI: *http://dx.doi.org/10.1001/jama.292.8.927*

8. Sluijs I, van der Schouw YT, van der AD, Spijkerman AM, Hu FB, Grobbee DE, et al. Carbohydrate quantity and quality and risk of type 2 diabetes in the European prospective investigation into cancer and nutrition-Netherlands (EPIC-NL) study. Am J Clin Nutr. 2010; 92: 905–11. DOI: *https://doi.org/10.3945/ajcn.2010.29620*

9. de Koning L, Malik VS, Rimm EB, Willett WC, Hu FB. Sugar-sweetened and artificially sweetened beverage consumption and risk of type 2 diabetes in men. Am J Clin Nutr. 2011; 93(6): 1321–27. DOI: *https://doi.org/10.3945/ajcn.110.007922*

10. Blaak EE, Antoine JM, Benton D, Bjorck I, Bozzetto L, Brouns F, et al. Impact of postprandial glycemia on health and prevention of disease. Obes Rev. 2012; 13(10): 923–84. DOI: *https://doi.org/10.1111/j.1467-789X.2012.01011.x*

11. Riccardi G, Rivellese AA, Giacco R. Role of glycemic index and glycemic load in the healthy state, in prediabetes, and diabetes. Am J Clin Nutr. 2008; 87(1): 269S–74S. DOI: https://doi.org/10.1093/ajcn/87.1.269S

12. Brand-Miller J, Hayne S, Petocz P, Colagiuri S. Low glycemic index diets in the management of diabetes: a meta-analysis of randomized controlled trials. Diabetes Care. 2003; 26(8): 2261–67. DOI: https://doi.org/10.2337/diacare.26.8.2261

13. American Dietetic Assosiation. Nutrition recommendation and interventions for diabetes (position statement). Diabetes Care. January 2008; 31(1): S61-S78. DOI: *https://doi.org/10.2337/dc08-S061*

14. Rizkalla SW, Taghrid L, Laromiguiere M, Huet D, Boillot J, Rigoir A, et al. Improved plasma glucose control, whole-body glucose utilization, and lipid profile on a low glycemic index diet in type 2 diabetic men. Diabetes Care. 2004; 27(8): 1866–72. DOI: https://doi.org/10.2337/diacare.27.8.1866

15. Lukitaningsih E, Rumiyati, Ika Puspitasari. Kajian glisemik indeks dan makronutrien dari umbi-umbian dalam upaya pencarian sumber pangan fungsional. Pharmacon. 2012; 13(1): 18-

23. https://publikasiilmiah.ums.ac.id/bitstream/handle/11617/3377/2012-13-1-18.pdf?sequence=1&isAllowed=y
16. Carolina A, Ilmi FN. Production of Indonesian *canna edulis* type iv resistant starch through acetylation modification. International Food Research Journal. 2016; 23(2): 491-97. http://www.ifrj.upm.edu.my/23%20(02)%202016/(6).pdf
17. Tshingani K, Donnen P, Mukumbi H, Duez P, Dramaix-Wilmet M. Impact of *Moringa oleifera lam.* leaf powder supplementation versus nutritional counseling on the body mass index and immune response of HIV patients on antiretroviral therapy: a single-blind randomized control trial. BMC Complementary and Alternative Medicine. 2017; 17:420-33. DOI: *https://doi.org/10.1186/s12906-017-1920-z*
18. Jaiswal D, Rai PK, Mehta S, Chatterji S, Shukla S, Rai DK, et al. Role of *Moringa oleifera* in regulation of diabetes-induced oxidative stress. Asian Pacific Journal of Tropical Medicine. 2013; 6(6): 426-32. DOI: *https://doi.org/10.1016/S1995-7645(13)60068-1*
19. Perkumpulan Endokrinologi Indonesia. Konsensus pengelolaan dan pencegahan diabetes melitus tipe 2 di Indonesia 2015. Jakarta: PB. Perkeni; 2015. https://pbperkeni.or.id/wp-content/uploads/2019/01/4.-Konsensus-Pengelolaan-dan-Pencegahan-Diabetes-melitus-tipe-2-di-Indonesia-PERKENI-2015.pdf
20. Mortensen A. Sweeteners permitted in the European Union: safety aspects. Scandinavian Journal of Food and Nutrition. 2006; 50(3): 104-16. DOI: https://doi.org/10.1080/17482970600982719
21. Franz MJ. Medical nutrition therapy for diabetes mellitus and hypoglycemia of nondiabetic origin. In: Mahan LK, Escottt-Stump S. Krause's food, and the nutrition care process. 13th edition. Philadelphia: Elsevier; 2012. 675-710.
22. Frost G. Carbohydrate: Glucose tolerance and the glycemic (glycaemic) index. In: Caballero B. Encyclopedia of food sciences and nutrition. New York: Academic Press; 2003. 2917-22.
23. Gallagher ML. Intake: The nutrient and their metabolism. In: Mahan LK, Escottt-Stump S. Krause's food, and the nutrition care process. 13th edition. Philadelphia: Elsevier; 2012. 32-41.

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Household food security and diet quality with chronic energy deficiency among preconception women

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ABSTRACT

Background: Chronic energy deficiency (CED) that occurs at risk preconception women during pregnancy increased low birth weight (LBW) in infants. Household food security and diet quality are factors that cause CED. Therefore, this study aims to determine the relationship between household food security and diet quality with CED preconception women.
Materials and Methods: We used a cross-sectional study design. The subject of 70 preconception women aged 16-35 years registered in the religious affairs office in Sumowono and Pringapus Subdistrict were selected by consecutive sampling method. Weight and height were measured to assess body mass index to determine CED. Household food security was measured using the Household Food Security Scale Module (HFSSM). Food intake data were obtained using the Semi-Quantitative Food Frequency Questionnaire (SQ-FFQ) and DQI-I (Diet Quality Index-International) to measure diet quality. Bivariate analyses were tested using Rank Spearman and Pearson Product Moment.
Results: The prevalence of subjects with CED risk was 48.6% and subjects with CED was 15.7%. 38.6% of subjects married at the age of 16-20 years, 75.1% of subjects had low household food security and 80% of subjects had low diet quality. There was no significant relationship between household food security and diet quality with CED, namely (p = 0.537) and (p = 0.711). The components of diet quality, namely variation, adequacy, moderation and balance also did not show a significant relationship with CED, respectively with p-value (p = 0.711), (p = 0.523), (p = 0.412), (p = 0.604)
Conclusions: There was no correlation between household food security and CED, also no correlation between diet quality and CED.

Key Words: CED; Diet quality; food security; Preconception woman

BACKGROUND

The preconception period is the period before pregnancy. Preconception women are women of reproductive age, who will have to be ready to become mothers. The preconception period needs different nutritional support compared to the previous period. The nutritional status of pregnant women is determined by the period before pregnancy, thus the poor nutritional status of preconception women will have an impact on the pregnancy. Preconception women are also defined as women of reproductive age who are also prone to CED.[1]

Chronic energy deficiency (CED) in women is defined as a condition when a person experiences a prolonged or chronic lack of calories and protein. CED is characterized by a mid-upper arm circumference (MUAC) of <23.5 cm.[2] Another indicator in defining CED is body mass index (BMI) of <18.5 kg/m^2.[3] Based on the results of Basic Health Research (Riskesdas) 2013, the prevalence of pregnant women aged 15-49 years who suffer from CED was 24.3%, while the prevalence of women of reproductive age who were not pregnant was 20.8%.[4] A preceding research stated that the prevalence of CED in pregnant women in Semarang District was 10.28%.[5] Another study conducted by Sumarmi also stated that the prevalence of CED among future brides in 4 sub-districts of Probolinggo District was 27.3%.[6]

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The impact of the incidence of CED among pregnant women in Indonesia includes the high rate of infant mortality (IMR) due to low birth weight (LBW) where the prevalence of LBW reached 10.2% in 2013.[7] Other impacts that can arise due to CED are bleeding, abnormal weight gain in pregnant women, and infectious diseases. Pregnant women with CED have a high risk of miscarriage, abortion, stillbirth, congenital defects, anemia in the baby, and die in the womb (Asphyxia intrapartum).[8]

The quality and quantity of diet are some of the factors that cause CED in preconception women. The quality of the diet is an important index to determine the intake of macro and micro-nutrients as well as dietary patterns that may affect the risk of diet-related diseases. According to researches that have been conducted in developing countries such as Indonesia and India, it is known that the quality of diet will affect the nutritional status of women of reproductive age (WRA) including brides.[9] Other studies have shown that a poor quality diet before pregnancy will affect the nutritional status of pregnant women which may lead to low birth weight (LBW) in children.[10]

The quality of one's diet is determined by the food security of the household, the better household food security, the better its diet quality. Food insecurity in WRA often results in the consumption of food that is not following their needs. This can cause a lack of energy, protein, and micronutrients which will affect the nutritional status of women of childbearing age, namely chronic energy deficiency (CED).[11,12]

Sumowono and Pringapus subdistricts are areas with a fairly high prevalence of infant mortality rate (IMR) and maternal mortality rate (MMR). Based on the Health Profile of Semarang District in 2016, there were 5 cases of MMR within 2011-2016. Meanwhile, during the same period, 70 and 49 cases of IMR were recorded in Sumowono and Pringapus districts respectively.[13] One of the risk factors of low birth weight (LBW) and also a major risk factor in infant mortality is CED during pregnancy.[14] The number of CED cases in Sumowono and Pringapus districts occurred due to the intake of protein and energy of their people which was relatively low compared to people in other areas in Central Java.[15] Therefore, this study aims to examine the relationship between household food security and diet quality with chronic energy deficiency (CED) in preconception women.

MATERIALS AND METHODS

This study was conducted between March-April 2018 in the Office of Religious Affairs (*Kantor Urusan Agama*/KUA) of Sumowono and Pringapus sub-districts, Semarang District. This is a cross-sectional study within the scope of Public Health Science. This study has been granted a permit by the Health Research Ethics Commission of the Faculty of Medicine, Universitas Diponegoro - Central Public Hospital Dr. Kariadi Semarang with No. 480 / EC / FK-RSDK / VII / 2018.

The minimum sample size was calculated using a correlative analytic formula and the 70 study participants were selected through the consecutive sampling method. The sample inclusion criteria include preconception women registered in the Office of Religious Affairs Sumowono and Pringapus sub-districts, aged 16-35 years, never and not currently pregnant (at the time of the data collection), and willing to fill in their information and sign a statement of willingness as research study participants.

The independent variables in this study were household food security and diet quality, while the dependent variable was chronic energy deficiency (CED). The cut-off point of CED is Mid-Upper Arm Circumference (MUAC) of <23.5 cm and has a BMI of <18.5 kg / m^2. The collected data in this study were study participants' general data, anthropometric data, household food security, and diet quality.

Household food security is a condition when people have the right to have physical and economic access at any time to obtain sufficient food to meet their needs for a productive and healthy life. The household food security was measured using the Household Food Security Scale Module (HFSSM) on study participants who live with no children aged under 18 years. Each of the questions from the HFSSM was assigned a 1 score if the study participants answered frequently or occasionally and a 0 score for never. Food security is categorized into 4 categories, namely high food security with 0 scores, moderate food security with a score of 1-2, low food security with a score of 3-5, and very low (food insecurity) with a score of 6-10.[16,17]

Diet quality is an assessment of food consumption that consists of 4 categories, namely variation, sufficiency, moderation, and overall balance based on dietary guidelines using the Diet Quality Index International (DQI-I) form. The total score of the DQI-I varies between 0 as the lowest score and 100 as the highest score. Diet quality is rated low if the score is ≤ 60 and is rated high if the score is > 60.[18] Data on the diet quality was collected through food intake interviews using the Semi-Quantitative Food Frequency (SQ-FFQ) form during the last month. Food photo books are used to minimize bias in intake data collection.

Data processing and analysis were carried by a computer program. Univariate analysis was performed to describe the characteristics of the subject. The bivariate analysis started with a normality test with the Kolmogorov-Smirnov test. Furthermore, bivariate analysis was carried out to determine the correlation between the independent and dependent variables with the Rank-Spearman and Pearson Product Moment tests.

RESULTS

Subject Characteristics

The participants' characteristics consist of data on age, education, occupation, nutritional status, household food security, and diet quality.

Table 1. Subjects characteristics

Characteristics	n	%
Age		
Adolescents (16-20 years old)	27	38.6
Adult (21-35 years old)	43	61.4
Educational Level		
Elementary School	8	11.4
Junior High School	23	32.9
Senior High School	29	41.4
Bachelor	10	14.3
Occupation		
Employment	45	64.3
Unemployment	25	35,7
Chronic Energy Deficiency (CED)		
CED (BMI <18.5kg/$m^{2,}$ MUAC< 23.5 cm)	11	15.7
Normal (BMI ≥18.5kg/$m^{2,}$ MUAC< 23.5 cm)	59	84.3
Risk of Chronic Energy Deficiency		
Risk of CED (MUAC< 23.5 cm)	34	48.6
Normal (MUAC ≥ 23.5 cm)	36	51.4
Household Food Security		
High Food Security	14	17.1
Medium Food Security	2	2.4
Low Food Security	53	75.1
Very low (Food Insecurity)	1	1.4
Diet Quality		
Low (score ≤60)	56	80
High (score >60)	14	20

A total of 38.6% of the study participants aged 16-20 years old, within the youth category. Most of the participants were junior and senior high school level, only 14.3% of the participants had an education up to diploma or bachelor's degree. The nutritional status based on BMI indicated that 11

participants (15.7%) suffered from CED and the other 84.3% of participants were normal The MUAC measurement showed that 48.6% of participants were at risk of CED and another 51.4% of the participants were normal. A total of 53 participants (75.1%) were in the low food security category. Most of the participants (80%) also had poor diet quality, and only 14 participants (20%) had good diet quality.

Table 2 describes the minimum, maximum, average, and median values of age, BMI, MUAC, household food security score, and diet quality score. The youngest study participant was 16 years old. The average BMI was 21.46 kg/m^2, which was normal. However, there was a participant who had a BMI score of 15.6 kg/m^2 which was classified as CED. On the other hand, there was a study participant who had a BMI score of 32.8 kg/m^2 which was classified as over-nutrition. The average MUAC score was 23.9 cm which was normal, but some study participants' MUAC were less than 23.5 cm. They were classified as at risk of CED. The median score of household food security was 3, which showed that most of the participants had low household food security. The average score of diet quality was 51.77, which was classified as low dietary quality.

Table 2. Minimum, maximum, average, and median values of age, BMI, MUAC, household food security score, and diet quality score

Variable	Minimum	Maximum	Mean±DS/Median
Age (years)	16	29	21±3.4[b]
BMI (kg/m^2)	15.6	32.8	21.45±3.23[b]
MUAC (cm)	17	30.7	23.8±2.83[b]
Household Food Security (score)	0	8	3[a]
Diet Quality (score)	28	75	52±11[b]

[a] Median [b] Mean (Deviation Standard)

Household Food Security

Table 3 showed that 72.9% of the participants were worried about not being able to buy food before they get another income and were only able to buy food to survive. A total of 57.1% of participants were unable to consume a balanced diet because they had no money. There were 21.4% of the participants who consciously reduced their eating portions due to financial constraints. Furthermore, there were 14.3% of the participants ate less than usual. Meanwhile, 5.7% of the participants were unable to buy food when they got hungry. A total of 17.1% of the participants lost their weight due to financial constraints to buy food, and 7.1% of the participants experienced not eating any food for the whole day because they did not have enough money to buy food.

Diet Quality

The diet quality consists of four aspects, i.e., variety, adequacy, moderation, and overall balance. Table 4 shows that the median score of the variation of the participants was 17 out of a maximum score of 20, which means that the participants were already consuming a variety of foods including overall variance and variety of protein. The minimum score (0) was given if a participant did not consume any fruit and vegetable. Moderation scores showed that the average consumption of total fat was 36%, which was relatively high compared to the recommendation of DQI-I, which is less than 30% energy needs. Additionally, the average score of saturated fat consumption was 16.9%, which was relatively high. It was also higher than the recommendation (less than 10%). The adequacy aspect, shown in table 5, consists of some components, i.e., vegetable, fruit, staple foods, fiber intake, protein intake, iron intake, calcium intake, and vitamin C intake. The results of this study showed that the consumption of vegetable, iron, and calcium groups was low, which lead to the low quality of the overall diet. On the other hand, the intake of staple food, protein, and vitamin C was adequate.

Table 3. Components of the HFSSM Questionnaire

Statements	Responses	n	%
Household Level			
Worried about not being able to buy food before getting next income	S, K	51	72.9
	T	19	27.1
Only buying food to survive and unable to stock on food	S, K	51	72.9
	T	19	27.1
Unable to consume a balanced diet	S, K	40	57.1
	T	30	42.9
Individual Level			
Having the experience to reduce an eating portion or skip meal time due to having not enough money to buy food	S, K	15	21.4
	T	55	78.6
Having the experience to eat less than you usually consumed due to having not enough money to buy food	S, K	10	14.3
	T	60	85.7
Feeling hungry, but do not have enough money to buy food	S, K	4	5.7
	T	66	94.3
Having an experience of weight loss due to having not enough money to buy food	S, K	12	17.1
	T	58	82.9
Having an experience to not eat all day long due to having not enough money to buy food	S, K	5	7.1
	T	65	92.9

S= happens often, K=sometimes happens, T= never happen

Table 4. Value of Minimum, Maximum, and Mean Diet Quality Subjects

Variable	Minimum	Maximum	Mean[a]±DS/Median[b]
Variety (score)	4	20	17[a]
Overall group food variety	1	5	4[a]
Within-group variety for Protein sources	1	6	6[a]
Adequacy (score)	11	38	22±6,3[b]
Vegetable group (servings/day)	0	4	0,5[a]
Fruit Group (servings/day)	0	12	1,2[a]
Grain Group (servings/day)	0.1	7.3	3.65±1.5[b]
Fiber intake (gr/day)	2	60.1	12.35[a]
Protein Intake (gr/day)	7.5	20	13.25±2.9[b]
Iron Intake (mg/day)	1.7	32.8	9.5[a]
Calcium Intake (mg/day)	41.1	1505.6	415.4[a]
vitamin C Intake (mg/day)	5.8	139.3	112.05[a]
Moderation (score)	3	24	12[a]
Total Fat (% total energy/day)	16.2	59	36±8.59[b]
Saturated Fat (% total energy/day)	3.6	37	16.9[a]
Cholesterol (mg/day)	26	921.9	240[a]
Natrium (mg/day)	94.5	4565.6	559.3[a]
Empty Calory Food (gr/day)	1	41	14±9.3[b]
Overall Balance (skor)	0	6	0.0[a]
Macronutrient Ratio (KH:P:L)	0	6	0.8[a]
Fatty Acid Ratio (PUFA:MUFA:SFA)	0	2	0.0[a]

An assessment by moderation category was used to evaluate food intake that indicated a link to chronic diseases and might need to be restricted, i.e. fats, saturated fats, cholesterol, sodium, and low-nutrient foods. This study showed that the moderation score was low, which means that the food intake of the participants was still not appropriate.

Table 6 shows the total consumption of fats, saturated fats, and junk food which then contributing to the score of the diet quality. According to the balanced nutritional guidelines, the intake of cholesterol and sodium of the participants was relatively good.

The last category was the balance category consisting of macronutrient balance and fatty acid ratio. The balance category evaluated the overall diet in terms of the proportion of energy sources and fatty acid composition. Tables 5 and 6 show that most participants consumed foods high in fat and enough for protein which led to the low intake of carbohydrates. Moreover, the intake of saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) were high (>10%). The recommendation stated that the intake of MUFA should be higher compared to the intake of PUFA.

Table 5. Description of Adequacy in Diet Quality

Variable	components	n	%
Adequacy			
Vegetable Group	Good (≥3-5 servings/day)	2	2.9%
	Adequate (<3-1.5 servings/day)	9	12.9%
	Less (<1.5 servings/day)	59	84.3%
Fruit Group	Good (≥2-3 servings/day)	22	31.4%
	adequate (<2-1 servings/day)	26	37.1%
	Less (<1 servings/day)	22	31.4%
Grain Group	Good (≥3-5 servings/day)	50	71.4%
	Adequate (<3-1.5 servings/day)	17	24.3%
	Less (<1.5 servings/day)	3	4.3%
Fiber Intake	Good (≥20-30 gram/day)	14	20%
	Adequate (<20-10 gram/day)	31	44.3%
	Less (<10 gram/day)	25	35.7%
Protein intake	Good (≥15% energi/day)	21	30%
	Adequate (<15-7.5 energy/day)	49	70%
Iron intake	Good (≥ 100 % RDA mg/day)	3	4.3%
	Adequate (<100%-50%RDA/day)	18	25.7%
	Less (<50 %RDA/day)	49	70%
Calcium Intake	Good (≥ 100 % RDA mg/day)	2	2.9%
	Adequate (<100%-50%RDA/day)	22	31.4%
	Less (<50 %RDA/day)	46	65.7%
Vitamin C intake	Good (≥ 100 % RDA mg/day)	45	64.3%
	Adequate (<100%-50%RDA/day)	11	15.7%
	Less (<50 %RDA/day)	14	20%
Carbohydrate Intake	Excessive (>100% carbohydrate need/day)	27	38.6%
	Adequate (80-100% (>100% carbohydrate need/day)	8	11.4%
	Less (<80% (>100% carbohydrate need/day)	35	50%
Energy intake	Excessive (>100% energy need/day)	27	38.5%
	Adequate (80-100% energy need/day)	18	25.7%
	Less (<80% energy need/day)	25	35.7%

Table 6. Description Moderation in Diet Quality

Variable	Components	n	%
Moderation			
Total Fat	Good (≤30% total energy/day)	17	24.3%
	excessive (>30% total energy/day)	53	75.7%
Saturated Fat	Good (≤10% total energy/day)	10	14.3%
	excessive (>10% total energy/day)	60	85.7%
Cholesterol	Good (≤300 mg/day)	44	62.9%
	excessive (>300 mg/dayi)	26	37.1%
Natrium	Good (≤2400 mg/day)	66	94.3%
	excessive (>2400 mg/day)	4	5.7%
Empty Calory Foods	Good (≤10% total energy/day)	22	31.4%
	excessive (>10% total energy/day)	48	68.6%

Relations between Household Food Security and Diet Quality with CED

Bivariate analysis was conducted to investigate the relationship between household food security and the quality of diet with CED in the study participants. Table 7 shows no significant link (p=0.537) between household food security and chronic energy deficiency.

Table 7. Relations between Household Food Security and Diet Quality with CED

Variable	CED	
	p	**r**
Household Food Security	0.537[b]	0.075
Diet Quality	0.711[a]	0.045

[a] Pearson [b]Rank Spearman

DISCUSSION

Study Participant Characteristics

This study consisted of 38.6% of adolescents (16-20) years. Marriage under the age of 20 years also occurred in Palestine where the prevalence of married women under the age of 18 was 41.4% while in India it was 44.5%.[19,20] Marriage under the age of 20 years is an important risk factor of adolescent pregnancy, where a pregnant young mother and suffers from CED would have a higher risk of perinatal death and give birth to low birth weight (LBW) babies.[21] Education was one of the underlying factors of early marriage in Indonesia.[22] Most of the participants were only graduated from junior and high school. Also, some of the participants only graduated from elementary school.

The prevalence of participants who were at risk of CED based on MUAC measurement was still high (48.6%). However, this prevalence was lower (15.7%) when assessed based on BMI measurement. The higher prevalence of CED based on MUAC measurement in this study was also reported by a preceding study conducted among brides in Probolinggo, which was only 27.3%, and a study conducted in Gorontalo, which was only 28.3%.[6,23] Meanwhile, the prevalence of CED in this study was lower compared to a study conducted in India which reported a prevalence of CED among preconception women of 32.1%.[21] We also found that 14.3% of the participants were over nutrition, which means that there was a double burden of malnutrition in this study.

Household Food Security

75.1% of the study participants in this study had low food security and 1.4% had food insecurity. The prevalence of low food security observed in this study is higher compared to the prevalence of food security among adult women in Malaysia which was only 43.5%.[24] The high prevalence of low food security may be affected by culture and habits among villagers who prioritize the food sufficiency of children. Besides children, working adults with an important role in the household, like fathers, become the main prioritize to get first and more food compared to other family members.[25]

The statement in the HFSSM questionnaire that contributed the highest score is the statement at the household level. A total of 72.9% of the participants were worried they could not buy food before they get another income and could only buy enough food to survive and not being able to stock up on food. Furthermore, 57.1% of the participants could not buy balanced food. This is due to 64.3% of the participants were working but only get low income as factory workers (regional minimum wage) in Semarang District. The other 35.7% of participants were unemployed thus dependent on their parents. The economy is the underlying factor for household food security. Low – middle-income households tend to have low food security. While high-income households often spend more on healthy food.[26]

Low income in a household and the number of family members that live in it will affect the household food security.[27] Low-income households with many dependent members lead to insufficient intake of each family member due to limited access to food. This condition was shown in the statement at the individual level where 21.4% of participants have experienced reducing of skipping a meal due to financial constraint. A total of 14.3% of participants have experienced eating less than usual and 5.7% of participants have experienced hunger due to financial constraints which led them to not be able to purchase food.

Diet Quality

Results show that the average score of diet quality among study participants was low. A total of 80% of the participants had a low-quality diet and the other 20% had high diet quality. The low-quality diet among participants was affected by the discrepancy between the nutrient intake and the recommendation.

The quality of diet consists of four categories which are variation, sufficiency, moderation, and overall balance. The variation evaluates the variation of the overall food and overall protein of the participants. Results show that in general, the participants consume varied food. Insufficiency, moderation, and overall balance categories, the results show a low mean which led to a low score of diet quality. Another study in Spain reported a similar finding, where female adolescents in the said study were found to consume varied food, but the other categories (sufficiency, moderation, and overall balance) contributed a low score to the total diet quality score.[28]

The sufficiency of vegetables, iron, and calcium in participants recorded a low mean. The average vegetable intake was only 0.5 portions/day compared to the recommended 3-5 portions/day. Similarly, a study in Spain also showed that the average vegetable and fruit intake among female adolescents was low.[28] Iron and calcium are important minerals needed during pregnancy. Most of the participants consumed iron and calcium-rich food which are not following the recommendation. A preceding study on pre-pregnancy women in Bogor supports this finding. The said study found that the iron and calcium intake pre-pregnancy among women was low.[29] Low intake of vegetable, fruit, and other micronutrients like iron and calcium can cause low diet quality among preconception women.[30]

The DQI-I score shows an imbalance of total fat, total saturated fat, and low-nutrients food compare to the requirement. Based on the DQI-I, the recommended consumption of total fat per day is <30% total energy/day, saturated fat is <10% total energy/day and low-nutrients food is <10% total energy/day. However, most of the participants still consumed high-fat and low-nutrient food like sugar, which contributes a low score to their diet quality score. A high intake of total and saturated fat resulted from the high consumption of fried food.

A low score was also found in the overall balance category. The high intake of fat among participants hinders the balance of other macronutrient intakes. Very few of the participants consumed total fat less than 30% of the total energy/day. This has an impact on the low intake of carbohydrates and protein. Results show that the consumption of carbohydrates and protein among the participants was relatively low compared to the recommendation even though many of the participants consumed sufficient protein. The high intake of fat increases the total energy intake which then affects the nutritional status.[31]

This study also reported an imbalance of fatty acids. The average intake of SFA was higher than PUFA and MUFA. This finding is in line with the findings in Iran which showed that the high intake of SFA was higher than PUFA and MUFA in female adolescents. This may be caused by the shift in the diet pattern where adolescents tend to consume

more high-energy foods like fast food and snack.[32] Fast food has become an easy and cheap option even though it contributes to excessive energy intake.

Relations between Household Food Security and Diet Quality with CED

Based on the test, household food security is not significantly correlated to CED among preconception women (p=0.536). Very limited studies evaluated household food security of preconception women using the HFSSM questionnaire. This study is supported by studies conducted in Bangladesh among pregnant women and in Kenya among women of reproductive age between 15-49 years which found no significant relations between household food security with nutritional status by BMI measurement.[33,34]Yet, the results of these studies contradicted the findings of a study conducted in Ethiopia which found a significant status between household food security and nutritional status by BMI measurement.[35]

In this study, household food security did not indicate low energy and protein intake. This can be seen from the high (and sometimes excessive) intake of energy and protein among the participants which was 64.2%. The high energy intake was acquired from sugar and fat intake and low fiber which tend to be low in micronutrients. Thus, the CED rate was not high as the participants were close to being over nutrient.

On the other hand, diet patterns in society also went through a shift where most households with low food security only paid attention to quantity and not the quality of the food. For example, by accessing fried food and low intake of vegetables and fruit. A study in Kenya backed this finding. It reported a shift in diet patterns in the household with low food security. Fried food was chosen because it is cheap and easy to consume compared to vegetables and fruit even though fried food is high in energy and is unhealthy.[34,36] Statements from most of the participants portray this by stating that they cannot consume a balanced diet due to financial constraints. Also, access to fried food is easier, as most of the participants work as factory workers where access to buy food was limited. A limited break time is a reason that the participants consumed an unhealthy diet that in prolonged time will cause overnutrition.[37]

Very limited studies evaluated the diet quality of preconception women using the DQI-I questionnaire. The overall diet quality of the participants in this study did not show a significant correlation with CED (p=0.711). This finding is supported by findings of other studies conducted in the USA among female adolescents which explains the quality diet of female adolescents did not have a significant relationship with their nutritional status based on BMI.[38] However, this finding contradicts a preceding study that reported a significant relationship between diet quality and BMI before pregnancy among pregnant women.[1] Another direct factor that may cause CED in preconception women is infection. During infection or another sickness, we tend to lose appetite thus lead to malnutrition. If it continues for a prolonged time, it can cause CED.[39,40]

This disagreement may happen because CED in preconception women is not only affected by household food security and diet quality. Energy and protein intake plays an important role in the incidence of CED among preconception women. A low protein and energy intake for a prolonged time can cause CED.[23] In this study, most of the participants had sufficient energy intake, moreover, 38.5% of the participants had excessive energy intake. Looking at the sufficiency of staple food, the average consumption of staple food of a participant was 3.65 portions/day and is categorized as sufficient. Also, the protein intake of the participants was categorized as sufficient and good, thus it can be concluded that by quantity the intake of the participants was adequate.

This study differs from the Central Java Individual Food Consumption Survey in 2014 which stated that women who live in the village tend to have low consumption of energy and protein thus causing malnutrition. However, in this study, the participants had high energy high-fat diets thus tend to be over nutrition. This is supported by the result that shows 14.35% of the participants had over nutrition. Diets that tend to be over nutritious is a low vegetable and fruit yet high fat intake.[15,28]

Diet quality is influenced by household food security, where a family with high food security will have a good diet quality.[12,41] A low household food security caused most of the participants in this study to consume cheap and easily accessed food like fried food. It was shown by the high intake of fat among

the participants in the moderation category which was more than 30% of total energy. A low moderation score will affect the overall balance score where most of the participants had a high intake of fat and impair the balance of other macronutrients. A high intake of fat will contribute to excessive energy thus cause a build-up of fat on the adipose tissue and plays an important role in the incidence of overnutrition.[42]

CONCLUSIONS

A total of 48.1% of participants were at risk of CED, but only 15% of preconception women were categorized as CED. The household food security of the participants was mostly low (75.1%). Most of the participants (80%) had low-quality diets where the mean diet quality was 51.77. there was no significant correlation between household food security and diet quality with CED.

REFERENCES

1. Shin D, Lee KW, Song WO. Pre-Pregnancy Weight Status Is Associated with Diet Quality and Nutritional Biomarkers during Pregnancy. Nutrients. 2016 Mar 11;8(3):1–12.
2. Stephanie P, Kartika SKA. Gambaran Kejadian Kurang Energi Kronik dan Pola Makan Wanita Usia Subur di Desa Pesinggahan Kecamatan Klungkung Bali 2014. E-Jurnal Medika Udayana. 2016 May 31;5(6):1–6.
3. Das S, Bose K. Body Mass Index, and Chronic Energy Deficiency among Adult Santals of Purulia District, West Bengal, India. Journal of Human Sciences. 2010 Jul 25;7(2):488–503.
4. Kementerian Kesehatan RI. Riset Kesehatan Dasar 2013 (RISKESDAS 2013). Indonesia: Kementerian Kesehatan Republik Indonesia; 2013.
5. Budiono I. Pengembangan Model Indeks Pembangunan Gizi. KEMAS: Jurnal Kesehatan Masyarakat. 2013 Jan 3;8(2):113–20.
6. Putri SI, Sumarmi S. Perbandingan Konsumsi Zat Gizi, Status Gizi, dan Kadar Hemoglobin Pengantin Wanita di Wilayah Pantai dan Pertanian Kabupaten Probolinggo. Media Gizi Indonesia. 2012 Jan;9(1):1–6.
7. Kementerian Kesehatan Republik Indonesia. Pusat Data dan Informasi Kementrian Kesehatan RI. Kementerian Kesehatan Republik Indonesia; 2014.
8. Rahmi L. Faktor-Faktor yang Berhubungan dengan Kekurangan Energi Kronik (KEK) pada Ibu Hamil di Puskesmas Belimbing Padang Tahun 2016. Jurnal Kesehatan Medika Saintika. 2017 Jun 12;8(1):35–46.
9. Arimond M, Elin L, Wiesmann D, Joseph M, Carriquiry A. Dietary Diversity as a Measure of Women's Diet Quality in Resource-Poor Areas: Results from Rural Bangladesh Site. Food and Nutrition Technical Assistance. 2008;58.
10. Potdar RD, Sahariah SA, Gandhi M, Kehoe SH, Brown N, Sane H, et al. Improving women's diet quality preconceptionally and during gestation: effects on birth weight and prevalence of low birth weight--a randomized controlled efficacy trial in India (Mumbai Maternal Nutrition Project). Am J Clin Nutr. 2014 Nov;100(5):1257–68.
11. Na M, Mehra S, Christian P, Ali H, Shaikh S, Shamim AA, et al. Maternal Dietary Diversity Decreases with Household Food Insecurity in Rural Bangladesh: A Longitudinal Analysis. J Nutr. 2016 Oct;146(10):2109–16.
12. Abraham S, Miruts G, Shumye A. Magnitude of chronic energy deficiency and its associated factors among women of reproductive age in the Kunama population, Tigray, Ethiopia, in 2014. BMC Nutrition. 2015 Jun 22;1(1):12.
13. Kementerian Kesehatan Republik Indonesia. Profil Kesehatan Provinsi Jawa Tengah Tahun 2016. Kementerian Kesehatan Republik Indonesia; 2016.
14. Kementerian Kesehatan Republik Indonesia. Profil Kesehatan Kabupaten Semarang 2014. Kementerian Kesehatan Republik Indonesia; 2014.
15. Santoso B, Sulistiowati E, Fajarwati T, Pambudi J. Survei Konsumsi Makanan Individu Provinsi Jawa Tengah 2014. Semarang: Badan Penelitian dan Pengembangan; 2014.
16. Usfar AA, Fahmida U, Februhartanty J. Household food security status measured by the US-Household Food Security/Hunger Survey Module (US-FSSM) is in line with coping strategy indicators found in urban and rural Indonesia. Asia Pac J Clin Nutr. 2007;16(2):368–74.
17. Adereti DT, Fasina OO. Gender analysis of food security status of rural households in Ondo

State, Nigeria. Russ Agricult Sci. 2017 Jul 1;43(4):353–60.
18. Guerrero MLP, Pérez-Rodríguez F. Diet Quality Indices for Nutrition Assessment: Types and Applications. In: Functional Food - Improve Health through Adequate Food [Internet]. IntechOpen; 2017 [cited 2020 Nov 16]. Available from: https://www.intechopen.com/books/functional-food-improve-health-through-adequate-food/diet-quality-indices-for-nutrition-assessment-types-and-applications
19. Ghrayeb FA, Rusli A M, Ismail I M, Ghrayeb NF, Al Rifai A. Prevalence of Early Marriage among Women in Rural Palestinian Community: A Cross-Sectional Study. International Medical Journal. 2015 Oct 3;22(4):291–4.
20. Raj A, Saggurti N, Balaiah D, Silverman JG. Prevalence of Child Marriage and its Impact on the Fertility and Fertility Control Behaviors of Young Women in India. Lancet. 2009 May 30;373(9678):1883–9.
21. Islam A, Islam N, Bharati P, Aik S, Hossain G. Socio-economic and demographic factors influencing nutritional status among early childbearing young mothers in Bangladesh. BMC Women's Health. 2016 26;16(1):58.
22. Rumble L, Peterman A, Irdiana N, Triyana M, Minnick E. An empirical exploration of female child marriage determinants in Indonesia. BMC Public Health. 2018 Mar 27;18(1):407.
23. Hubu N, Nuryani N, Hano YH. Pengetahuan, Asupan Energy dan Zat Gizi Berhubungan dengan Kekurangan Energy Kronis pada Wanita Prakonsepsi. GJPH. 2018 Apr 19;1(1):015.
24. Atiqah AN, Norazmir M, Anuar MK, Fahmi MM, Norazlanshah H. Food security status: it's association with inflammatory marker and lipid profile among young adult. [Internet]. undefined. 2015 [cited 2020 Nov 16]. Available from: /paper/Food-security-status%3A-it%27s-association-with-marker-Atiqah-Norazmir/557339a0f893c6e1e813a700a3ad236a335e5b50
25. Cordeiro LS, Wilde PE, Semu H, Levinson FJ. Household food security is inversely associated with undernutrition among adolescents from Kilosa, Tanzania. J Nutr. 2012 Sep;142(9):1741–7.
26. Hackett M, Melgar-Quiñonez H, Taylor CA, Alvarez Uribe MC. Factors associated with the household food security of participants of the MANA food supplement program in Colombia. Arch Latinoam Nutr. 2010 Mar;60(1):42–7.
27. Ahmed UI, Ying L, Bashir MK, Abid M, Zulfiqar F. Status and determinants of small farming households' food security and role of market access in enhancing food security in rural Pakistan. PLoS One. 2017;12(10):e0185466.
28. Mariscal-Arcas M, Romaguera D, Rivas A, Feriche B, Pons A, Tur JA, et al. Diet quality of young people in southern Spain evaluated by a Mediterranean adaptation of the Diet Quality Index-International (DQI-I). Br J Nutr. 2007 Dec;98(6):1267–73.
29. Madanijah S, Briawan D, Rimbawan R, Zulaikhah Z, Andarwulan N, Nuraida L, et al. Nutritional status of pre-pregnant and pregnant women residing in Bogor district, Indonesia: a cross-sectional dietary and nutrient intake study. Br J Nutr. 2016;116 Suppl 1:S57-66.
30. Shamim AA, Mashreky SR, Ferdous T, Tegenfeldt K, Roy S, Rahman AKMF, et al. Pregnant Women Diet Quality and Its Sociodemographic Determinants in Southwestern Bangladesh. Food Nutr Bull. 2016 Mar;37(1):14–26.
31. Austin GL, Ogden LG, Hill JO. Trends in carbohydrate, fat, and protein intakes and association with energy intake in normal-weight, overweight, and obese individuals: 1971-2006. Am J Clin Nutr. 2011 Apr;93(4):836–43.
32. Mohseni-Takalloo S, Mirmiran P, Hosseini-Esfahani F, Azizi F. Dietary Fat Intake and Its Relationship with Serum Lipid Profiles in Tehranian Adolescents. JFNR. 2014 Jun 16;2(6):330–4.
33. Mridha MK, Matias SL, Arnold CD, Dewey KG. Factors associated with nutritional status and dietary practices of Bangladeshi adolescents in early pregnancy. Ann N Y Acad Sci. 2018 Feb 18;1416:66–76.
34. Keino S, Plasqui G, van den Borne B. Household food insecurity access: a predictor of overweight and underweight among Kenyan

women. Agriculture & Food Security. 2014 Jan 28;3(1):2.
35. Abdu J, Kahssay M, Gebremedhin M. Household Food Insecurity, Underweight Status, and Associated Characteristics among Women of Reproductive Age Group in Assayita District, Afar Regional State, Ethiopia [Internet]. Vol. 2018, Journal of Environmental and Public Health. Hindawi; 2018 [cited 2020 Nov 16]. p. e7659204. Available from: https://www.hindawi.com/journals/jeph/2018/7659204/
36. Gulliford MC, Mahabir D, Rocke B. Food insecurity, food choices, and body mass index in adults: nutrition transition in Trinidad and Tobago. Int J Epidemiol. 2003 Aug;32(4):508–16.
37. Nobrega S, Champagne N, Abreu M, Goldstein-Gelb M, Montano M, Lopez I, et al. Obesity/Overweight and the Role of Working Conditions: A Qualitative, Participatory Investigation. Health Promot Pract. 2016 Jan;17(1):127–36.
38. Feskanich D, Rockett HRH, Colditz GA. Modifying the Healthy Eating Index to assess diet quality in children and adolescents. J Am Diet Assoc. 2004 Sep;104(9):1375–83.
39. Dobner J, Kaser S. Body mass index and the risk of infection - from underweight to obesity. Clin Microbiol Infect. 2018 Jan;24(1):24–8.
40. Katona P, Katona-Apte J. The Interaction between Nutrition and Infection. CLIN INFECT DIS. 2008 May 15;46(10):1582–8.
41. Leung CW, Epel ES, Ritchie LD, Crawford PB, Laraia BA. Food Insecurity Is Inversely Associated with Diet Quality of Lower-Income Adults. Journal of the Academy of Nutrition and Dietetics. 2014 Dec;114(12):1943-1953.e2.
42. Coelho DF, Pereira-Lancha LO, Chaves DS, Diwan D, Ferraz R, Campos-Ferraz PL, et al. Effect of high-fat diets on body composition, lipid metabolism and insulin sensitivity, and the role of exercise on these parameters. Brazilian Journal of Medical and Biological Research. 2011 Oct;44(10):966–72.

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Correlation between wrist circumference with blood pressure and creatinine level among elderly

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ABSTRACT

Background: Hypertension is a highly prevalent health problem which incidence is greatest among the elderly. Hypertension may increase creatinine level and leads to other health problems like diabetes mellitus, kidney damage, and cardiovascular disease. Wrist circumference is a simple anthropometric measurement that can be used to identify hypertension and increasing level of serum creatinine.
Objectives: To analyze the correlation of wrist circumference with blood pressure and creatinine level among the elderly.
Materials and Methods: This was a cross-sectional study with a purposive sampling method. Subjects of this study were 84 women aged 60 years old or above at Unit Rehabilitasi Sosial Pucang Gading Semarang. The independent variable of this study was wrist circumference, and the dependent variables were systolic blood pressure, diastolic blood pressure, and creatinine level. The result was analyzed using the Spearman-rho test.
Results: The participants of this research were 49% women aged 60-65 years old, with an average age was 65.5 years old. The prevalence of hypertension was 61.9%. Most hypertension incidence in this research was caused by high systolic blood pressure (50%), and the rest was caused by high diastolic blood pressure (3.9%) and both (46.1%). The level of creatinine was normal with an average level was 0.75 mg/dL. There was no correlation of wrist circumference with systolic blood pressure systolic (r=0.15; p=0.19), diastolic blood pressure (r=0.1; p=0.38), and creatinine serum (r=0.18; p=0.09) among elderly.
Conclusions: There was no correlation of wrist circumference with blood pressure and creatinine level among the elderly.

Keywords: Wrist circumference; Blood pressure; Creatinine level; Elderly

BACKGROUND

According to World Health Organization (WHO), the elderly are individuals with the chronological age of 60 years or more. The Elderly is the last phase of the human life cycle and is initiated by the aging process. This aging process is contributing to frailty which posing the elderly to diseases, both infectious and non-infectious ones.[1] Elderly population in Indonesia is 9.7% based on Riskesdas 2019 and was predicted to be increased more than 2 folds in 2050. Based on Riskesdas 2018 data, the most prevalent disease for the elderly in Indonesia is hypertension.[2]

Hypertension is a condition in which a person has systolic blood pressure ≥140 mmHg and/or diastolic blood pressure ≥of 90 mmHg.[3] Hypertension increase the risk of diabetes and coronary heart disease. Moreover, hypertension could lead to chronic kidney disease through the increased blood creatinine level.[4] Prevalence of hypertension in Indonesia is 34.1% where 59.4% of citizens with the age of >54 years are having hypertension.[2] Hypertension prevalence in Central Java is 37.6%, even higher than national prevalence.[5] Health Department of Central Java stated hypertension prevalence in Semarang City in 2018 is 6.3%, whereas much as 14.9% of the citizen with the age of >45 years are having hypertension.[6]

Hypertension is preventable and treatable by early detection. Hypertension is usually detected by performing a blood pressure examination using a sphygmomanometer. However, a simple anthropometry measurement that is quick and easy could be considered as a parameter to detect hypertension, one of them is wrist circumference.[7-9] Measurement of waist circumference is easier to be

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done since it does not require ratio calculations and trained individuals.[10] Inflammation state in hypertension indirectly decreases the bone mineral density, including on wrist circumference.[9]

Besides as a predictor of hypertension, many studies show the correlation between waist circumference and creatinine levels. A large-scale study conducted on 2.400 respondents in Iran shows that simple anthropometric measurement as wrist and neck circumference has a significant correlation with creatinine level and able to predict the risk of chronic kidney disease in adolescents and adults.[11]

The population-based study associated with the correlation between waist circumference with blood pressure and creatinine levels in Indonesia is still limited, especially among the elderly in Semarang City. The purpose of this study is to analyze whether waist circumference is associated with blood pressure and creatinine levels among the elderly.

MATERIALS AND METHODS

This study is an observational study with a cross-sectional design among 84 elderly in Unit Rehabilitasi Sosial Pucang Gading Semarang from May to July 2019. Samples were collected via the purposive sampling method. Institutional Review Board of Ethical Clearance (*Komisi Etik Penelitian Kesehatan*/KEPK) from the Medical Faculty of Diponegoro University has approved this study and publish ethical clearance141/EC/FK-RSDK/1V/2018.

Inclusion criteria in this study are women with the age of 60 years or more, communicative, and willing to be involved in this study by signing informed consent. Exclusion criteria are the history of/currently taking long-term corticosteroid medication, routinely consuming calcium supplements, and having chronic diseases such as diabetes, kidney failure, liver disease, gastrointestinal disturbance, and thyroid dysfunction.

The dependent variable in this study is wrist circumference. Wrist circumference measurement was performed using measuring tape with 0,1 cm accuracy. The measuring tape was wrapped around the wrist from the Lister tubercle on the distal radius to the distal ulnae on the right hand in a sitting position. The independent variables are blood pressure and creatinine level. Blood pressure measurement was performed twice using a digital sphygmomanometer. Blood sampling was performed by laboratory personnel and creatinine levels were tested using a photometer automatic chemistry analyzer.

Data analysis was performed using the computer-based statistic program SPSS, consisting of univariate and bivariate analysis. Univariate analysis was performed to describe the subject's characteristics such as age, body mass index (BMI), blood pressure, and creatinine levels. Bivariate analysis was performed to analyze the correlation between waist circumference with blood pressure and creatinine levels by the Spearman correlation test. Coefficient correlation (r) approaching 0 shows a weak correlation among variables and r value approaching -1 or 1 shows a strong correlation. P-value <0.05 shows a significant correlation between variables.

RESULTS

Table 1 shows the anthropometry and biochemistry characteristics of the subjects in this study. Based on Table 1, most of the subjects are 60-65 years old (58.3%) with an average of 65.5 years old. Forty-three subjects (51.2%) are obese.

The prevalence of hypertension in this study is 61.9%. As much as 50% of hypertension in the subjects of this study is marked by high systolic pressure. Hypertension with high diastolic blood pressure is only 3.9% and the rest (46.1%) is caused by high systolic and diastolic blood pressure. The biochemical test shows creatinine levels on most subjects are still in the normal range with an average of 0.75 mg/dL.

Table 2 shows the correlation between waist circumference and systolic blood pressure, diastolic blood pressure, and creatinine level to analyze the strength and direction of the variables.

Statistical analysis shows there is no correlation between waist circumference and systolic blood pressure, diastolic blood pressure, and creatinine level with a p-value > 0.05.

DISCUSSION

The prevalence of hypertension in this study, whether based on systolic or diastolic blood pressure or both, is 61.9%. This result is in accordance and able to show highly prevalent hypertension among the elderly in Indonesia that reach 59.4% in 2018.[2] This number shows the importance of a quick and good indicator to detect hypertension.

Previous studies show that waist circumference correlated with hypertension and increased creatinine level.[11-13] One of the factors influencing bone growth is a hormone called insulin-like growth factor 1 (IGF-1). This hormone is

stimulated by growth hormone and synthesized in the liver. Insulin-like growth factor 1 stimulates nitric oxide (NO) formation by endothelial cells and cells in blood vessels smooth muscle and stimulates Na-K-ATPase pump, therefore causing blood vessels relaxation and decrease blood pressure.[14,15] Wrist circumference which is one of the indicators of bone surface dimension could be used to predict hypertension risk and increased creatinine level. [16]

Table 1. Baseline Characteristics

	Numbers (%)
Total subjects	84
Age (years)	
60 – 65	49 (58.3%)
>65	35 (41.7%)
Body Mass Index (kg/m^2)	
< 18.5	2 (2.4%)
18.5 – 22.9	30 (35.7%)
23 – 24.9	9 (10.7%)
≥ 25	43 (51.2%)
Systolic Blood Pressure (mmHg)	
Normal	34 (40.5%)
Hypertension	50 (59.5%)
Diastolic Blood Pressure (mmHg)	
Normal	58 (69.1%)
Hypertension	26 (30.9%)
Creatinine Level (mg/dL)	
Normal	80 (95.2%)
High	4 (4.8%)
Wrist Circumference (cm)	15.35 ± 0.14

Note: BMI: Body Mass Index

Table 2. Correlation between wrist circumference with blood pressure and creatinine level

Variable	**Coefficient correlation (r)[a]**	**Sig. (*p-value)***
Systolic blood pressure (mmHg)	0.15	0.19
Diastolic blood pressure (mmHg)	0.10	0.38
Creatinine level (mg/dL)	0.18	0.09

Note: [a]pearman test

Physiologically, aging will cause the thickening and hardening of blood vessels. This condition is associated with chronic inflammation in blood vessels and increased oxidative stress thus decreasing the elasticity of blood vessels and inhibits blood flow. The blockaded blood vessels will increase heart workload in pumping blood, therefore increase blood pressure.[17] Blocked blood vessels in the kidney will cause a decreased glomerular filtration rate. This will result in decreased creatinine excretion and the risk of increased creatinine in blood among the elderly.[18]

There was not found a significant correlation between waist circumference with blood pressure and creatinine level in this study. This was presumably caused by the influence of several factors such as physical activity, food intake, previous history of the disease, and other factors that were not included in this study.

Increased physical activity will decrease the catecholamine level in the blood. This condition will indirectly decrease the sympathetic nervous system which responds to the decreased blood flow rate and vasodilatation.[19] Moreover, blood pressure and creatinine levels are easily influenced by food intake. Elderly with poor food intakes such as consuming high calories food, high sodium, protein, fat and low in potassium and fiber will increase the risk of increased blood pressure and creatinine level.[20-25] High sodium and low potassium food will influence fluid volume and blood vessel elasticity which contribute to increased blood pressure.[20, 21]

High protein intake, especially animal-based protein is associated with hypertension and kidney failure.[22] Fiber is known for its potency in increasing insulin sensitivity and endothelial cells which will contribute to blood vessel elasticity.[23]

High fat and dense calorie food will increase excess caloric intake and fat deposition in blood vessels that will result in blocked blood flow from and to organs, and disturbing its physiological condition.[24, 25] Obesity and hyperglycemia will increase the risk of increased blood pressure and blood creatinine level. Inflammation caused by obesity and hyperglycemia will increase free radicals and decrease adiponectin, and result in the damage and elasticity impairment of the blood vessels.[24,25] This could also increase blood pressure and blood creatinine levels if kidney blood vessels are affected.[17, 18]

CONCLUSIONS

Wrist circumference is not correlated with blood pressure and creatinine levels in this study. This is presumably caused by several factors such as physical activity, food intake, and previous history of the disease which could influence the correlation between waist circumference with blood pressure and creatinine level.

Further studies should be carried out to validate the correlation between waist circumference with blood pressure and creatinine level among the elderly by considering the food intake and physical activity of the elderly.

REFERENCES

1. World Health Organization. World Report on Ageing and Health. Geneva, Switzerland: WHO; 2015.
2. Kementerian Kesehatan RI. Laporan Nasional Riset Kesehatan Dasar 2018. Kementerian Kesehatan RI. Jakarta:2018.
3. Chobanian A V, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL, et al. The Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. Journal of the American Medical Association. 2003;289:2560-2571.
4. Ghaffari S, Pourafkari L, Tajlil A, Sahebihagh MH, Mohammadpoorasl A, Tabrizi JS, et al. The Prevalence, Awareness, and Control Rate of Hypertension among Elderly in Northwest of Iran. Journal of Cardiovascular and Thoracic Research. 2016;8(4):176–82.
5. Dinas Kesehatan Jawa Tengah. Profil Kesehatan Provinsi Jawa Tengah Tahun 2018. Semarang, Jawa Tengah: Dinkes Jateng; 2018.
6. Dinas Kesehatan Kota Semarang. Profil Kesehatan Kota Semarang Tahun 2018. Semarang, Jawa Tengah: DKK Semarang; 2018.
7. Kalantari S, Khalili D, Asgari S, Fahimfar N, Hadaegh F, Tohidi M, et al. Predictors of Early Adulthood Hypertension during Adolescence: A Population-based Cohort Study. BMC Public Health. 2017;17(1):1–8.
8. Zampetti S, Campagna G, Lucantoni F, Marandola L, D'Onofrio L, Chiesa C, et al. Wrist Circumference is Associated with Increased Systolic Blood Pressure in Children with Overweight/Obesity Article. Hypertension Research. 2018;41(3):193–7.
9. Mohebi R, Mohebi A, Sheikholeslami F, Azizi F, Hadaegh F. Wrist Circumference as A Novel Predictor of Hypertension and Cardiovascular Disease: Results of A Decade Follow Up in A West Asian Cohort. Journal of the American Society of Hypertension. 2014;8(11):800–7.
10. Shafiee G, Qorbani M, Heshmat R, Djalalinia S. Wrist Circumference as a Novel Predictor of Obesity in Children and Adolescents: the CASPIAN-IV Study. Journal of pediatric endrocinology & metabolism. 2018;31(7):1-9.
11. Mehrkash M, Heshmat R, Qorbani M, Motlagh ME, Djalalinia S, Zamani S, et al. Association of Neck, Wrist and Hip Circumferences with Kidney Function in Children and Adolescents: The CASPIAN- V Study. Journal of Pediatric Research. 2019;6(3):234–41.
12. Mitrea A, Soare A, Popa SG, Tudor MN, Mota M, Pozzilli P. Wrist Circumference: an Independent Predictor of Both Insulin Resistance and Chronic Kidney Disease in an Elderly Population. Romanian Journal of Diabetes Nutrition and Metabolic Disease. 2013;20(3):323–9.
13. Li Y, Liu Y, He J, Ma P, Yu L, Sun G. The Association of Wrist Circumference with Hypertension in Northeastern Chinese Residents in Comparison with other Anthropometric Obesity Indices. PeerJ. 2019;2019(8):1–16.
14. Hunt KJ, Lukanova A, Rinaldi S, Lundin E, Norat T, Palmqvist R, et al. A Potential Inverse Association Between Insulin-Like Growth Factor I and Hypertension in a Cross-Sectional Study. Annals of Epidemiology. 2006;16(7):563–71.

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15. Zhang L, Curhan GC, Forman JP. Plasma Insulin-like Growth Factor-1 Level and Risk of Incident Hypertension in Nondiabetic Women. Journal of Hypertension. 2011;29(2):229–35.
16. Masoud MI, Masoud I, Kent RL, Gowharji N, Hassan AH, Cohen LE. Relationship between Blood-Spot Insulin-like Growth Factor 1 Levels and Hand-Wrist Assessment of Skeletal Maturity. American Journal of Orthodontics and Dentofacial Orthopedics. 2009;136(1):59–64.
17. Buford TW. Hypertension and Ageing. Ageing Research Reviews. 2016; 26: 96-111.
18. Weinstein JR, Anderson S. The Aging Kidney: Physiological Change. Advances in Chronic Kidney Disease. 2010; 17(4): P302-307.
19. Kokkinos PF, Giannelou A, Athanasiosmanolis, Pittaras A. Physical Activity in the Prevention and Management of High Blood Pressure. Hellenic Journal of Cardiology. 2009;50:52-9.
20. Appel LJ. The Effects of Dietary Factors on Blood Pressure. Cardiology Clinics. 2017;35(2):197–212.
21. Naismith DJ, Braschi A. The Effect of Low-Dose Potassium Supplementation on Blood Pressure in Apparently Healthy Volunteers. British Journal of Nutrition. 2003;90(1):53-60.
22. Kamper A-L, Strandgaard S. Long-Term Effects of High-Protein Diets on Renal Function. Annual Review of Nutrition. 2017;37(1):347–69.
23. Streppel MT, Arends LR, Veer Pvt, Grobbee DE, Geleijnse JM. Dietary Fiber and Blood Pressure: A Meta-analysis of Randomized Placebo-Controlled Trials. Arch Intern Med. 2005;165:150-6.
24. Kim DH, Kim C, Ding EL, Townsend MK, Lipsitz LA. Adiponectin Levels and the Risk of Hypertension: a Systematic Review And Meta-Analysis. Hypertension. 2013;62(1):27-32.
25. Donmezi S, Gokalp O, Dogan M, Vural H, Yigit B. Effects of Short-Term Hyperglycemia on the Vasoconstriction of the Aorta. Turkish Journal of Medical Sciences. 2014;44:941-5.

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Catfish (Clarias sp.) as an animal protein source to improve serum albumin levels of hemodialysis patients

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ABSTRACT

Background: Hemodialysis patients often experience hypoalbuminemia complications, which occur mainly due to decreased synthesis due to inflammation, lack of protein intake, the fluid status of patients, and losses from the dialysate. Another problem in hemodialysis is malnutrition, with a prevalence between 23–73% globally. Gastrointestinal disorders such as nausea, vomiting, and decreased appetite also often occur in hemodialysis. Therefore, hemodialysis patients need to get nutritional support, which can be given in the form of catfish abon, one of the local Indonesian food.
Objective: To determine the effectiveness of the use of catfish as a source of animal protein to improve the albumin levels of hemodialysis patients.
Materials and Methods: This was a quasi-experimental study with a pre-post test design. This study involved 34 hemodialysis patients as subjects, with inclusion criteria, were routinely two times a week, aged >18 years, willing to be the subject and follow the research procedures, have albumin levels ≥3.0 g/dL, and no catfish allergies. Patients with anasarca edema, experiencing complications of diabetes mellitus and malignancy were excluded. The dependent variable was albumin content, while the independent variable was catfish as an animal protein source. Data were analyzed univariate and bivariate by Fisher's Exact test.
Results: Fisher's Exact test results on the effectiveness of using catfish as an animal protein source to improve albumin levels of hemodialysis patients showed p-value=0.048.
Conclusion: The use of catfish as an effective animal protein source significantly affected on improving albumin levels in hemodialysis patients.

Keywords: Nutritional support; Catfish (Clarias sp.); Albumin levels; Hemodialysis patients.

BACKGROUND

Chronic kidney disease is a global widespread epidemic disease, which a prevalence rate is 5-15%. The incidence rate of end-stage renal disease patients requiring dialysis is also increasing (1). Basic Health Research in Indonesia shows that the prevalence of chronic kidney disease nationally increased from 0.2% in 2013 to 0.38% in 2018. Province Special Region of Yogyakarta, where the Panembahan Senopati Hospital is located, ranked 12th nationally for the prevalence of chronic kidney disease (2).

Panembahan Senopati Bantul Hospital is a large type B hospital, which obtained a hospital-level plenary accreditation certificate. Panembahan Senopati Bantul Hospital is one of the hospitals that has a Hemodialysis Unit in Bantul Regency. Based on data from the 2013 Panembahan Senopati Bantul General Hospital annual report, the number of chronic kidney disease patients undergoing hemodialysis is increasing every year. In 2011, the number of routine hemodialysis patients was 111 people, in 2012, it increased to 125 people, in 2013, it became 142 people, and in 2014 it increased again to 144 people. The hemodialysis patient is accommodated in a room (Hemodialysis Unit) with a capacity of 22 beds.

Hemodialysis patients often experience hypoalbuminemia complications, which occur mainly due to decreased synthesis due to inflammation, lack of protein intake, the fluid status of patients, and losses from dialysate (3,4). Another problem that often arises in hemodialysis is malnutrition, with a prevalence between 23–73%

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globally (5). Gastrointestinal disorders such as nausea, vomiting, and decreased appetite also often occur in hemodialysis patients (6).

Protein-energy malnutrition is a major risk factor for mortality and inflammation; the presence of co-morbid conditions like cardiovascular disease increases this risk further (7). Both low protein intake and a high state of inflammation are associated with low serum albumin in maintenance hemodialysis patients (8). Therefore, hemodialysis patients need to get nutritional support, one of them with catfish.

Catfish have many advantages in terms of nutritional content but are still often underestimated by the community because of conventional processing. Catfish can be processed into various types of food products, one of which is catfish *abon*, so it is expected to increase the acceptability and intake of hemodialysis patients. *Abon* is one of the local food in Indonesia. This study was conducted to determine the effectiveness of the use of catfish as a source of animal protein to improve the albumin levels of hemodialysis patients.

MATERIALS AND METHODS

This research was conducted at the Hemodialysis Unit of Panembahan Senopati Bantul Hospital in January–December 2017. This research type was a quasi-experimental design with a pre-post design. This study involved 34 subjects with inclusion criteria for routine hemodialysis patients twice a week, aged >18 years, willing to be the subject of research and follow the research procedures, have albumin levels ≥3.0 g/dL, and there was no catfish allergy. Whereas patients with anasarca edema, complications of diabetes mellitus, and malignancy were excluded from the study.

An experiment in the form of providing nutritional support for catfish *abon* for 21 days, based on the half-life of albumin. The nutritional content of catfish *abon* which is given as nutritional support is shown in Table 1. With a protein requirement of 1.2 g/kg body weight per day, nutritional support for hemodialysis patients can be given protein additions of ±0.2 g/kg body weight per day. The protein content of catfish *abon* based on proximate test results is 54.89 grams per 100 grams of catfish *abon*. With the nutritional support needs of hemodialysis patients of 0.2 g/kg ideal body weight/day, the need for *abon* is 0.36 g/kg ideal bodyweight/day.

Table 1. Nutrient content of catfish *abon* in the study

Nutrient	Unit	Nutrient content
Energy	kcal	421.294
Protein	g	54.894
Fat	g	25.159
Carbohydrate	g	326.091
Crude fiber	g	0.127
Sodium	ppm	1619.25
Potassium	ppm	421.294

Before being given an intervention (pre-intervention), subjects first measured levels of albumin. Then the subjects were given the intervention to provide nutritional support in the form of catfish *abon* for 21 days and were asked to record all the food and drinks they consume every day in the food record questionnaire. To ensure subjects consume catfish *abon* according to the nutritional support needs of protein and record food and drink intake every day, the researchers did the monitoring by sending a reminder in the form of a Short Message Service reminder. Messages sent to the subject only contained a reminder to consume *abon* given in the study but did not contain material related to dietary therapy on hemodialysis. Nutrition monitoring using SMS is proven to improve dietary adherence of hemodialysis patients (9). If the patient adheres to the given diet, the nutritional status of the patient will increase and can indirectly increase the albumin levels (10,11). This is related to changes in behavior which is the most influential factor on adherence to the diet of hemodialysis patients (12).

The contents of the SMS were sent to subjects only to remind and ensure subjects to eat catfish *abon* and record their food and drink intake from sources other than *abon* regularly every day. The SMS does not contain material related to the hemodialysis diet so as not to cause bias related to nutritional counseling provided by the hospital nutritionist. After 21 days post-intervention, the subject's albumin level was measured again.

The dependent variable of this study was albumin levels, while the independent variable was the use of catfish as an animal protein source. Albumin level was on a nominal scale with two categories, i.e. hypoalbuminemia when the albumin levels were <3.50 g/dL and normal when the albumin levels were ≥3.50 g/dL. In this study, there are external variables that are thought to affect albumin levels, namely patient intake. Patient's intake was defined as the overall energy, protein, fat, and carbohydrate intake of hemodialysis patients obtained by the food record method, then processed using the Nutrisurvey program, compared to the patient's needs calculated individually, and presented. The calculation of its needs using the formula according to Pernefri, namely energy needs of 35 kcal/kg BW/day, protein requirements of 1.2 g/kg BB/day, fat 20% of total energy needs, and carbohydrates are the remaining total energy requirements.

The data obtained were then analyzed univariately to determine the frequency distribution of each variable and bivariate analysis using the Fisher's Exact test. This research has obtained Ethical Clearance from the Health Research Ethics Commission, Faculty of Health Science, Universitas Respati Yogyakarta No: 330.4/FIKES/PL/II/2017 dated 15 February 2017.

RESULTS

Hemodialysis patients routinely come 2 times a week with a fixed schedule, namely Monday–Thursday, Tuesday–Friday, and Wednesday–Saturday. The hemodialysis patient schedule is divided into 3 shifts, namely the morning shift (07.00–11.00 WIB), afternoon (11.00–15.00 WIB), and evening (16.00–20.00 WIB).

Subjects in this study were patients with a diagnosis of chronic kidney disease (CKD) with hemodialysis routinely 2 times a week. The number of subjects there were 34 people taken by purposive sampling technique from 3 shifts (morning, afternoon, evening) on Monday–Thursday, Tuesday–Friday, Wednesday–Saturday. Subject characteristics in this study include age, gender, education, and complication of the disease, which are shown in Table 2.

Table 2. Frequency distribution of subject characteristics

Subject characteristics	Category	Frequency (n)	Percentage (%)
Age	17 – 25 years	1	2,9
	26 – 35 years	4	11,7
	36 – 45 years	11	32,3
	46 – 55 years	9	26,4
	56 – 65 years	7	20,5
	≥ 65 years	2	5,8
Total		34	100
Gender	Man	15	44,1
	Woman	19	55,8
Total		34	100
Education	Primary school	13	38,2
	Junior high school	5	14,7
	Senior high school	14	41,1
	University	2	5,8
Total		34	100
Complication of disease	With complications	19	55,8
	No complications	15	44,1
Total		34	100

Based on Table 2, the majority of subjects belonging to the adult age category (11 people or 32.3%). Based on gender, there were more female subjects (19 people; 55.8%) than male subjects (15 people; 44.1%). Based on the level of education, the majority of subjects had a high school / vocational education (14 people or 41.1%) and at least were university graduates, namely 2 people or 5.8%. Based on complications, there were more subjects with disease complications (19 people; 55.8%) than subjects without disease complications (15 people; 44.1%). Complications of the subject's diseases include hypertension, gastrointestinal disorders (gastritis), gout, and lupus.

In this study, subjects were given nutritional support for catfish *abon* for 21 days, under the need of subjects that was 0.36 g/kg BB/day. The body weight used to calculate the needs of catfish *abon* is

the post hemodialysis body weight of each subject. Subjects consume all catfish *abon* given and controlled by sending a Short Message Service to the subject to remind the subject to consume the catfish *abon*. Even based on the results of the interview with the subjects, they liked the catfish *abon* and wanted to be given the catfish *abon* for a longer time after the study was completed. It means that the catfish *abon* product has good acceptance. Variable utilization of catfish as an animal protein source can be divided into pre-intervention and post-intervention.

Table 3 shows that in the pre-intervention condition, subjects with normal albumin levels were higher (73.53%) than subjects who experienced hypoalbumin (26.47%). Similarly, in the post-intervention condition, subjects with normal albumin levels were more (88.24%) than subjects who experienced hypoalbumin (11.76%). Table 3 shows that the number of subjects with normal albumin levels increased in the post-intervention condition compared to the pre-intervention.

Table 3. Frequency distribution of subject based on albumin levels

Data retrieval time	Category	Frequency (n)	Percentage (%)
Pre-intervention	Hypoalbumin	9	26.47
	Normal	25	73.53
Total		34	100
Post-intervention	Hypoalbumin	4	11.76
	Normal	30	88.24
Total		34	100

Total nutrient intake and non-catfish *abon* nutrient intake during the intervention in hemodialysis patients

The results of this study consider external variables that are thought to affect the results of the study, namely the patient's nutritional intake. The patient's nutritional intake is divided into the total nutrient intake during the intervention and the patient's nutritional intake from sources other than catfish *abon*.

The total nutrient intake during the intervention in this study included intake from catfish *abon* plus intake from non-catfish *abon*. Nutrient intake was obtained from the average intake of subjects for 21 days obtained by the Food Record method, processed using the Nutrisurvey program, then compared with the patient's needs calculated per individual. The intake is categorized to be low if the intake <80% needs, good if the intake 80–110% needs, and high if >110% needs.

Table 4. Frequency distribution of subject based on total nutrient intake during the intervention

Variable	Category	Frequency (n)	Percentage (%)
Energy intake	Low	13	38.2
	Good	18	52.9
	High	3	8.8
Total		34	100
Protein intake	Low	18	52.9
	Good	14	41.1
	High	2	5.8
Total		34	100
Fat intake	Low	21	61.7
	Good	11	32.3
	High	2	5.8
Total		34	100
Carbohydrate intake	Low	12	35.2
	Good	18	52.9
	High	4	17.6
Total		34	100

Table 4 shows that the majority of subjects consumed energy in the good category, as many as 18 people (52.9%). Most subjects consume protein in the less category, as many as 18 people (52.9%). Most of the subjects consume fat with fewer categories, namely as many as 21 people (61.7%). Most subjects consume carbohydrates in the good category, namely as many as 18 people (52.9%).

The intake of non-*abon* nutrients during the intervention was the intake of subjects sourced from food and beverages other than *abon* catfish. The intake of non-*abon* nutrients was obtained from the average intake of non-*abon* subjects for 21 days obtained by the Food Record method, processed using the Nutrisurvey program, then compared with the patient's needs calculated per individual. The intake of non-*abon* nutrients is categorized to be less if the intake <80% needs, both if the intake 80–110% needs, and more if >110% needs.

Table 5. Frequency distribution of subject based on nutrition intake from non-catfish *abon* source during the intervention

Variable	Category	Frequency (n)	Percentage (%)
Energy intake	Low	17	50
	Good	15	44.1
	High	2	5.8
Total		34	100
Protein intake	Low	28	82.3
	Good	5	14.7
	High	1	2.9
Total		34	100
Fat intake	Low	25	73.5
	Good	7	20.5
	High	2	5.8
Total		34	100
Carbohydrate intake	Low	12	35.2
	Good	18	52.9
	High	4	11.7
Total		34	100

Table 5 based on the intake of energy nutrients shows that the majority of subjects consume energy from non-catfish *abon* sources with fewer categories, as many as 17 people (50%). Based on the intake of protein nutrients from non-catfish *abon* sources the majority of subjects consumed protein in the less category, as many as 28 people (82.3%). Based on the intake of non-catfish *abon* fat nutrients, the majority of subjects consumed less fat, namely 25 people (73.5%). Based on the intake of non-*abon* carbohydrate nutrients, the majority of subjects who consumed carbohydrates in the good category, namely as many as 18 people (52.9%).

Effectiveness of catfish *abon* as nutritional support to albumin levels of hemodialysis patients

Fisher's Exact test results in Table 6 show that the use of catfish as an effective animal protein source has a significant effect on improving albumin levels of hemodialysis patients with p-value=0.048 (p-value<0.05).

Table 6. Bivariate analysis result of the effectiveness of catfish *abon* as nutritional support to albumin levels of hemodialysis patients using Fisher's Exact test

Category of albumin levels		Post-intervention		Total	*p-value*
		Hypoalbumin	Normal		
Pre-intervention	Hypoalbumin	3 (33.30%)	6 (66.70%)	9 (100.00%)	0.048
	Normal	1 (4.00%)	24 (96.00%)	25 (100.00%)	
Total		4 (11.80%)	30 (88.20%)	34 (100.00%)	

DISCUSSIONS

This study was dominated by adult subjects. This study was consistent with the general description of CKD patients undergoing hemodialysis in Indonesia, as reported by the Indonesian Renal Registry, which in 2011 found 89% of CKD patients undergoing hemodialysis aged 35–70 years with the most age groups of 45–54 years which are 27%.

Age is one of the factors that can affect an individual's health status. At the age of 40–70 years, glomerular filtration rate will progressively decrease to 50% from normal, i a decrease in the ability of the kidney tubules to reabsorb and concentrate urine, decrease the ability to empty the bladder thereby increasing the risk of infection and obstruction, and decreasing fluid intake which is a risk factor for kidney damage (13). Gender and age affect the incidence of glomerulonephritis which is one of the risk factors for chronic kidney failure (14).

The results of this study, more female subjects than male subjects. These results are similar to previous studies that the sex of patients with chronic kidney failure who performed hemodialysis were more women, as many as 33 people (52.4%), compared to 30 men (47.6%) (15).

Most of the subjects in this study had high school/vocational education and at least a university education graduate. In patients who have higher education will have broader knowledge also allows patients to be able to control themselves in overcoming the problems faced, have high self-confidence, experienced, and have precise estimates of how to handle events and easily understand what is happening recommended by health workers, will be able to reduce anxiety so that it can help the individual in making decisions (16). Knowledge of cognitive is a very important domain for the formation of action, behavior based on knowledge will be more lasting than those not based on knowledge (17).

Based on complications, the number of subjects with complications is higher than subjects without complications. Complications of the subject's diseases include hypertension, gastrointestinal disorders (gastritis), gout, and lupus.

Albumin levels of Hemodialysis Patients

In this study, there was an increase in the number of normal albumin subjects who experienced an increase in the condition of post-intervention compared to pre-intervention. Albumin is a very important serum in the body which is the main determinant of blood plasma osmotic pressure. The effect caused by decreased albumin will cause a shift in fluid from the intravascular space (18).

Hypoalbumin in patients with chronic kidney failure who have undergone hemodialysis can be caused by severe malnutrition caused by the inflammatory process in patients with chronic kidney failure. The presence of inflammation is associated with anorexia in dialysis patients. Chronic inflammation can also result in a rapid decrease in skeletal muscle protein and other tissues, reducing muscle and fat resulting in hypoalbumin (19). In addition to the inflammatory process of hemodialysis which removes protein and vitamins along with the dialysate which during hemodialysis runs it will lose 10–12 grams of amino acids, glucose will also be released via dialysate (20).

Albumin deficiency occurs when production is reduced and loss of albumin. More protein is lost than is made by a healthy liver (21). Decreased albumin levels in patients undergoing hemodialysis are influenced by malnutrition due to the inflammatory process that still occurs due to lack of hemodialysis time (22).

Total nutrient intake and non-catfish *abon* nutrient intake during the intervention in hemodialysis patients

Nutrient intake includes energy, protein, fat, and carbohydrates. The total nutrient intake during the intervention in this study included intake from catfish *abon* plus intake from non-catfish *abon*. The intake of non-catfish *abon* nutrients during the intervention was the intake of subjects sourced from food and beverages other than catfish *abon*.

Patients with chronic kidney failure with hemodialysis are recommended high protein intake to maintain nitrogen balance and replace amino acids lost during the hemodialysis process, ie 1–1.2 g/kg BW/day with 50% protein should be of high biological value because protein intake is very necessary remembering its function in the body. The effect of protein intake plays an important role in the prevention of nutrition of patients with chronic kidney failure because the symptoms of the uremic syndrome are caused by the accumulation of body protein catabolism. Therefore, the better the protein intake, the better it is in maintaining its nutritional status (23).

Cultivated fat intake 30% of calorie intake. On the one hand, fat intake is sufficient to meet calorie needs, while on the other hand fat also worsens kidney function and increases morbidity due to atherosclerosis. Consumption of complex carbohydrates such as rice, bread, sweet potatoes,

and cassava can spur the removal of excess uric acid in the blood (24).

Food intake of hemodialysis patients refers to the level of deterioration in kidney function. Food intake that must be limited in consumption is, protein intake is limited to 1–1.2 g/kg/day, potassium intake is limited to 40–70 meq/day, given the decreased function of potassium excretion and excretion of urea nitrogen by the kidneys. Meanwhile, the number of calories given is 30–35 kcal/kg BB/day (25).

Effectiveness of catfish *abon* as nutritional support to albumin levels of hemodialysis patients

In this study after being tested using Fisher's Exact, it was shown that providing nutritional support for catfish *abon* was effective in influencing the albumin levels of hemodialysis patients. Catfish *abon* has never been given as a form of dietary therapy in hemodialysis patients that is needed to replace the protein lost during hemodialysis and prevent catabolism of protein so that it can maintain serum creatinine levels within normal limits, overcome hypoalbuminemia, and enhance immunity.

Nutrient-based dietary guidelines emphasize animal-based protein foods for preventing and managing protein-energy wasting in hemodialysis patients (26). Fish is an animal food that contains proteins of good quality due to the complete content of essential amino acids. Fish can be extracted to obtain plasma protein (sarcoplasmic) containing albumin and another nutrient that has the potency to improve hypoalbuminemia condition (27).

There is a similar study with the basic ingredients of processed fish which are nutritional support in hemodialysis patients, the fish used is cork fish filtrate. Giving an extra intradialytic diet can maintain and increase serum albumin levels, as a result of protein intake during the hemodialysis process so that it can replace the amino acids lost especially during hemodialysis (25). The high intensity of use of cork fish (snakehead fish) to produce various products of fish albumin has put the natural stock of this fish species under great pressure. Therefore, it is necessary to find alternative sources of fish albumin other than the cork fish (28). Catfish is one of the sources of animal protein that has a high biological value, has a high protein content, and can be accessed by the public at a low price. The protein from catfish contains albumin, which can improve the albumin levels of hemodialysis patients if consumed with an adequate amount.

CONCLUSIONS

The use of catfish as an effective animal protein source has a significant effect on improving albumin levels in hemodialysis patients. Based on these conclusions, it is recommended to the hospital conduct continuous monitoring of the intake of hemodialysis patients, especially animal protein sources, to maintain the nutritional status of hemodialysis patients. Further research needs to be done on nutritional support for hemodialysis patients, with the use of local food in Indonesia.

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REFERENCES :

1. Ghorbani A, Hayati F, Karandish M, Sabzali S. The prevalence of malnutrition in hemodialysis patients. J Renal Inj Prev 2020;9(2):1–6.
2. Ministry of the Health Republic of Indonesia. Hasil utama riskesdas 2018. Jakarta: Ministry of the Health Republic of Indonesia; 2018.
3. Rivai AT. Status albumin serum pasien penyakit ginjal kronik yang menjalani hemodialisis di rumah sakit cipto mangunkusumo pada bulan februari 2009 dan hubungannya dengan lama hemodialisis [Artikel Penelitian]. Jakarta: Universitas Indonesia; 2009.
4. Wi JW, Kim N. Assessment of malnutrition of dialysis patients and comparison of nutritional parameters of CAPD and hemodialysis patients. Biomed Sci Letters 2017;23(3):185–193.
5. Qureshi AR, Alvestrand A, Divino-Filho JC, Gutierrez A, Heimburger O, Lindholm B, Bergstrom J. Inflammation, malnutrition, and cardiac disease as predictors of mortality in hemodialysis patients. JASN 2002;13:S28–S36.
6. Suhardjono. Buku ajar ilmu penyakit dalam jilid 2 edisi 3. Jakarta: Balai Penerbit FKUI; 2001.
7. Sridhar NR, Josyula S. Hypoalbuminemia in hemodialyzed end-stage renal disease patients: risk factors and relationships - a 2-year single-center study. BMC Nephrol 2013;14(242):1–9.
8. Kim Y, Molnar MZ, Rattanasompattikul M, Hatamizadeh P, Benner D, Kopple JD, Kovesdy CP, Kalantar-Zadeh K. Relative contributions of inflammation and inadequate protein intake to hypoalbuminemia in patients on maintenance

hemodialysis. Int Urol Nephrol 2013; 45:215–227.
9. Widiany FL, Afriani Y. Pemantauan gizi dengan SMS reminder efektif meningkatkan kepatuhan diet pasien hemodialisis. Jurnal Gizi Klinik Indonesia 2019;15(3):89–94.
10. Widiany FL, Afriani Y. Pemberian SMS reminder efektif memperbaiki status gizi antropometri pasien hemodialisis. Ilmu Gizi Indonesia 2017;01(01):49–53.
11. Widiany FL, Afriani Y, Rohman A. The effectiveness of nutritional monitoring by short message service reminder to albumin level of hemodialysis patients. J Nutr Sci & Diet 2018;4(3):Summer.
12. Widiany FL. Faktor-faktor yang mempengaruhi kepatuhan diet pasien hemodialisis. Jurnal Gizi Klinik Indonesia 2017;14(2):72–79.
13. Lajuck KS, Moeis ES, Wongkar MCP. Status gizi pada pasien penyakit ginjal kronik stadium 5 yang menjalani hemodialisis adekuat dan tidak adekuat. Jurnal e-Clinic 2016;4(2).
14. Price SA, Wilson LA. Patofisiologi konsep klinis proses-proses penyakit edisi 6 volume 2. Jakarta: EGC; 2006.
15. Damayanti P, Apen, Lisa A. Karakteristik penderita gagal ginjal kronis yang dilakukan hemodialisis; 2014.
16. Kamaluddin R, Rahayu E. Analisis faktor-faktor yang mempengaruhi kepatuhan asupan cairan pada pasien gagal ginjal kronik dengan hemodialisis di RSUD Prof. Dr. Margono Soekarjo Purwokerto. Jurnal Keperawatan Soedirman (The Soedirman Journal of Nursing) [series online] 2009;4(1). Available from: URL: http://jks.fikes.unsoed.ac.id/index.php/jks/article/view/218.
17. Notoatmojo S. Metodologi penelitian kesehatan. Jakarta: Rineka Cipta; 2002.
18. Sacher, Ronald A. Tinjauan klinik hasil pemeriksaan laboratorium. Jakarta: EGC; 2004.
19. Mardiana N. Nutrisi pada penderita dialisis. Prosiding dalam Meeting and Symposium [series online] 2008. Available from: URL: http://www.b11nk.wordpress.com
20. Yuwono A. Kualitas hidup menurut spitzer pada penderita gagal ginjal terminal yang menjalani hemodialisis di RSU DR. Kariadi Semarang. Artikel [series online] 2000. Available from: URL: http://www.eprint.undip.ac.id
21. Sibuea H, Panggabean M, Gultom. Ilmu penyakit dalam. Jakarta: PT Rineka Cipta; 2005.
22. Arinta, Rihantoro T. Hubungan lama menderita gagal ginjal kronik dengan kadar albumin pada pasien yang menjalani hemodialisis. Jurnal Keperawatan 2015;11(1).
23. Almatsier S. Penuntun diet. Jakarta: PT Gramedia Utama; 2005.
24. Sustrani L, Syamsir A, Iwan H. Asam urat, informasi lengkap untuk penderita dan keluarganya edisi 6. Jakarta: Gramedia; 2004.
25. Sulistyowati E, Hadi H, Susetyowati, Atma G. Pemberian diet ekstra formula komersial dan diet ekstra filtrat ikan gabus intradialisis serta pengaruhnya terhadap kadar serum albumin dan kreatinin pasien dengan hemodialisis di RSUD Dr. Saiful Anwar Malang. Jurnal Gizi Klinik Indonesia 2008;5(2).
26. St-Jules DE, Goldfarb DS, Popp CJ, Pompeii ML, Liebman SE. Managing protein-energy wasting in hemodialysis patients: A comparison of animal- and plant-based protein foods. Seminars in Dialysis 2018:32(1).
27. Mustafa A, Widodo MA, Kristianto Y. Albumin and zinc content of snakehead fish (Channa striata) extract and its role in health. IEESE International Journal of Science and Technology (IJSTE) 2012;1(2):1–8.
28. Fatma N, Nurilmala M, Metusalach, Nurfaidah, Astuti N. Low economic benefits of fish as source of fish albumin besides cork fish. Advances in Economics, Business and Management Research 2019;141:56–59.

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Effects of mixture powder of black rice (Oryza sativa L indica), red beans (Phaseolus vulgaris L), and moringa leaves (Moringa oleifera L) on blood glucose concentration in hyperglycemic Rats

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ABSTRACT

Background: Diabetes mellitus, increased blood glucose or hyperglycemia, is associated with increased oxidative stress and cardiovascular diseases. This condition will further cause carbohydrate and fat metabolism change, resulting in the decreased antioxidant defense system. Black rice, red beans, and moringa leaves contain oleic acid, butyric amino acid, antioxidants, phytic acid, and arginine, which can improve insulin sensitivity, and blood glucose homeostasis.
Objective: This study aimed to analyze the effect of betamelor (black rice, red beans, and moringa leaves) on blood glucose in rats
Materials and Methods: The design of this research was experimental research with pre-and post-control group design. A total of 20 Sprague Dawley female rats were divided into four groups, namely standard feed (PS), 80% feed of betamelor (PB8), 50% feed of betamelor (PB5), and 20% feed of betamelor (PB2). Betamelor intervention was given as much as 5% of weight for 28 days. Fasting Blood Glucose (FBG) levels were measured using the GOD-PAP method. Blood glucose data were analyzed by Analysis of Variance (ANOVA) at a 95% confidence level and using Duncan's test.
Results: There were differences in FBG between groups after the intervention of betamelor. The results showed that after 28 days of intervention, betamelor decreased the serum glucose concentration from 122.69 mg / dL to 97.70 mg / dL (20.37%) in the PB8 group and from 123.91 mg / dL to 113.28 mg / dL (8.58%) in the PB5 group, but the standard diet (PS) increased by 5.73%. This result can be applied to reduce blood glucose levels in obese and patients with metabolic syndrome.
Conclusions: There was a significant effect of giving a mixture of black rice, red beans, and Moringa leaves on fasting blood sugar in rats.

Keywords: Black rice; Red beans; Moringa leaves; Fasting blood glucose

BACKGROUND

The changes in lifestyle and diet of carbohydrate-based foods into high-fat content foods leads to the rise of degenerative diseases such as coronary heart disease, hypertension, and diabetes mellitus. An increase in blood glucose or hyperglycemia is a sign of a metabolic disorder known as diabetes mellitus and is associated with the rise of oxidative stress and complications in the vascular system. It will further cause the alteration in carbohydrate and fat metabolism resulting in an impairment of the oxidation defense system. In which this condition stimulates the increased formation of Reactive Oxygen Species (ROS) induce β-pancreatic cell dysfunction.1

One of the antioxidant defense systems is Superoxide Dismutase (SOD), the enzyme involved in the earliest and most potent detoxification in cells.2 The antioxidant enzyme itself is an essential enzyme which is able to eliminate radicals, then may protect cells against toxic of aerobic metabolism byproducts.3 The use of oral medicine of diabetes mellitus accompanied by the use of natural ingredients already become common practice yet, and around 1050 anti-Diabetes Mellitus plants have been studied.4 Some plants commonly used as a source of functional foods and already developed to help in controlling blood sugar are black rice, red beans, and moringa leaves. Ingredients contain oleic acid, amino butyric acid, antioxidants, pitic acid and arginine which are proven to reduce the rate of oxidative stress, thereby increasing antioxidants in the body.

Following Walter and Marchesan (2011), phenolic compounds are concentrated higher in the black rice pericarp.5 Bioactive compounds work as antioxidants in rice, including phenolic, flavonoid,

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anthocyanin, proanthocyanidin, tocopherol, tocotrienol, γ- oryzanol and phytate.6 Aleuron and endospermic in black rice could produce the high-intensity anthocyanin result a deep purple to dark color.7 However, black rice contains less protein and fiber, and this lack can be corrected by substitution of red beans and Moringa leaves. Kidney beans contain fat (15.80%), dietary fiber (3.60%), and carbohydrates (49.00%).8 The soluble fiber in red beans may reduce blood glucose.9 These beans also possess a high antioxidant capacity. The antioxidant capacity of EC50 red bean skin extract reaches 294.78 mg/ml.10 Al-Malki et al. (2015) found that antioxidant and antidiabetic activity in Moringa leaf extract showed potential as an antidiabetic agent in streptozotocin induced mice.17 Tantipaiboonwong et al. (2017), who have reported the antihyperglycemic and anti-hyperlipidemic effects of red rice extract and black rice extract to streptozotocin induced mice. Kidney beans contain arginine about 600 mg/100g.25 Arginine acts as an antidiabetic by regenerating pancreatic β cells to improve the stimulation of insulin secretion.26 Moringa leaves contain 6.7% protein, 1.7% fat and 14.3% carbohydrates.

Rat is one of rodents often used as models in studying blood sugar levels. This is because rats have a significant similarity in hematology and genome with humans.11 Studies related to black rice, red beans and Moringa leaves, which associated with the potential and influence on health, are still rarely performed. It is critical to assess whether there is an impact of giving black rice, red beans, and moringa leaves on blood sugar levels in rats.

This study aims to determine the effect of feeding developed from black rice flour, red beans, and Moringa leaves on rat blood glucose. Precisely, the objectives of this study, i.e., 1) determine differences in body weight 2) determine the average consumption of rat feeding during the intervention period; 3) analyze the effect of treatment on reducing feed consumption; 4) investigate the impact of treatment on rat blood glucose.

MATERIALS AND METHODS

This was an experimental study using a complete random design (CRD) with a pre-post control group. This study was conducted in March-May 2020, involving several laboratories. Making the black rice, red beans, and moringa leaves flours were carried out in the Food Processing Laboratory of Kupang's Health Polytechnic of Health Ministry's. The feed production was carried out at the Animal Feed Industry Laboratory, Kupang Agricultural Polytechnic. The intervention towards the experimental animal and blood collection was executed at the Biosciences Laboratory of the University of Nusa Cendana Kupang, while blood analysis conducted at the Health Laboratory of East Nusa Tenggara. The proximate analysis was carried out at the Saraswanti Indo Genetech Laboratory in Bogor, West Java.

The number of experimental animals used in this study was 20 rats, which were calculated according to the Federer formula (1997): (n-1) (t-1) ≥15 (n is the number of samples needed and t is the number of treatments). All those 20 rodents were divided into four groups, namely five rats of the standart feed (PS), five rats intervened by 80% mixture powder (PB8), five rats intervened by 50% mixture powder (PB5), and five rats intervened by 20% mixture powder (PB2). The animals used were Sprague Dawley female rats met the inclusion criteria, such as normal and healthy, aged 5-6 months, the weight of 150-200 g, open eyes, reddish-white skin, agile, and have soft, clean, dense, smooth, not fall, and shiny-haired. The exclusion criteria were that the rat died during the intervention, behavior changes (did not want to eat, drink and limp), weight loss of > 5%. Equipment used for rat care consists of a cage filled with husks equipped with food containers and drinking bottles, iron cage enclosures, digital scales for weighing rats and leftovers, and a set of cage cleaning tools. This study had received ethical approval from the Animal Ethics Commission, Faculty of Veterinary Medicine, University of Nusa Cendana No.168 / UN15.8.1 / PP / 2020.

The first stage of the research was the feed formulations from black rice, red beans, and moringa leaves. The composition of ingredients was following the diet of people with diabetes mellitus. Producing mixture powder began with making black rice flour, red beans flour, and moringa leaves powder, then sieved with 80 mesh size. Feed material in the form of bravo 512 standard solid feed was mixed until homogeneous. After that, a little water added until the feed mixture was half wet, then formed to obtain pellet feed. The wet pellet then dried in an oven (T 40ºC), then put into a plastic bag and kept in a refrigerator until the intervention time to experimental rats.

During the adaptation period, intended to ensure the experimental animals were in a healthy condition before the intervention, which was carried out in individual cages, given standard feed, and ad libitum drinking water for about ten days. Bravo 512 standard feed contains 12% water, 19.5-21.5% crude protein, min 5% crude fat, max 5% crude fiber, max 7% ash, 0.9 - 1.1% calcium, 0.6 – 0.9% phosphorus, and energy

about 3125kcal/Kg. Intake and residual feed were weighed and recorded every day, and body weighing was done once a week. A high glucose feed of 2cc/200g of body weight/day was given through sonde for two weeks to create hyperglycemic rats. The treatment was performed for 28 days, i.e., subjected a standard feed, 80% mixture powder, 50% mixture powder, and 20% mixture powder. Furthermore, those rodents fasted for 10 - 12 hours, then two cc of blood samples per each were taken through the eye veins (ocular arteries) using a microhematocrit capillary tubes after anesthetized with ketamine 10 mg/kg rat. Blood serum glucose analysis was executed according to the GOD-PAP method. The data of blood glucose were analyzed by Analysis of Variance (ANOVA) at a 95% confidence level. If it shows the significance response, then Duncan's Multiple Range Test was applied. Data processing and analysis were performed using Microsoft Excel and SPSS for Windows version 22.0.

RESULTS AND DISCUSSION

Proximate Composition

The results of the proximate analysis of black rice, red beans, and moringa leaves can be seen in Table 1. It shows that the ingredients used complement nutritional value each other mainly on the parameters of carbohydrate, protein, fat and dietary fibre.

Table 1. Proximate Composition of Mixture Powder

Parameters	PB Mixture 80%	PB Mixture 50%	PB Mixture 20%
Protein (g)	6.3 ± 0.91	10.1 ± 1.26	13.87 ± 2.03
Total fats (g)	0.8 ± 0.17	1.54 ± 0.49	2.28 ± 0.94
Carbohydrate (g)	25.3 ± 3.21	36.5 ± 2.64	47.9 ± 5.16
Fibre (g)	18.6 ± 2.33	13.8 ± 1.75	9.1 ± 1.02
Energy (kcal)	133.5 ± 18.01	200.2 ± 20.01	267.6 ± 37.22

Body Weight

Body weighing was done weekly to determine the amount of standard feed, high glucose feed (weeks 1 and 2), and mixture powder feed (weeks 3 to 6). The effect of feeding treatment on the body weight of rats can be seen in table 2. At the end of acclimatization, the average body weight of the rats did not have a significant difference between treatment groups. During the high-fructose diet for 7 days, the body weight of the rats increased significantly compared to the acclimatization period, but not significantly between treatments. During the mixture powder intervention period for 21 days, the body weight of the rats experienced a significant increase compared to the high fructose diet and there were also significant differences between the treatment groups. It was probably caused by the influence of blood drawn through the eye veins (ocular arteries), which results in pain and low appetite, recognized from the decrease in feed intake. In the following days, there was a slow improvement in body weight in line with an increase in feed intake.

Table 2. Body weight of rats fed a high-fat diet and treated with mixture powder for 10 weeks

Group	Acclimatization mean ± SD (g)	HGD mean ± SD (g)	Intervention of mixture powder mean ± SD (g)	P[2] (Acclimatization)	P[1] HGD1-Intervention
PS	190.82 ± 8.81	202.33 ± 5.79	227.62 ± 4.95	0.048	0.000
PB8	192.27 ± 6.47	201.48 ± 7.40	210.13 ± 3.19		
PB5	190.38 ± 6.72	202.18 ± 4.51	219.68 ± 3.57		
PB2	189.91 ± 7.63	203.76 ± 6.24	224.27 ± 5.51		
P[1]	0.821	0.294	0.008		

HGD : High Glucose Diet, [1]One way Anova test, [2] Paired t-test

Table 3. Concentration (mg/dL) of fasting blood glucose of female Sprague dawley rats that were administered graded doses of mixture powder

Group	Acclimatization mean ± SD (mg/dl)	HGD mean ± SD (mg/dl)	P[2]	△Increase (mg/dl) (%)	P[1]
PS	78.41 ± 2.60	124.10 ± 3.71	0.000	45.69 ± 1.51 (58.27%)	0.071
PB8	80.12 ± 1.53	122.69 ± 2.06	0.000	42.57 ± 2.07 (53.13%)	
PB5	79.64 ± 1.29	123.91 ± 3.49	0.000	44.27 ± 1.92 (55.58%)	
PB2	78.29 ± 2.31	124.53 ± 3.84	0.000	46.24 ± 1.89 (59.06%)	
P[1]	0.369	0.493			

HGD : High Glucose Diet, [1] One way Anova test, [2] Paired t-test

Table 4. Concentration (mg/dL) of fasting blood glucose of female Sprague dawley rats that were administered graded doses of betamelor.

Group	Acclimatization mean ± SD (mg/dl)	HGD mean ± SD (mg/dl)	P[2]	△Increase (mg/dl) (%)	P[1]
PS	124,10 ± 3,71	131,21 ± 2,46	0,004	7,11 ± 2,18 (5,73%)	0,007
PB8	122,69 ± 2,06	97,70 ± 1,92	0,000	-24,99 ± 1,84 (-20,37%)	
PB5	123,91 ± 3,49	113,28 ± 1,17	0,000	-10,63 ± 2,35 (-8,58%)	
PB2	124,53 ± 3,84	126,96 ± 2,38	0,042	2,43 ± 1,72 (1,95%)	
P[1]	0,493	0,493			

HGD: High Glucose Diet, [1] One-way Anova test, [2] Paired t-test

Fasting Blood Glucose Level Before and After Observation

Blood glucose levels of the rats at the end of the aclimatization stage and after HGD feeding for each group can be seen in Table 3. Significantly increased blood sugar levels occurred in all groups after intervened by HGD with a significant value (p> 0.05) based on the analysis of one-way test Anova. The average value of fasting blood glucose levels in the intervention of mixture powder 80% (PB8) and mixture powder 50% (PB5) experienced a significant decrease (p <0.05). While the response by standard feed as control and mixture powder 20% (PB2) experienced an increase in blood glucose levels. There were differences in rat blood glucose levels between groups after the administration of mixture powder based on the *Kruskal Wallis test*. The PB8 treatment group experienced the most substantial decline in blood glucose levels which was -24.99 ± 1.84 (-20.37%) mg/dl (Table 4). Concentration (mg/dL) of fasting blood glucose of female Sprague dawley rats that were pre and post administered graded doses of *mixture powder*.

Body weight in mice increased significantly for all groups from the acclimatization period to the high fructose diet phase and the high fructose diet phase to the betamelor intervention phase.Weight loss in the experimental animal was significantly increased for all groups on the acclimatization, high glucose diet, and mixture powder intervention. There was no significant difference in body weight among the groups in either phase of acclimatization or a diet high in glucose, but there were substantial differences in the intervention period. The bodyweight improvement was due to the consumption of standard and high glucose feed before the intervention. The differences in the different treatments during the intervention cause the alterations that have a significant effect on the amount of energy intake so that excess consumption will be stored as fat

reserves and increase bodyweight. In the acclimatization phase, the standard feed intake of each group was not significantly different. In the phase of the provision of a high glucose diet, there were significant differences between the treatment groups and the standard feed, the feeding intake of the control group had the highest compared to other groups.

Significant differences in feed intake occurred between groups. The control group had a higher feed intake than the treatment group. The consumption of the control group tended to remain since acclimatization. The treatment group of 50% and 20% mixture powder increased but not significant, and the 80% mixture powder group experienced a decrease in feed intake. It was due to the administration of mixture powder in the treatment groups that contains a high fibre composition, lead to full quickly compared to the standard feed group. Reducing intake in the intervention phase compared to the conditioning phase of treatment groups was due to the high fibre effect of diet caused by the rise of the hormone leptin, so the appetite decreased in the intervention phase.12 In the treatment groups, the standard feed intake was lower than in the conditioning period because it was in the adaptation stage. During the intervention phase, the usual feed intake of treatment groups was lower than the other groups because the stomach was quickly filled due to high fibre feeding.

Blood glucose levels were elevated in the test animals as the result of conditioning the provision of a high glucose diet after a period of adaptation completed with values > 120 mg/dl. It can be interpreted as metabolic syndrome, according to Crescenzo (2014) in Italia, the metabolic syndrome can be realized by high glucose feeding to increase blood glucose levels.13 Excess fructose intake in the liver will be metabolized into fat. Fructose may lead to a failure of insulin signal to reduce the synthesis of glycogen and increase gluconeogenesis, resulting in blood glucose elevation.13

Furthermore, this study revealed that the average fasting blood glucose values in the PB8 and PB5 groups experienced a significant slope after mixture powder intervention ($p < 0.05$). The results of this study were in accordance with Mbikay's literature review study (2012), the administration of moringa leaves for diabetes mellitus causes a potentially reducing blood sugar levels.14 Compounds that was an essential key allegedly in the antihyperglycemic activity in extracts of moringa (M. oleifera) is oleic acid. Oleic acid, or commonly called omega-9, is one of the best types of fat. Oleic acid also is known as one of monounsaturated oil (MUFA) because the body can synthesize from the nutritional compounds consumed, so it also including in non-essential fatty acids.14

In the research conducted by Aly et al. (2016) using GC-MS analysis, identified eight fatty acids contained in Moringa seed oil including palmitic acid, stearic acid, arachidonic acid, behenic acid, palmitoleic acid, linoleic acid, linolenic acid, and oleic acid.15 Besides, there were more than 50 phytochemical contents possessed in Moringa seed oil. According to Busari et al. (2015), the hypoglycemic activity of Moringa seed oil is caused by the presence of fatty acids in the oil.16 Monounsaturated fatty acids have a tendency to improve the function of beta-cell secretors, reduce the disruption of beta-cell activity and proliferation. Al-Malki et al. (2015) found that antioxidant and antidiabetic activity in Moringa leaf extract showed potential as an antidiabetic agent in streptozotocin-induced mice.17

Blood glucose levels in this study also found a decrease might be caused by the presence of black rice extract in the mixture flour. It is in line with the study by Tantipaiboonwong et al. (2017), who have reported the antihyperglycemic and anti-hyperlipidemic effects of red rice extract and black rice extract to streptozotocin-induced mice. The results of the study revealed that consumption of black rice extracts of 50 mg/kg body weight or red rice extract of 100 mg/kg body weight significantly reduced blood sugar levels after eight weeks.18 Observation by Chaiyasut et al. (2017) showed that germinated black rice extract could increase the aminobutyric acid content, total antioxidant capacity, and levels of antioxidant enzymes in diabetic rats and showed antidiabetic activity both before and after administration of streptozotocin in rats as animal models.19

A study conducted by Chung, et al. (2016) also found that pigmented rice has inhibitory enzymes responsible for diabetes. The results showed that those pigments contained phenol and had a significant enzyme inhibitory activity. Bioactive compounds that cause pigmentation in rice are anthocyanin and proanthocyanidin.20 Kasim et al. (2005) revealed that black rice has the highest antioxidant activity compared to the red and white rice. Naturally, black rice contains pigments with high antioxidant activity (anthocyanin).21 Hosoda, et al (2018) reported that black rice and brown rice contain anthocyanin and proanthocyanidin, which are potentially used as antioxidants sources other than as a starch source in ruminants.22 Bioactive compounds in pigmented rice

may reduce the oxidative stress, prevent cancer, cardiovascular, complications and potential diabetes, and others.5,22

In this study, mixture powder as a mixture of flours in an intervention feed in which one constituent is red beans, so the slope of fasting blood sugar values in an animal model may be related to the red beans. Kidney beans (*Vigna angularis*) are good sources of fiber, every 100 grams of dried red beans provides about 4 grams of fiber consisting of both soluble and insoluble fiber.23 Soluble fiber significantly lowering blood sugar, due to the soluble fiber is able to substantially reduce the glycemic response 23,24. This beans like other legumes, contain several components of inhibitor substances such as phytic acid, tannin, trypsin inhibitors, and oligosaccharides. The diet treatment using this legume may reduce glucose absorption by 48.43%. As for the soy diet, it can reduce absorption by 45.84% .25 Water-soluble fiber can form a viscous solution; the higher viscosity in the intestine leads the slower absorption of glucose by the small intestine. The viscous soluble fiber can reduce postprandial blood sugar and insulin levels. Based on the results of several studies, three different doses of red beans might lower blood glucose levels in male Wistar rats that given glucose load.24

Kidney beans contain arginine about 600 mg/100g.25 Arginine acts as an antidiabetic by regenerating pancreatic β cells to improve the stimulation of insulin secretion.26 Arginine as an amino acid of hormonal terminal GLP-1 (Glucagon-Like-Peptide-1) served as proinsulin gene expression and insulin synthesis stimulator. After insulin secretion occurs, glucose levels in the circulation immediately decrease; thus, the effect of GLP-1 will disappear by itself.27 Kidney beans; a staple food recommended for diabetic patients contains high protein and fiber, and low carbohydrates. Starch granules from these legumes have a unique structure and hydrolysis of starch in the small intestine relatively slow, thus postprandial glycemic response will be delayed.27 Yao's (2014) finding that the mechanism of red beans in hyperglycemia related to the effect of protein and phenolic compounds in it. Besides, this experiment also proved that extruded red bean protein and polysaccharides play a role in α-glucosidase inhibition.28 The subjects used were mice with *Sparague Dawley* strains, could not study the mechanism of diabetes mellitus. in further research, humans can use as samples. Further research is necessary to test the complete phytochemical contents of mixture powder of black rice, red beans, moringa leaves and to analyze its effect on other anti-inflammatory and antioxidant parameters in diabetes mellitus to deepen our understanding on the role and mechanisms of mixture powder of black rice, red beans, moringa leaves in diabetes mellitus

CONCLUSIONS

According to this study, the mixture of black rice, red beans, and moringa leaves as mixture flour reduce the fasting blood glucose reduction in rats significantly. Further research is needed to examine the complete phytochemical content of mixed black rice, kidney bean, moringa leaf powder and analyze their effect on anti-inflammatory and other antioxidant parameters in diabetes mellitus to deepen understanding of the role and mechanism of mixed powder black rice, kidney beans, moringa leaves in diabetes mellitus.

REFERENCES

1. Retnaningsih C, Darmono, Widianarko B, Muis SF. Peningkatan aktivitas antioksidan superoksida diamutase pada tikus hiperglikemi dengan asupan tempe koro benguk (Mucuna pruriens L).Agritech. 2013: 33(2):154–161.
2. Ighodaro OM, Akinloye OA. First line defence antioxidants-superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPX): Their fundamental role in the entire antioxidant defence grid. Alexandria Med J. 2017;54(4):287–293
3. Mansuroglu B, Derman S, Yaba A, Kizilbey K. Protective effect of chemically modified SOD on lipid peroxidation and antioxidant status in diabetic rats. Int J Biol Macromol. 2014;72:79–87
4. Subramoniam, A. Anti-Diabetes Mellitus Plants. Boca Raton: CRC Press; 2016
5. Walter M, Marchesan E.Phenolic Compounds and Antioxidant Activity of Rice. *Brazilian A Bio Tech*. 2011; Vol. 54 (2): 371–377.
6. Goufo P, Trindade H. Rice Antioxidants: Phenolic Acids, Flavonoids, Anthocyanins, Proanthocyanidins, Tocopherols, Tocotrienols, γ-Oryzanol, and Phytic Acid. *F Sci Nutr*. 2013; Vol. 2 (2): 75–104.
7. Wanti S, Andriani M.A.M, Parnanto N.H.R. Pengaruh Berbagai Jenis Beras terhadap Aktivitas Antioksidan pada Angkak oleh *Monascus purpureus. Biofarmasi. 2015;*Vol. 13 (1): 1–5
8. Audu SS, Aremu MO. Effect of process- ing on chemical composition of red kidney bean

(*Phaseolus vulgalris* L.) flour. PJN. 2011;10(11):1069-1075.
9. Messina. Nutrition and health benefit of dried bean. Am J Clin Nutr. 2014; 100 (suppl): 437S-442S.
10. Fidriannyi I, Nuraini P, Marlia SW. Antioxidant activities, total flavonoid, phenolic, carotenoid of various shells extracts from four species of legumes. Asian J Pharm Clin Res. 2014; 7(4):42-46.
11. Dieman VV, Trindale EN, Trindale MRM. Experimental model to induce obesity in rats. Acta Cir Bras. 2006;21(6):425-429.
12. Izadi, V., Saraf-Bank, S., & Azadbakht, L. Dietary intakes and leptin concentrations. *ARYA atherosclerosis.*2014;*10*(5), 266–272.
13. Crescenzo R, Bianco F, Coppola P, Mazzoli A, Tussellino M, Carotenuto R, et al. Fructose supplementation worsens the deleterious effects of short-term high-fat feeding on hepatic steatosis and lipid metabolism in adult rats. Exp Physiol. 2014;99(9):1203-13
14. Mbikay, Majambu. Therapeutic Potential of *Moringa oleifera* Leaves in Chronic Hyperglycemia and Dyslipidemia: A Review. Front Pharmacol. 2012; 3:24
15. Al-Malki, A.L, dan El Rabey, H.A. The Antidiabetic Effect of Low Doses of Moringa oleifera lam. Seeds on Streptozotocin Induced Diabetes and Diabetic Nephropathy in Male Rats. Biomed research International. 2015: 1-13.
16. Tantipaiboonwonga P, Pinthaa K, Chaiwangyena W, Chewonarinb T, Pangjitc K, Chumphukama O,et.al. Anti hyperglycaemic and Anti hyperlipidaemic Effects of Black and Red Rice in Streptozotocin Induced Diabetic Rats. *ScienceAsia.* 2017;Vol. 43: 281– 288.
17. Chaiyasut C, Sivamaruthi B.S, Pengkumsri N, Keapai W, Kesika P, Saelee M, Tojing P, et.al. Germinated Thai Black Rice Extract Protects Experimental Diabetic Rats from Oxidative Stress and Other Diabetes-Related Consequences. *Pharmaceuticals*. 2017; Vol. 10 (3): 2–16.
18. Chung S.I, Kang M.Y, Lee S.C. Inhibitory Effect of Germinated Pigmented Rice on Key Metabolic Enzymes Associated with Diabetes and Hyperglycemia. *In JF Nutr Scie.* 2016; Vol. 5 (4): 32–40.
19. Kasim E, Astuti S, Nurhidayat N. Karakterisasi Pigmen dan Kadar Lovastatin Beberapa Isolat *Monascus purpureus*. *Biodiversitas*. 2005;6 (4): 245–247.
20. Hosoda K, Sasahara H, Matsushita K, Tamura Y, Miyaji M, Matsuyama H. Anthocyanin and proanthocyanidin contents, antioxidant activity, and *in situ* degradability of black and red rice grains. Asian-Australas J An Scie. 2018; Vol. 31 (8): 1213–1220.
21. Rimbawan.Indeks glikemik pangan: cara mudah memilih pangan yang menyehatkan. Jakarta: Penebar Swadaya;2004.
22. Audu SS, and MO Aremu. Effect of Processing on Chemical Composition of Red Kidney Bean (Phaseolus vulgaris L .) Flour. Pakistan J Nutr. 2011;10 (11):1069–75.
23. Monti LD, MCC asiraghi, E Setola, E Galluccio, MA Pagani, L Quaglia, et al,. L-Arginin Enriched Biscuit Improve Endothelial Function and Glucose Metabolism: A Pilot Study in Healthy Subjects and A Cross-over Study in Subjects with Impaired Glucose Tolerance and Metabolic Syndrome. Met Clin Ex. 2013;62: 255-264.
24. Yao Y, Cheng X, Ren G. Alpha-Glucosidase inhibitory activity of protein-rich extracts from extruded adzuki bean in diabetic KK-Ay mice. Food Func. 2014;5(5):966–971.
25. Liu, Y., Wang, Q., Li, S., Yue, Y., Ma, Y., & Ren, G. Convenient food made of extruded adzuki bean attenuates inflammation and improves glycemic control in patients with type 2 diabetes: a randomized controlled trial. Ther clin risk man.2018;14, 871–884
26. Yao Y, Cheng X, Wang L, Wang S, Ren G. A determination of potential alpha-glucosidase inhibitors from Azuki Beans (Vigna angularis) Int J Molecular Sci. 2011;12(10):6445–6451.

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The effectiveness of Islamic comic media in increasing the attitude of healthy breakfast among students

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ABSTRACT

Background: As much as 73.4% of children aged ≥ 5 years in Indonesia have breakfast with the low nutritional quality of food consumption, as evidenced by the prevalence of 95.5% of children's less consumption of vegetables and fruits. Breakfast habits affect fiber and micronutrient levels. Communication and nutritional information can increase accuracy in breakfast behavior from childhood. Effective and efficient media are needed. So far, the media for children has prioritized illustrations without paying attention to the cultivation of moral values.
Objectives: This study aimed to determine the effect of Islamic comic media on improving healthy breakfast attitudes among students.
Materials and Methods: The research design was a quasi-experimental type. Subjects were taken by a simple random sampling method. Ninety-six students were divided into three groups. Group A was given nutrition education using Islamic comic media about health breakfast (n = 32). Group B was given nutrition education using Islamic comic strip media about health (n = 32). Group C was a control group (n = 32). The study used two types of nutrition promotion media: comics and comic strips, which are stories about healthy eating in Islam. The time for nutrition education intervention through comics and comic strips is 2x ± 35 minutes. The pretest was ± 25 minutes with ± 10 minutes explaining the instructions for filling out the questionnaire. The posttest was ± 25 minutes with a discussion for ± 10 minutes—nutritional attitudes related to health breakfast by answering a validated questionnaire. We used the Lickert scale to categorized nutritional attitudes. Statistical test was done by Wilcoxon and Mann Whitney test.
Results: The distribution of nutrition attitudes of respondents increased after the provision of Islamic comics, both comics and comic strips about breakfast. The nutritional attitudes increased significantly in the Islamic comic media group (p = 0.000) and the Islamic comic strip media group (p = 0.000), from 78.75 to 92.96 and 78.61 to 92.88, respectively.
Conclusions: There was a relationship between counseling and Islamic comic media regarding healthy breakfast towards the nutritional attitude of elementary school-age students.

Keywords: Nutritional attitude; Islamic comics; Breakfast

BACKGROUND

In developing countries, the prevalence of obesity in children is known to have increased by 60% since 1980.[1] The habit of not having breakfast causes the child to experience hypoglycemia, dizziness, trembling, fatigue and difficulty concentrating.[2] Breakfast also affects appetite control, satiety, and energy expenditure, and weight management.[3] Long-term impact, resulting in nutritional status, decreasing health and stamina of children, and hindering the improvement of the nation's human resources In Indonesia. It is known that 69.6% of Indonesian children have not had breakfast according to the recommended balanced nutrition guidelines and 73.4% have breakfast with the nutritional quality of food consumption is low. The consumption of various foods greatly affects nutritional status.[4]

Breakfast has a positive influence on cognitive and academic outcomes.[5] Breakfast is important for elementary school age were a period of high brain productivity in absorbing various lessons.[6] The habit of not having breakfast also affects fiber and micronutrient levels. Children who regularly eat a healthy breakfast with vegetables and fruit have higher levels of fiber and micronutrients.[7] Low consumption of fruits and vegetables can result from a lack of motivation.[8]

Habits and appropriateness of children's breakfast can be formed with nutritional education.[9] Nutritional education with a medium can accelerate understanding, resulting in motivation according to the message received.[10] Therefore, an educational process needs to be supported by appropriate media.[11] Elementary school-aged students are more interested

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in media and picture books or cartoons than media and books that contain only writing.[12] Cartoon media has been shown to increase children's fruit or vegetable intake.[13]

Media is very important in the educational process. This is because a medium can increase motivation and enthusiasm for learning psychologically.[14] Someone who has good nutritional knowledge, then dominantly has a good nutritional attitude too. Student learning intentions can be shaped by using learning comics. Meanwhile, in comic media, besides being preferred by students, it is also proven to be effective in increasing students' knowledge. The effectiveness of comics as a learning medium was proven positive with a significant increase in the level of knowledge in the experimental group.[15] A significant increase also occurred in the attitude of the experimental group. Students' interest in learning media in the form of audiovisuals illustrates good acceptance.[16] Educational media for children nowadays put forward good illustrations without paying attention to the cultivation of moral values.[17] The learning media should not only prioritize changes in knowledge but also contain moral values.[18] Cultivation of noble morals through comic book characters also plays an important role, because children like to imitate and look after. [19] This study aims to analyze the influence of Islamic comic media on healthy breakfast on elementary school students' attitudes.

MATERIALS AND METHODS

This study used a quasi-experimental design. Location of research at Public Madrasah Ibtidaiyah (MIN) 1 Teluk Lingga, East Kutai district, East Kalimantan, Indonesia. This study was carried out in December 2018 - February 2019, which was adjusted to the active schedule of students of MIN 1 Teluk Lingga, Sangata City, East Kutai district, East Kalimantan, Indonesia. The school criteria taken were Islamic elementary (Madrasah Ibtidaiyah / SDIT), had A accreditation by the official madrasah accreditation organizing body, and had never received counseling or nutritional education. MIN 1 Teluk Lingga is a school that meets the above criteria with the largest student population.

The population in this study were all students of MIN 1 Teluk Lingga, as many as 647 students. Consists of 21 classes with various levels. The inclusion criteria for the selection of research subjects were subjects who had never received nutritional education about healthy breakfast, were willing to take part in the research until it was finished, and were aged 10-12 years. Sampling calculation using the Slovin formula to obtain a sample size of 96 students, then formed 3 groups, namely, group A given nutritional education with Islamic comic media about healthy breakfast (n = 32), group B was given nutritional education using Islamic comic strip media about healthy breakfast and group C was a control group (n = 32).

The variables studied included elementary school students' attitudes regarding healthy breakfast before and after the comic education intervention using a questionnaire that had been tested for validation. The questionnaire totaled 16 attitude statements. Lickert used to determine the level of a person's attitude by providing "agree" and "disagree" statements.[20] Attitude data was categorized as good if the answer is 76% -100% correct, categorized sufficient if the answer is correct 51% -76%, and categorized as less if the answer is correct 25-50%.[21]

This study used two types of nutrition promotion media, in the form of comics and comic strips, which are stories about healthy breakfast according to Islamic eating habits. The main characters used are elementary school-age children with additional characters in the form of mothers and teachers at school. Characters are designed with noble morals with a good speech to provide good role models for respondents.

The types of images used in both media were 2D animation based on Graphical User Interfaces (GUIs). The tools used in the media are the Samsung Galaxy Tab A.8.0 Tablet and the rubber nib Stylus Pen with applications such as Medibang Pro and Adobe Photoshop. The comic was later printed on A4 ivory paper.

This research was conducted in five stages which were carried out for five consecutive days. The first stage was pretest data collection on the three groups carried out on the first day. The second stage was carried out with nutrition education intervention in group 1 using comic strip media and in group 2 using comic strip media which was conducted on the second day. The third stage was posttest data collection from the three groups with the same questions in the three groups which were carried out on the third day. The fourth stage was the repetition of the second stage, while the fifth stage was the repetition of the third stage.

The time for nutrition education intervention through comics and comic strips was 2x ± 35 minutes. The pretest was carried out for ± 25 minutes with ± 10 minutes of explaining the instructions for filling out the questionnaire. Posttest was carried out for ± 25 minutes with a discussion for ± 10 minutes.

The analysis used the Wilcoxon test with Mann-Whitney post hoc because the data was not normal. The Wilcoxon test aims to determine whether there is a significant difference between the attitude values before and after the provision of comic strip media, comic strips, and without giving media to each treatment group. Furthermore, the Mann-Whitney post hoc test was conducted to determine which intervention or treatment was the most effective. This study was endorsed by the health research ethics committee of Dr. Moewardi Hospital, Faculty of Medicine, Sebelas Maret University with number 395 / III / HREC / 2019

Figure 1. Comic cover

The nutrition promotion media created were Islamic comics in the form of comics and comic strips, with the title "Sarapan Sehat, yuk!" (Let's have healthy breakfast!), The first edition. The place of manufacture was in Sangata on December 30th, 2018. The total page consists of 9 pages for comics. The language used is Indonesian. The story setting is Home and School. The main characters of this comic are Ali and Aisyah, while the additional characters are Budi, Umi, and Bu Guru. Summary of story: Every morning, Umi always told Ali and Aisyah to have breakfast, but they didn't know why they had to have breakfast every morning. One day at school, their friend Budi fainted during a ceremony because he wasn't having breakfast. After being explained by the teacher, Ali and Aisyah knew the benefits of breakfast. Since then, Ali and Aisyah always had breakfast happily and also helped Umi cook breakfast. Comic bonus: the 'Kata Nabi' comic about the prohibition of denouncing food and blowing hot food. Reference comics: Comic "Ayo Sarapan Sehat" (Come have a healthy breakfast), by PERGIZI, and comic "Sarapan, yuk!" (Let's have breakfast) by the POMPI club (BPOM).

Figure 2. Islamic Comic Characters and Storylines

The main characters used are elementary school-age children with additional characters in the form of mothers and teachers at school. Parents are the first and foremost educators, whose roles are role models and provide the best care and education for children. Characters are designed with noble morals with a good speech to provide good role models for respondents.

On the first day, the pretest questions were distributed to the research group at different lesson hours. The pretest time lasted for 35 minutes, the first 10 minutes, the researcher used to explain the aims and objectives of the study by filling out the questionnaire instructions. The time to answer the pretest questions was 25 minutes. The nutrition promotion media provision intervention was given the next day.

Nutrition promotion media were given in 2 days with 2 hours of subjects per day. The time used was 70 minutes. On the first day, students were asked to listen to several students who led the story (storytelling) in front of the class and matched the conversation with the comic that each student had held. This was done to add an audio function that makes it easier to remember during the presentation of the material. Because comics are printed media that do not have an audio function. Then, the researcher asked three students to retell the essence of the story they understood by bringing their respective comics.

The researcher asked each student to read their comics without being guided by anyone at the second meeting. This is to train the critical feeling that students have in understanding the implicit message in the story presented. The researcher gave 30 minutes to read the comic and asked the students to read it twice to strengthen students' understanding of the story content. At the end of the meeting, the researcher asked three students to give their opinion about the message conveyed by comics and provide the essence of the story, this time without bringing comic media.

RESULTS

Based on the data analysis carried out, the distribution of respondents age and gender characteristics
was described in the following table:

Table 1. Distribution of Respondents Age and Gender Characteristics

Variable	A%	B%	C%	Total%
Age				
10	65.7	34.3	53.1	51
11	34.3	65.7	46.9	49
Gender				
Male	50	50	43.7	48
Female	50	50	56.3	52

Note: A = The group that was given nutrition education using Islamic comic media about healthy breakfast (n = 32); B = The group that was given nutrition education using Islamic comic strip media about healthy breakfast (n = 32); C = Control group (n = 32).

Differences in Attitude Values Between Research Groups

The data was not normally distributed related to the results of the distribution on the normality test of the mean data difference between the pretest and posttest attitude values. Furthermore, the analysis was carried out with the Wilcoxon test with the Mann Whitney post hoc test, to determine which groups had differences. The results showed that between groups A and B the result was p = 0.000 (<0.05).

Table 2. Attitudes of Respondents Before and After Intervention

	Pengukuran	n	Mean	Median (Minimum-Maximum)	p Value
A	Before the Intervention	32	78.75	90.62 (50.00-100.00)	0.000
	After the Intervention	32	92.96	95.31 (76.56-100.00)	
B	Before the Intervention	32	78,61	88.81 (50.00-100.00)	0.000
	After the Intervention	32	92,88	95.31 (68.75-100.00)	
C	Before the Intervention	32	80,46	85.15 (48.43-98.43)	0.148
	After the Intervention	32	82,46	84.37 (50.00-100.00)	

DISCUSSION

Based on the results obtained, it can be seen that there were differences in attitude scores in groups A and B compared to group C which is the control group. This result was in line with previous research conducted by Widayanti et al. which states that there was a significant increase in attitude scores (<0.05) in elementary school students who were given nutrition promotion media in the form of comics.[22] This result was also supported by previous research conducted by Jefri and Kharis (2017) in which there was a significant increase in attitude levels (> 0.05) in elementary school students who were given promotional media in the form of comic strips.[6] In line with the research of Ridha et al. (2009) stated that comics have a strong influence on social information processing to create changes in attitudes.[23]

The main role of comics in instruction is their ability to create interest in students. The effectiveness of comics as a learning medium was proven positive with an increase in the level of knowledge that significantly shaped attitudes in the experimental group.[15] Students' interest in learning media in the form of audiovisuals illustrates good acceptance.[16] Comic media and comic strips have several main functions as follows: 1) Attention function, which attracts and directs students' attention to concentrate on the content of the lesson related to the visual meaning displayed; 2) The compensatory function, visual media that provides context for understanding the text, helps students who are weak in reading to organize information in the text and recall and understand the content of the lesson that is presented with text or presented verbally;[24] 3) Affective function, where visual images or symbols can evoke emotions and attitudes of students, for example, information relating to social or racial issues;[25] 4) Cognitive function, where visual symbols or images facilitate the achievement of goals to understand and remember the information contained in images.[26]

Based on the results of the mean difference between the pre and post-education attitude values in group A was 14.21 with a pretest value of 78.75 increasing to 92.96 and the mean difference between the pre and post-education attitude values in group B was 14.27 with a value of 78.61 increasing to 92.88. The results of the Mann Whitney test in groups A and B were p = 0.000. It can be concluded that there was a significant increase in attitudes after the provision of nutrition promotion media in the form of Islamic comic strips and comic strips in groups A and B. In group C, the difference in the mean value of attitude increased by 2.0 but based on the Wilcoxon test there was no increase in attitude in group C, which is the control group.

Changes in the attitude value of the experimental group that increased significantly also reflected an increase in the learning achievement of respondents. Motivation to learn is the point the respondent's result is obtained by each individual as a level of ability expressed in the form of cumulative values or numbers. The importance of increasing the respondent's nutritional attitude after giving the media is an indication that the moral values of the characters conveyed in Islamic comics have succeeded in giving a significant effect in increasing the respondent's attitude.[27] This is based on the nature of the respondents who are children of primary school age who like to imitate and nurture.[27] Factors that influence attitude formation include the amount of information a person has.[28] The more attractive the educational media used, the higher the increase in knowledge that shapes the respondent's attitude.[29]

CONCLUSIONS

The respondent's attitude increased after counseling by providing Islamic comic media about healthy breakfast. There was an effect of the provision of Islamic comic media and Islamic comic strips regarding healthy breakfast on increasing attitudes.

REFERENCES

1. Gupta N, Goel K, Shah P, Misra A. "Childhood obesity in developing countries: epidemiology, determinants, and prevention," *Endocr. Rev.*, vol. 33, no. 1, pp. 48–70, 2012, DOI: 10.1210/er.2010-0028.
2. Huang CJ, Hu HT, Fan YC, Liao YM, Tsai PS. "Associations of breakfast skipping with obesity and health-related quality of life: Evidence from a national survey in Taiwan," *Int. J. Obes.*, vol. 34, no. 4, pp. 720–725, 2010, doi: 10.1038/ijo.2009.285.
3. Gwin JA, Leidy HJ. "A review of the evidence surrounding the effects of breakfast consumption on mechanisms of weight management," *Adv. Nutr.*, vol. 9, no. 6, pp. 717–725, 2018, DOI: 10.1093/advances/nmy047.
4. Perdana F, Hardinsyah d. "Analisis Jenis, Jumlah, Dan Mutu Gizi Konsumsi Sarapan Anak Indonesia (Analysis of Type, Amount, and Nutritional Quality of Breakfast among Indonesian Children)," *J. Gizi Dan Pangan*,

vol. 8, no. 1, pp. 39–46, 2013, [Online]. Available: http://download.portalgaruda.org/article.php?article=127426&val=199&title=ANALISIS JENIS, JUMLAH, DAN MUTU GIZI KONSUMSI SARAPAN ANAK INDONESIA.

5. Yaslam Babaeer L, Wraith D. "A systematic review of the effect of habitual breakfast for A systematic review of the effect of habitual breakfast for adolescents aged 11-19 years on academic performance adolescents aged 11-19 years on academic performance," *J. Adolesc. Fam. Heal.*, vol. 9, no. 1, p. 7, 2018, [Online]. Available: https://scholar.utc.edu/jafh/vol9/iss1/7/.
6. Jefri M, Kharis W. "Pengaruh Pendidikan Kesehatan Menggunakan Media Komik Strip Terhadap Pengetahuan," *J. Penelit. Keperawatan*, vol. 3, no. 2, pp. 96–99, 2017.
7. Coulthard JD, Palla L, Pot GK. "Breakfast consumption and nutrient intakes in 4-18-year-olds: UK National Diet and Nutrition Survey Rolling Programme (2008-2012)," *Br. J. Nutr.*, vol. 118, no. 4, pp. 280–290, 2017, doi: 10.1017/S0007114517001714.
8. Albani V, Butler L, Traill W, Kennedy O. "Breakfast consumption and nutrient intakes in 4–18-year-olds: UK National Diet and Nutrition Survey Rolling Program," *Br. J. Nutr*, vol. 120, no. 2, pp. 398–408, 2017.
9. Damayanti AY, Setyorini IY, Fathimah F. "Gambaran Tingkat Pengetahuan Gizi Seimbang pada Santriwati Remaja Putri di Pondok Pesantren," *Darussalam Nutr. J.*, vol. 2, no. 2, p. 1, 2018, doi: 10.21111/dnj.v2i2.2437.
10. Cita DA. "Pengembangan Materi dan Daya Terima Pendidikan Gizi Melalui Media Visual Terhadap Pengetahuan Gizi Anak Usia Sekolah Dasar," 2014.
11. Kementrian Kesehatan RI. "Pedoman PGSKesehatan," *Pedoman Gizi Seimbang*, pp. 1–99, 2014.
12. Daryanto. *Manajeman Pemasaran : Sari Kuliah*. Bandung: Satu Nusa, 2013.
13. Kraak VI, Story, M. "Influence of food companies' brand mascots and entertainment companies' cartoon media characters on children's diet and health: A systematic review and research needs," *Obes. Rev.*, vol. 16, no. 2, pp. 107–126, 2015, DOI: 10.1111/obr.12237.
14. Rusman. *Belajar dan Pembelajaran Berbasis Komputer*, 1st ed. Bandung: Alfabeta, 2013.
15. Pritandhari M, Ratnawuri T. "Penerapan Komik Strip Sebagai Media Pembelajaran Mata Kuliah Manajemen Keuangan Mahasiswa Univeristas Muhammadiyah," *J. Pendidik*, vol. 3, no. 1, pp. 1–13, 2015.
16. Johan H, Pradana TD. "Pengaruh Penyuluhan Media Audio Visual Video Terhadap Perilaku Cuci Tangan Pakai Sabun Pada Siswa Kelas III Di SDN 027 Samarinda," *J. Husada Mahakam*, vol. 4, no. 6, pp. 352–356, 2018.
17. Yuliana L. *Urgensi Penanaman Nilai-Nilai Moral Pada Anak Usia Dini*. Bandung: Alfabeta, 2014.
18. Najuah. "Strategi Pembelajaran Berbasis Pendidikan Nilai Dalam Pembelajaran IPS Di Sekolah," in *Prosiding : Seminar Nasional Tahunan*, 2017.
19. Al-Ayouby MH. "Dampak Penggunaan Gadget Pada Anak Usia Dini (Studi di PAUD dan TK.Handayani Bandar Lampung)," *Skripsi*, vol. 1, no. 1, pp. 1–10, 2017, doi: 10.1037/0022-3514.51.6.1173.
20. Swarjana K. *Metode Penelitian Kesehatan*, 1st ed. Yogyakarta: ANDI, 2015.
21. Arikunto S. *Prosedur Penelitian Suatu Pendekatan Praktik*, Revisi 7. Jakarta: Arikunto, 2013.
22. Widajanti L, Suryawati C, Sugihantono A. "The Effect of Healthy and Nutritious Snack Food Comic to Increase Knowledge and Attitudes of Elementary School Children Ages, Indones.," *J. Public Heal.*, vol. 6, no. 1, pp. 22–27, 2009.
23. Ridha A, Dian P, Mayarestya NP. "Pengaruh media komik terhadap pengetahuan kesehatan mata pada anak," *J. Perikan (J. Fish. Sci.)*, vol. XII, no. 1, pp. 20–27, 2009.
24. Ajizah N, Suherman HA, Susilawati D. "Penggunaan Media Pembelajaran Untuk Meningkatkan Gerak Dasar Lari Sprint Pada Siswa Kelas V Sdn Sukajadi Kecamatan Situraja Kabupaten Sumedang," *Sp Vol 1*, vol. 1, no. 1, pp. 35–44, 2016.
25. Doron E. "Fostering Creativity in School-

Aged Children Trought Perspective Taking and Visual Media Based Short Term Intervention Program," *J. Think. Ski. Creat.*, vol. 23, no. 27, pp. 150–160, 2016, DOI: https://doi.org/10.1016/j.tsc.2016.12.003.

26. Dean J. "Sorted for Memes and Gifs: Visual Media and Everyday Digital Politics.," *Polit. Stud. Rev.*, vol. 17, no. 3, pp. 255–266, 2019, DOI: 10.1177/1478929918807483.
27. Naujah. "Strategi Pembelajaran Berbasis Pendidikan Nilai Dalam Pembelajaran IPS Di Sekolah," in *in Seminar Nasional Tahunan*, 2017, p. 1.
28. Azwar S. *Sikap Manusia : Teori dan Pengukurannya*, 2nd ed. Yogyakarta: Liberty, 2016.
29. Hartono NP, Wilujeng CS, Andarini S. "Nutritional Education of Healthy Snack Choice Between Lecture and Comic Method," Indonesia," *J. Hum. Nutr*, vol. 2, no. 2, pp. 76–84, 2015.

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Application of the general message of balanced nutrition during the pandemic coronavirus disease 2019 (COVID-19) in Mataram City

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ABSTRACT

Background: Coronavirus Disease 2019 (COVID-19) in 2020 became the limelight worldwide, including in Indonesia. Infection caused by this virus can be prevented by social distancing, physical distancing, and a healthy lifestyle. Maintaining nutritional intake by implementing balanced nutrition guidelines is very important to enhance the immune system.
Objectives: This study aimed to determine the description of the application of balanced nutrition guidelines during the COVID-19 pandemic in the community in Mataram City.
Materials and Methods: This research was a descriptive study with a survey approach, and the data taken were cross-sectional. This research was conducted in May-June 2020 in the community in the Mataram City taken by purposive sampling with inclusion criteria, namely women and men of productive age, domiciled in the city of Mataram, married status, and at least elementary school education. Respondents in this study were 460 respondents who were calculated using the Slovin formula. Data on the application of balanced nutrition guidelines were obtained using a questionnaire. Data from the results of this research were analyzed using descriptive methods.
Results: There are as many as >90% of the community has implemented a general message of balanced nutrition in addition to the fifth message. As many as 89.9% of respondents did not apply the fifth general message of balanced nutrition in limiting consumption of sweet, salty, and fatty foods. The majority of applications >50% of general messages of balanced nutrition were applied by respondents aged 21-29 years, female, high education and work status.
Conclusions: The application of the general message of balanced nutrition during the epidemic COVID-19 in Mataram City was implemented optimally, but this application was not carried out in the fifth message in limiting consumption of sweet, salty, and fatty foods.

Keywords: COVID-19; Application; General Message for Balanced Nutrition

BACKGROUND

In early 2020, various parts of the world including Indonesia were shocked by the pandemic caused by the Coronavirus Disease 2019 (COVID-19). Coronavirus disease 2019 (COVID-19) is a disease caused by the acute respiratory syndrome coronavirus (SARS-CoV-2). On March 11, 2020, the World Health Organization (WHO) declared the Coronavirus outbreak in 2019 to be a pandemic [1]. This is because this disease has rocked more than 200 countries in various parts of the world including Indonesia. In Indonesia, on June 18, 2020, there were 42.762 cases infected with this virus with a cure rate of 39,3% [2], while in West Nusa Tenggara Province there were 1.022 cases infected by this virus. This happened because the spread of Coronavirus Diseases 2019 was relatively fast and difficult to detect [3]. Various efforts have been recommended to cut the spread of the virus through social distancing and physical distancing so that all activities such as work, discussions, and meetings are normally carried out online [4].

The restrictions that occur during the pandemic have an impact on the decline in the economy, food availability, nutritional intake, and people's lifestyle. Efforts and strategies continue to be made by the government to stabilize this condition, either by providing direct cash assistance or providing food supplies [1]. Availability of foodstuffs, a healthy

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lifestyle, and fulfillment of balanced nutritional intake are now things that need to be prioritized and carried out optimally. However, as before known, the problem of unhealthy food consumption habits among Indonesians including West Nusa Tenggara Province before the COVID-19 pandemic is still quite high and has not been resolved optimally. This showed in the results of the 2018 Basic Health Research in West Nusa Tenggara Province which noted that as many as 7.1% of the people did not consume fruits and vegetables, 31.2% had less activity, 53.1% did not wash their hands properly, > 30% consuming sweet and fatty foods and 10% with the habit of consuming salty foods [5]. The application of general guidelines for well-balanced nutrition during the COVID-19 pandemic plays an important role in the health of the body. Fulfilling the need for micronutrients, micro (micronutrients), food safety, physical activity and adequate water consumption in the body can help the hormones forming, leukocytes, immunoglobulins, and lymphocytes in the immune system improving the function of the heart, lungs and maintaining balance in the body during the COVID-19 pandemic [6].

According to the Indonesian Ministry of Health (2020), the infection caused by the Coronavirus in 2019 has an impact on increasing body temperature, so that compliance of balanced and healthy nutritional intake tends to increase to support the immune system. The adoption of unhealthy eating habits and lifestyles by not washing hands properly, unbalanced food consumption, and inadequate physical activity during the pandemic will have an impact on decreasing the body's defense system and increasing viral infections. Therefore, understanding and implementing a healthy lifestyle that refers to the guidelines for well-balanced nutrition is the main asset for obtaining a healthy, strong body defense and avoiding various diseases caused by viruses during the COVID-19 pandemic [7].

Previous research has shown that the majority of toddler mothers in Babakan District do not apply the four pillars of balanced nutrition obtained by using the interview method [8]. However, further, in this study, research will be carried out about the description of the application of general guidance for balanced nutrition in more detail on the application of 10 general messages of balanced nutrition, especially during the COVID-19 pandemic. The application of the general message of balanced nutrition is obtained by using a questionnaire in the form of scoring on each general message of balanced nutrition. The goal of this research is that people notice and apply a healthy lifestyle based on balanced nutrition guidelines in their daily life. In addition, it is also hoped that health workers and the government will continue to give education and socialization about general guidance for balanced nutrition to improve the health status of the community. This study aims to describe the application of balanced nutrition guidelines during the COVID-19 pandemic in the people of Mataram City, West Nusa Tenggara.

MATERIALS AND METHODS

This research was a descriptive study using a survey approach method. This research was conducted from May to June 2020 in Mataram City, West Nusa Tenggara. The populations in this study were all people in West Nusa Tenggara Province with the number of respondents calculated using the Slovin formula (α = 5%) as many as 460 respondents. The sampling technique in this study was carried out by using purposive sampling with inclusion criteria consisting of women and men of productive age, live in Mataram city, married and or have been married and at least primary school education. Data on the application of balanced nutrition general messages were collected using a questionnaire for the application of 10 general messages of balanced nutrition which were applied during the COVID-19 pandemic with the help of google form media. The questionnaire used in this study refers to a balanced nutrition behavior questionnaire with a Cronbach α reliability value of 0.725 and has been valid [9]. The general message of balanced nutrition contained in this study consists of (1) being grateful and enjoying the diversity of food, (2) consuming lots of vegetables at least 3 times a day and consuming fruit at least 2 times a day, (3) get used to consuming side dishes that contain high protein, (4) get used to consuming a variety of staple foods, (5) limit consumption of sweet, salty and fatty foods, (6) get used to breakfast, (7) get used to drinking enough and safe water in the amount of 8 glasses or ± 2 liters per day, (8) getting used to reading labels on food packaging, (9) washing hands with soap and running water and, (10) doing adequate physical activity and maintaining a normal body

weight with a duration of activity and exercise as much as 30 minutes per day. The data obtained from the results of this study were analyzed descriptively on each general message in the guidelines for balanced nutrition.

RESULTS

Respondent Characteristics

Data on the characteristics of respondents in this study consisted of age, gender, education, and occupation. The results of this research show that most of the respondents in this research were in the age range 21-29 years and were female. In terms of education, most of the respondents are at the middle level with a working status. The data on the characteristics of respondents in this research can be seen in Table 1.

Application of The General Message of Balanced Nutrition

Data collection on the application of balanced nutrition general messages in this research refers to 10 general messages of balanced nutrition in the balanced nutrition guidelines regulated by the Ministry of Health of the Republic of Indonesia in 2014. The application data for balanced nutrition general messages are categorized in the category of applied and not applied. The applied category is given to respondents who apply according to the instructions on every general message of balanced nutrition, while in the not applied category it is given to respondents who do not apply according to the instructions in every general message of balanced nutrition. The results of application 10 general messages on balanced nutrition in the guidelines for balanced nutrition can be seen in Table 2.

Table 1. Respondent Characteristics

Category	n	%
Age		
≤ 20 years	75	16,3
21 - 29 years	238	51,7
30 – 49 years	142	30,9
> 49 years	5	1,1
Sex		
Male	155	25,0
Female	345	75,0
Education		
Primary	7	1,6
Middle	80	17,4
Higher	373	81,0
Work Status		
Work	316	68,7
Not Work	144	31,3

Based on the results of the analysis in Table 2 it is known that during the COVID-19 pandemic 449 (97.6%) respondents had applied the general message of balanced nutrition in being grateful and enjoying a variety of foods. The application of getting into the habit of consuming vegetables and fruit also shows that most of the 454 (98.7%) respondents have adopted the habit of consuming vegetables and fruit every day. This happened because most respondents said that during the COVID-19 pandemic respondents gained a lot of knowledge from various information sources that advocated a healthy lifestyle with enough consumption of fruits and vegetables to maintain endurance and avoid viruses and various diseases, besides that the habit of consuming fruits and vegetables has also become a habit that has been done before, but during this pandemic, the consumption of fruits and vegetables has increased.

The general message of balanced nutrition on the habit of consuming side dishes that contain high protein and the habit of consuming a variety of staple foods has mostly been applied as many as 456 (99.1%) and 454 (98.7%) respondents have applied this general message. This is because most respondents said that the availability of high protein and staple foods such as chicken, sea fish, rice, noodles, and bread in West

Nusa Tenggara Province is quite available and easy to find in traditional markets and other shopping centers so that access to these foods is easy to get. Respondents did not apply restrictions on consuming sweet, salty, and fatty foods during the COVID-19 pandemic. As many as 98.9% of respondents are accustomed to consuming sweet, salty, and fatty foods, so the general message of balanced nutrition in getting used to limiting consumption of sweet, salty, and fatty foods is not applied. Most of the reason respondents did not apply this message was that it had become a habit. Sweet foods that are often consumed include syrup, flavored drinks, jam, candy, and the use of sugar, salty foods can be salted fish and salted eggs, while fatty foods consumed are fried and fast food.

Table 2. Application of 10 General Messages on Balanced Nutrition in The Guidelines for Balanced Nutrition

General Messages on Balanced Nutrition	Application				Total	
	Applied		Not Applied			
	n	%	n	%	n	%
1. Being grateful and enjoying the diversity of food	449	97,6	11	2,4	460	100
2. Consuming vegetables and fruit	454	98,7	6	1,3	460	100
3. Consuming side dishes that contain high protein	456	99,1	4	0,9	460	100
4. Get used to the consumption of a variety of staple foods	454	98,7	6	1,3	460	100
5. Limit consumption of sweet, salty and fatty foods	5	1,0	455	98,9	460	100
6. Get used to breakfast	450	97,8	10	2,2	460	100
7. Get used to drinking enough and safe water	446	96,9	14	3,1	460	100
8. Get used to reading labels on food packaging	445	96,7	15	3,3	460	100
9. Get used to washing hands with soap and running water	458	99,6	2	0,4	460	100
10. Doing enough activity and maintaining a normal body weight	421	91,3	39	8,6	460	100

The habit of drinking enough and safe drinking water of ± 8 glasses (2 liters) during the COVID-19 pandemic has been implemented by 446 (96.9%) respondents in this study, this habit has also been practiced before the COVID-19 pandemic. In addition to the habit of drinking enough and safe water, breakfast habits have also been carried out during this pandemic as many as 450 (97.8%) of respondents have adopted the habit of having breakfast. This happens because most respondents said that the habit of eating breakfast is done because to maintain a healthy lifestyle and this breakfast habit accidentally occurs because all activities and work are done at home so that the opportunity to have breakfast can be done.

Judging from the application of the general message of balanced nutrition in the habit of reading labels on food packaging during the COVID-19 pandemic, it is known that 445 (96.7%) respondents applied the general message, but a small proportion did not apply it, while in the application it was used to wash their hands using soap, and running water during the COVID-19 pandemic, it is known that most of the 458 (99.6%) respondents applied this general message. In the application of general messages in getting used to doing adequate physical activity and maintaining normal body weight, there were 39 (8.6%) respondents who did not apply this general message during the pandemic. This is because many respondents have the reason that office work and online assignments and meetings make physical activity limited.

Data on the application of the general message of balanced nutrition were also analyzed descriptive based on the characteristics of the respondents consisting of age, gender, education, and occupation. The application of this balanced nutrition general message is categorized in the category of applying

message, apply 50% of messages, and didn't apply the message. The category of applying messages is given to respondents if they have applied >50% - 100% or 6-10 messages of balanced nutrition according to the advice, while in the category of applying 50% of the messages are given to respondents if they have applied 50% of messages or 5 of 10 general nutritional messages balanced according to recommendations and the category of not applying message is given to the respondent if they do not apply the 10 general messages of balanced nutrition according to the advice. The results of descriptive analysis of the application of the general message of balanced nutrition based on the characteristics of the respondent during the COVID-19 pandemic can be seen in Table 3.

Based on the results of the analysis in Table 3, it shows that the majority of respondents who applied the general message of balanced nutrition during the COVID-19 pandemic were in the age range of 21-29 years as many as 235 (98.7%) respondents and as many as 6 (8.0%) respondents in ≤ 20 years old who apply 50% of the general message of balanced nutrition. Judging by gender, as many as 341 (9.8%) female respondents applied the general message of fully balanced nutrition, and seen at the education level, most of the higher education levels implemented the general message of fully balanced nutrition as many 365 (97.8%). The application of the general message of balanced nutrition during the COVID-19 pandemic has also been fully implemented by most respondents with a working status of 140 (97.3%) of respondents.

DISCUSSION

The infection caused by the coronavirus disease 2019 (COVID-19) causes many life changes, including lifestyle, so that the nutritional balance before and after infection is very important to pay attention to [7]. The implementation of the general guidance for balanced nutrition established by the Ministry of Health of the Republic of Indonesia in 2014 is one way that can be done to improve body health. These guidelines aim to offer behavioral guidelines in consuming food and beverages and living a healthy life. The results of these studies indicate that > 90% of respondents have implemented the general message of balanced nutrition properly during the COVID-19 pandemic except for the fifth message by limiting sweet, salty, and fatty foods. In general, the application of general guidelines for well-balanced nutrition during the COVID-19 pandemic can offer benefits for the health of the body.

Table 3. Application of Balanced Nutrition General Message based on Respondent Characteristics

Characteristics	Application						Total	
	Applying Messages		Applying 50% of Messages		Didn't applying Message			
	n	%	n	%	n	%	n	%
Age								
≤ 20 years	69	92.0	6	8,0	0	0	75	100
21 - 29 years	235	98.7	3	1.3	0	0	238	100
30 – 49 years	139	98.6	3	1.4	0	0	141	100
> 49 years	5	100	0	0	0	0	5	100
Sex								
Male	107	93.0	8	7.0	0	0	115	100
Female	341	98.8	4	1.2	0	0	345	100
Education								
Primary	7	100	0	0	0	0	7	100
Middle	76	95.0	4	5.0	0	0	80	100
Higher	365	97.8	8	2.2	0	0	373	100
Job Status								
Work	308	97.5	8	2.5	0	0	316	100
Not Work	140	97.3	4	2.7	0	0	144	100

The habit of consuming a variety of foods, both staple foods, and fruits and vegetables, getting used to breakfast, and reading packaging labels on food during the COVID-19 pandemic, the need for macronutrients, micronutrients, and food safety in the body are met. In addition, the fulfillment of protein intake, physical activity, and adequate water consumption in the body can effectively help hormones forming, leukocytes, immunoglobulins, and lymphocytes which can support the immune system, improve the function of the heart, lungs and maintain balance in the body during the COVID-19 pandemic [10].

Prevention of deficiency and excess due to nutrition caused by bad food habits and lifestyle during a pandemic requires an understanding and application of good nutrition with the principle of balanced nutrition [11,12]. In general, actions in the application of food consumption and healthy living carried out by each person will pass through various stages including response, mechanism, and adaptation [13]. The difference that occurs in each action is consuming everyone's foods is strongly influenced by the presence of internal factors and external factors. Psychological conditions, availability of foodstuffs, and the area of residence are risk factors for changes in eating behavior for everyone [14]. This is in line with the application of the general message of balanced nutrition carried out during the COVID-19 pandemic in this study, the psychological state and the availability of food ingredients during this pandemic period have made people pay more attention to a healthy lifestyle that is carried out daily so that body health is maintained. In addition, the government has made an appeal and education to wash hands with soap and clean water as well as improve the clean and healthy lifestyle of the community. However, the application of limiting the consumption of sweet, salty, and fatty foods has not been implemented optimally with >90% of respondents in this study still consuming large amounts, so this is a special concern that needs to be done to maintain a healthier intake.

The limitation made on the consumption of sweet, salty, and fatty foods in the general message of balanced nutrition is an effort to support health. Excessive consumption of sweet, salty, and fatty foods will have an impact on the emergence of various degenerative diseases such as diabetes mellitus, heart disease, and hypertension as well as changes in nutritional status so that with these diseases the body's defenses will become weak in the face of the virus during the COVID-19 pandemic [15,16,17]. The results of this study show that 98.9% of respondents do not practice limiting their consumption of sweet, salty, and fatty foods. This is in line with the results of the 2018 Basic Health Research which showed that consumption of sweet, salty, and fatty foods respectively was 37.6%, 10.0%, and 32.4%, which are still quite high in West Nusa Tenggara [6]. The emergence of various types of food and beverages on some televisions, shopping centers, habits, preferences, and environmental factors makes it easy to access these foods, thus affecting food consumption. In addition to food restrictions, the application of the habit of washing hands using soap and running water during the COVID-19 pandemic is also highly recommended. Washing hands using soap and running water is one of the efforts that can be done in reducing the spread of the coronavirus, so this application is very important.

In general, food consumption behavior and habits that are implemented by a person are strongly influenced by various factors, including internal and external factors. Internal factors consisting of age, gender, education, and occupation can affect a person's healthy behavior. The age factor can affect the knowledge a person has [18]. With increasing age, it greatly affects the development, needs, and mindset that exists in him, so that this situation affects the habit of implementing the general message of balanced nutrition [19]. The results of these studies indicate that as many as 98.7% of respondents aged 21-29 and gender apply the general message of well-balanced nutrition. One of the perceptions and patterns of thought that exist in each person is greatly influenced by the increase in age. Everyone will play an active role in social life and prepare for the adjustment to old age that occurs at the age of 20 to 35 years. Throughout that age, everyone will tend to spend more time acquiring knowledge, so that intellectual abilities, problem-solving, and knowledge acquisition tend to increase and result in good behavior in everyday life [20].

The educational factor is one of the tools used to produce a change in a person, therefore through education, a person can find out everything that has not been known before. Education is a process carried out to gain knowledge, understanding, and how to behave according to your needs [21]. The results of this

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study show that as many as 365 (97.8%) respondents with a higher education level have implemented >50% of the general message of well-balanced nutrition. This research is in line with research on mothers with children in Manado who reported that mothers who are at the higher education level have knowledge of well-balanced nutrition and will continue to learn and apply this knowledge related to nutrition to their children and their families [22]. In addition, the results of research in South Kalimantan also show that providing education related to good nutrition to a person will improve healthy behavior [23].

An internal characteristic that factors into a person's behavior is work. The work that is owned by each person influences lifestyle and consumption behavior, including behavior in implementing general guidance for balanced nutrition [15]. The results of this study show that as many as 308 (97.5%) respondents with working status have implemented> 50% general messages of well-balanced nutrition. This research is in line with research at SMK Surabaya which reports that the work factor of parents is one of the factors that can influence food consumption behavior in the household so that the habit of providing and implementing food for each individual and family is very much determined by education, work and income [24].

CONCLUSIONS

Based on the results of this research it was concluded that >90% of the people in Mataram City applied the general message of balanced nutrition in the first to fourth messages and the sixth to tenth messages properly during the pandemic period. However, >90% of people in Mataram City do not apply the fifth general message of balanced nutrition, namely limiting the consumption of sweet, salty, and fatty foods. Judging by the characteristics, most of the people aged 21-29 years, female sex, middle education, and work applying >50% general nutrition messages during the COVID-19 pandemic. The application of healthy living habits based on general guidelines for balanced nutrition for both self and family during this pandemic is very necessary to be maintained to support a healthy body. The role of health workers and the government is very much needed to continue to offer education and information related to balanced nutrition to support and become a habit in having a healthy lifestyle.

REFERENCES

1. Djalante, R., Lassa, J., Setiamarga, D., Sudjatma, A., Indrawan, M., Haryanto, B., Warsilah, H. Review and Analysis of Current Responses to COVID-19 in Indonesia: Period of January to March 2020. Progress in Disaster Science Journal: 2020; 6 (100091): 1-9.
2. Kementerian Kesehatan Republik Indonesia. Situasi Covid-19, Kondisi 18 Juni 2020. Kementerian Kesehatan Republik Indonesia; 2020.
3. Sekertariat Daerah Pemerintah Provinsi Nusa Tenggara Barat. Press Release Covid-19. Nusa Tenggara Barat; 2020.
4. Lingam, L., & Suresh, S. R. COVID-19, Physical Distancing and Social Inequalities: are we all Really in This Together. The International Journal of Community. 2020; 2(2): 173-190.
5. Badan Penelitian dan Pengembangan Kesehatan Kementerian Kesehatan RI. Laporan Nasional Riskesdas 2018. Jakarta; 2018.
6. Wu, D., Lewis, E. D., Pae, M., & Meydani, S. Nutritional Modulation Of Immune Function: Analysis Of Evidence, Mechanisms, and Clinical Relevance. Frontiers in Immunology Journal. 2019; 9(3160): 1-19.
7. Kementerian Kesehatan Republik Indonesia. Panduan Gizi Seimbang Pada Masa Pandemi Covid-19. Jakarta; 2020.
8. Fitri, Z., & Salam, A. Penerapan Pedoman Gizi Seimbang Pada Ibu Balita di Kelurahan Babakan. Jurnal Gizi Prima. 2017; 2(2): 148-153.
9. Nuryani. Validitas dan Reabilitas Kuesioner Pengetahuan, Sikap dan Perilaku Gizi Seimbang Pada Remaja. Jurnal Gizi dan Kesehatan. 2019; 3(2): 37-46.
10. Kementerian Kesehatan Republik Indonesia. Pedoman Gizi Seimbang. Jakarta; 2014.
11. Pratami, T. J., Widajanti, L., & Aruben, R. Hubungan Penerapan Prinsip Pedoman Gizi Seimbang denga Status Gizi Mahasiswa S1 Departemen Ilmu Gizi Universitas Diponegoro. Jurnal Kesehatan Masyarakat. 2016; 4(4): 561-570.
12. Agustina, & Permatasari, P. Hubungan Pengetahuan dan Penerapan Pesan Gizi seimbang Remaja dalam Pencegahan Anemia Gizi Besi. Jurnal Ilmiah Kesehatan Masyarakat. 2019; 11(1): 1-9.

13. Perdani, Z. P., Roswita, H., & Nurhasanah. Hubungan Praktik Pemberian Makan dengan Status Gizi Anak usia 5 Tahun di Pos Gizi Desa Tegal Kunir Lor Mauk. Jurnal KFT. 2016; 1(2): 17-29.
14. Stankevich, A. Explaining the Consumer Decision-Making Process: Critical Literature Review. Journal of International Businees Research and Marketing. 2017; 2(6): 7-14
15. Adriaansz, P. N., Rottie, J., & Lolong, J. Hubungan Konsumsi Makanan dengan kejadian Hipertensi pada lansia di Puskesmas Ranomuut Kota Manado. Jurnal Keperawatan. 2016; 4(1): 1-9.
16. Anjangsari, K. N., & Isnawati, M. Hubungan Konsumsi Softdrink, lingkar pinggang dan aktivitas fisik dengan kadar glukosa darah puasa pada wanita dewasa. Jurnal Of Nutrition College. 2015; 4(2): 162-170.
17. Soraya, D., Sukandar, D., & Sinaga, T. Hubungan Pengetahuan gizi, tingkat kecukupan zat gizi dan aktivitas dengan status gizi pada guru SMP. Jurnal Gizi Indonesia. 2017; 6(1): 29-36.
18. Rahman, N., Dewi, N. U., & Armawaty, F. Faktor-faktor yang berhubungan dengan perilaku makan pada remaja SMA Negeri 1 Palu. Jurnal Preventif. 2016; 7(1): 1-64.
19. Yusra, V. D., Machmud, R., & Yenita. Faktor-faktor yang berhubungan dengan tingkat pengetahuan wanita usia subur tentang SADARI di Nagri Painan. Jurnal Kesehatan Andalas. 2016; 5(3): 697-704.
20. Silalahi, E., Syarifuddin, & Sudibyo, M. Faktor-faktor yang mempengaruhi terhadap pengetahuan tentang lingkungan pada siswa tingkat SMP/MTs dan SMA/MAN Adiwiyata di Kota Labuhanbatu. Jurnal Pendidikan Biologi. 2016; 5(3): 146-153.
21. Putri, R. M., & Lasri. Pekerjaan, Tingkat Pendidikan dan Pengetahuan Ibu Pra Sekolah tentang Gizi Seimbang. Jurnal Care. 2016; 4(3): 78-87.
22. Wowor, P., Engkeng, S., & Kalesaran, A. F. Faktor faktor yang berhubungan dengan perilaku konsumsi jajanan pada pelajar di Sekolah Dasar Negeri 16 dan Sekolah Dasar Negeri 120 Kota Manado. Jurnal Kesmas. 2018; 7(5): 17-23.
23. Irnani, H., & Sinaga, T. Pengaruh Pendidikan Gizi terhadap pengetahuan, praktik gizi seimbang dan status gizi pada anak sekolah dasar. Jurnal Gizi Indonesia. 2017; 6(1): 58-64.
24. B, V. N., & Wahini, M. Faktor- Faktor yang mempengaruhi pola konsumsi makan remaja (Kasus di sekolah Menenengah kejuruan Negeri 8 Surabaya). Jurnal Boga. 2014; 3(3): 47-50.

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Antibacterial potential of red dragon fruit peel yogurt (hylocereus spp.) against bacillus subtilis bacteria in hypercholesterolemic wistar rats

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ABSTRACT

Background: Fruit peel is a part of red dragon fruit that weighed 30-35% of the fruit weight and has not been used optimally. Red dragon fruit peel contains fiber, vitamin, flavonoid, tannin, alkaloids and has the potential as an antibacterial. Red dragon fruit peel can be processed into yogurt.
Objectives: This research examined the antibacterial potential of red dragon fruit peel yogurt against Bacillus subtilis in hypercholesterolemic Wistar rats.
Materials and Methods: Materials tested in this study were negative control, positive control, and caecum of hypercholesterolemic Wistar. This study used chloramphenicol as the positive control (K+) and DMSO 10% as the negative control (K-). The K1; K2; K3 were orally administered with 1.8 mL; 2.7 mL; 3.6 mL of red dragon fruit peel yogurt, respectively. Red dragon fruit peel yogurt was administered daily for 28 days. Caecum was collected and tested for antibacterial activity using disk diffusion (Kirby Bauer). The Bacillus subtilis was obtained from the Microbiology Laboratory of Center for Food and Nutrition Studies Universitas Gadjah Mada.
Results: The average inhibition zone in K-; K+; K1; K2; K3 were 0.00±0.00 mm; 11.5±1.41 mm; 11.5±0.96 mm; 10.13±0.66 mm; 10.38±1.12 mm, respectively. The experimental animal groups, which received 2.7 mL and 1.8 mL of red dragon fruit peel yogurts, showed a significant difference compared to the positive control group (p= 0.026 and p=0.021, respectively). When the dose was increased to 3.6 mL, it showed no statistical difference in results (p=1.000).
Conclusions: Red dragon fruit peel yogurt has an antibacterial potential against Bacillus subtilis.

Keywords: Antibacterial; Bacillus subtilis; Caecum; Red dragon fruit peel; Yogurt.

BACKGROUND

Antibacterial is a substance produced by microbes or other sources that can inhibit growth and even eradicate pathogenic bacteria that are harmful to humans. Bacterial infection is one of the health problems in Indonesia. Gastrointestinal disorders are complaints caused by bacterial infection.[1] Pharmacological therapy using antibiotics is often chosen to treat bacterial infection problems. Bacterial infection therapy using antibiotics can cause problems. Antibiotics are components produced by microbes that can kill or inhibit the growth of other microbes. Antibiotics are also defined as a natural molecular inhibitor.[2] The problem that arises from antibiotic therapy is the occurrence of resistance. This resistance event is triggered by the use of antibiotics that are not following the proper dosage, thus causing pathogenic microorganisms to become resistant and infection therapy to be ineffective. Bacillus subtilis can survive in various environmental conditions on land, water, and even in anaerobic conditions inside the digestive tract. These bacterial spores can even be blown by the wind to allow long-distance migration.[3] Bacillus subtilis can activate its host defense system so that the host becomes resistant to pathogens. Bacillus subtilis is a gram-positive bacterium commonly found in rotten bread.[4]

The discovery of new antibacterial compounds derived from natural ingredients can be an alternative solution to overcome the problem of antibiotic resistance. The sensitivity of each microorganism to a drug determines the lowest drug levels that can inhibit the growth of microorganisms in vitro. These antibacterial compounds can be obtained from plants. Plants contain compounds that have antibacterial potential with a new mechanism of action and have not yet experienced resistance. One of the plants that can function as an antibacterial is red dragon fruit.

Red dragon fruit is one type of dragon fruit species widely found and consumed in Indonesia. Dragon fruit is commonly known as pitaya, comes from the *Cactaceae* family [5] and is a tropical fruit.

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Pitaya has a CAM (photosynthetic crassulacean acid metabolism pathway) which allows efficient use of water.[6] Dragon fruit peel is a part of dragon fruit that weighed 30-35% of the fruit weight and has not been used optimally.[7] Red dragon fruit peel contains polyphenols and other active compounds. Active compounds other than polyphenols that can be found on red dragon fruit peel are alkaloids, terpenoids, flavonoids, saponins, tannins, carotenes, and phytoalbumins. Polyphenols such as flavonoids are more commonly found in the peel of red dragon fruit than in the flesh. Polyphenols as phenol components have many antioxidant activities. Betalain is a component associated with anthocyanins and is a reddish pigment found in red dragon fruit peel.

Processing of products from red dragon fruit peel which are considered as waste has already begun. To date, people know that red dragon fruit peel has many properties such as antioxidants, anti-hypercholesterolemia, and anti-diabetics. In research on antibacterial activity of n-hexane fraction from red dragon fruit peel against Staphylococcus aureus ATCC 25923, it was known that the inhibition zone diameter of the n-hexane fraction at concentrations of 20 and 40 mg/mL are 11.17±1.69 mm and 12.80±1.11 mm respectively.[8] Another research using ethanol extract of red dragon fruit peel (Hylocereus polyrhizus) on Staphylococcus aureus ATCC 25923 in vitro showed that the minimum inhibitory concentration was obtained at a concentration of 25% and the best inhibition zone was produced by an extract with a concentration of 100%.[9] Other research on red dragon fruit peels also stated that red dragon fruit peel extract can inhibit the growth of Salmonella pullorum with an average inhibition zone diameter at concentrations of 60 mg/mL, 40 mg/mL, and 20 mg/mL are 9.6 mm, 9.4 mm and 9.3 mm, respectively.[10] Research on the potential of red dragon fruit peel has not been done much, and it can be a great opportunity to explore red dragon fruit peel. Septiana's pre-clinical research shows that steeping dragon fruit peel can reduce plasma MDA levels in Sprague Dawley dyslipidemic rats. MDA is one of the parameters for free radical compounds. An increase in MDA value indicates an increase in oxidative stress in body cells.[11] Mardiana's research on red dragon fruit peel shows that red dragon fruit peel can be processed into yogurt and has the potential to be a functional food.[12] Yogurt can support human health and survival and improve dysbiosis due to aging.[13] Red dragon fruit added to yogurt with concentrations of 3%, 5%, and 7% showed antimicrobial activity with moderate to high inhibition zones because the growth of E. coli reached the range of 6.06 - 15.29 mm. [9]Research on the antibacterial activity of red dragon fruit peel is only limited to the extraction process and there has been no further research related to processed dragon fruit peel products. This study aims to determine the antibacterial properties of red dragon fruit skin yogurt against Bacillus subtilis in hypercholesterolemic rats.

MATERIALS AND METHODS

Materials and Tools

This research uses experimental animal handling and facilities of CNFS laboratory, Universitas Gadjah Mada, Yogyakarta based on the Guidelines for Care and Use of Laboratory Animals. The ethical letter was obtained from the Committee on Ethics of Health, Universitas Negeri Semarang number 140/KEPK/EC/2019. House of Experimental Rats CNFS, Universitas Gadjah Mada, Yogyakarta Indonesia is the provider of Wistar rats used in this study. The inclusion criteria were male Wistar rats, active, aged 8-12 weeks, weighed 150-240 grams with fasting blood glucose level <110 mg/dL, while the exclusion criteria were Wistar rats with anatomical abnormalities and had a weight loss of more than 10% during the study. During the study, no Wistar rats were dropped out because all animals were able to follow the intervention until the end.

The laboratory environment is conditioned to have a dark/light cycle with a ratio of 12 hours:12 hours with a room temperature of 25±1°C to maintain room humidity. The cages were always kept clean by removing feces every day, to minimize the stress of Wistar rats during the study. Wistar rats were placed in individual cages made of stainless steel and received standard feed with ad libitum water access. Wistar rats had an adaptation period of seven days before the study was carried out.

Hypercholesterolemia Induction

Hypercholesterolemic conditions in Wistar rats were obtained through single intraperitoneal injection of feed powder made of 1% cholesterol and 0.5% cholic acid given for 14 days.[14]Cholesterol and cholic acid powder were obtained from Sigma Aldrich, Japan.

Experimental Design

Materials used in this study were caecum from each group of hypercholesterolemic Wistar rats. The study used were K-: DMSO 10% as negative control; K+: chloramphenicol as positive control; K1: caecum of hypercholesterolemic Wistar rats received red dragon fruit peel yogurt as much as 1.8 mL/kg b.wt/day; K2: caecum of hypercholesterolemic

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Wistar rats received red dragon fruit peel yogurt as much as 2.7 mL/kg b.wt/day and K3: caecum of hypercholesterolemic Wistar rats received red dragon fruit peel yogurt as much as 3.6 mL/kg b.wt/day. Other materials required in this study were aqua dest, nutrient agar, chloramphenicol, and Dimethylsulfoxide (DMSO).

The tools used were separating funnel, rotary evaporator, ose needle, petri dish, vennier caliper, incubator, and autoclave. Tools such as separating funnels, ose needles, petri dishes, and vernier caliper were sterilized in an autoclave for 15 minutes at 121°C by regulating a pressure of 1.5 atm having previously been washed clean, dried, and wrapped in paper.

Procedures for Producing Red Dragon Fruit Peel Yogurt

Red dragon fruit peel yogurt was produced based on research by Mardiana[13] and Putriningtyas.[15] Production of red dragon fruit peel yogurt begins with making a starter. Culture rejuvenation or yogurt starter making was carried out using Lactobacillus bulgaricus or Streptococcus thermophilus, each of which was inoculated in full cream pasteurized milk for 18 hours at 42°C. The next step is selecting or sorting the red dragon fruit peel. Fresh red dragon fruit peel is separated from the pulp and cleaned using running water. Red dragon fruit peel is steamed for 15 minutes to remove the distinctive aroma of the peel and reduce the pectin content in the red dragon fruit peel. Red dragon fruit peel is blended with full cream milk in a ratio of 1:4. Suspension of steamed red dragon fruit peel with full cream milk is then added with 10% sucrose, and covered with aluminum foil for the next pasteurization process at a temperature of 75-85^{o}C for 15 minutes. The result of the pasteurized mixture of steamed red dragon fruit peel, full cream milk, and sucrose is cooled to a temperature of 35-37^{o}C then added with 5% Lactobacillus bulgaricus and 5% Streptococcus thermophilus yogurt starter. The last step in making red dragon fruit peel yogurt is incubation at 42^{o}C for seven hours.

Testing the Antibacterial Activity of Red Dragon Fruit Peel Yogurt

The antibacterial activity test of the sample was carried out on Bacillus subtilis obtained from the Microbiology Laboratory of the Center for Food and Nutrition Studies, UGM. Bacillus subtilis was rejuvenated by inoculating pure culture into nutrient agar and incubated at 37°C for 24 hours. Antibacterial activity was measured using the Disc diffusion method (Kirby Bauer).[16] Bacteria from the rejuvenation media were spread on nutrient agar. Empty test discs that have been soaked for 15 minutes in each caecum group of hypercholesterolemic Wistar rats were hygienically placed on the surface of the nutrient agar. The empty discs were immersed in 10 mL of caecum from each group of hypercholesterolemic rats. Media containing test discs were incubated at 37°C for 24 hours and then the diameter of the clear zone was measured using a vernier caliper. The negative control used was DMSO 10%, which was dripped as much as 20 µL on a paper disc. The positive control used in this study was chloramphenicol 0.5 mg. Chloramphenicol is a broad-spectrum antibiotic so it is appropriate to inhibit the growth of gram-positive and negative bacteria. All numerical data were expressed as mean±standard deviation from twice measurements. One-way analysis of variance (ANOVA) with Tukey's test was used to determine significant differences ($p < 0.05$) between means.

RESULTS

Determination of the antibacterial activity of red dragon fruit peel yogurt was carried out by the Kirby-Bauer disc diffusion method, which is the determination of the sensitivity of bacteria to certain substances that may have antibacterial activity, using paper discs. The results of the antibacterial activity of red dragon fruit peel yogurt against Bacillus subtilis in each group can be seen in table 1.

The average inhibition zone of K+; K-; K1; K2; K3 are each 11.50±1.41 mm; 0.00±0.00 mm; 11.50±0.96 mm; 10.13±0.66 mm; 10.38±1.12 mm. DMSO 10% as negative control has no inhibition zone. The categories of antibacterial inhibitory strength are inhibition zone diameter of 5 mm or less is categorized as weak, inhibition zone diameter of 5-10 mm is categorized as a medium, inhibition zone diameter of 10-20 mm is categorized as strong while the diameter of more than 20 mm is categorized as very strong.[10] The positive control using chloramphenicol (K1), which is the caecum of the experimental animal group which received 1.8 mL of red dragon fruit peel yogurt for 28 days, shows the widest inhibition zone.

The DMSO negative control group shows significant differences in the inhibition zone of each treatment group ($p<0.05$). Based on these results, it can be concluded that the red dragon fruit peel yogurt has antibacterial activity against Bacillus subtilis. The difference in the results of the antibacterial activity test in each group can be seen in table 2. Table 2 shows that there is no difference between the positive control group and the experimental animal group that received red dragon fruit peel yogurt at a dose of 1.8 mL and 3.6 mL ($p> 0.05$). The

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experimental animal group that received red dragon fruit peel yogurt with a dose of 1.8 mL shows a significant difference when compared to the group that received yogurt with a dose of 2.7 mL (p= 0.021). Table 2 also shows that there is no statistical difference when the dose is increased to 3.6 mL (p> 0.05).

Table 1. Difference Mean

Group	Deuteronomy1 (mm)	Deuteronomy2 (mm)	Mean±SD (mm)
K-	0	0	0.00±0.00
K+	12.0	11.0	11.50± 1.41
K1	11.25	11.75	11.50± 0.96
K2	10.0	10.25	10.13± 0.66
K3	10.5	10.25	10.38± 1.12

K- (DMSO); K+ (chloramphenicol); K1 (caecum derived from red dragon fruit peel yogurt at a dose of 1.8 mL); K2 (caecum derived from red dragon fruit peel yogurt at a dose of 2.7 mL); K3 (caecum derived from red dragon fruit peel yogurt at a dose of 3.6 mL)

Table 2. Post Hoc

Group	Group	p*
K-	K1	0.001*
	K2	0.001*
	K3	0.001*
K+	K-	0.001*
	K1	0.219
	K2	0.026*
	K3	0.084
K1	K2	0.021*
	K3	0.370
K2	K3	1.000

p significant post hoc test (p<0,05); K- (DMSO); K+ (Chloramphenicol); K1 (caecum derived from red dragon fruit peel yogurt at a dose of 1.8 mL); K2 (caecum derived from red dragon fruit peel yogurt at a dose of 2.7 mL); K3 (caecum derived from red dragon fruit peel yogurt at a dose of 3.6 mL)*

DISCUSSION

Red dragon fruit peel yogurt in this study was made using Lactobacillus bulgaricus and Streptococcus thermophilus as starters. Probiotic activity in yogurt is largely determined by nutrient availability, inoculation levels, incubation temperature, fermentation time, storage conditions, pH, sugar concentration, and content of milk solids. Lactic acid bacteria in yogurt can break down complex carbohydrates into simple carbohydrates thus producing the final product in the form of lactic acid. Lactic acid as a result of fermentation metabolites shows the ability to inhibit microbial growth.[17]

This study used DMSO in negative control and chloramphenicol in positive control. DMSO is a good extract solvent because it can dissolve without affecting the growth of the bacteria so that it does not interfere with the results of the observation. Polar and nonpolar compounds can be dissolved by DMSO.[18] Chloramphenicol gives an inhibitory area, so this shows that chloramphenicol is still sensitive to Bacillus subtilis.[19]Gram-positive bacteria have better antibacterial sensitivity than gram-negative bacteria. The composition and structure of cell walls of gram-positive bacteria are more susceptible to chemical components than gram-negative bacteria.[20] This sensitivity is due to differences in the structure of cell walls. Gram-positive bacteria have a relatively simple cell wall structure, making it easier for antimicrobial compounds to enter the cell and find targets to work. The structure of the cell wall of gram-negative bacteria is relatively more complex and consists of three layers, the outer layer is a lipoprotein, the middle layer is a lipopolysaccharide, and the inner layer is peptidoglycan. The middle layer in the form of lipopolysaccharide acts as a barrier against various antibacterial bioactive materials while the inner layer in the form of peptidoglycan has a high-fat content.[1] The results in table 2 show that there is no difference in the three administration of dragon fruit peel yogurt with chloramphenicol positive control. There was no

significant difference in the group that received 2.4 mL and 3.6 mL of red dragon fruit peel yogurt, so it can be assumed that administering 1.4 mL of red dragon fruit peel yogurt can also be used as antibacterial because it has the same antibacterial activity as the 3.6 mL. The process of making yogurt can trigger the production process of proteolytic enzymes by lactic acid bacteria. This proteolytic enzyme can cut peptide bonds in milk protein to form free amino acids and peptides.

Some peptides show antibacterial activity. Potential peptides that have antibacterial activity derived from milk proteins include Leu-Arg-Leu-Lys-Lys-Tyr-Lys-Val-Pro-Gln-Leu. These peptides are produced by the hydrolysis of pepsin in cow casein.[21] Antibacterial activity of these peptides also depends on various properties such as peptide charges, amphipathicity, and hydrophobic and hydrophilic conditions. This antibacterial activity can also be caused by the presence of several cationic peptides which are believed to interact with lipopolysaccharide anion bonds in cell membranes. This peptide can replace divalent cations such as Ca^{2+} and Mg^{2+}, distorting to the cell's outer membrane bilayer because these two minerals are important to support the integrity of the cell's outer membrane. This distortion in the outer membrane results in membrane lysis and ultimately cell death.[22]

Antibacterial effectiveness of red dragon fruit peel yogurt is possible due to the presence of polyphenols in the peel of red dragon fruit, alkaloids, and terpenoids. The antibacterial activity of phenol compounds is carried out through a protein denaturation process and results in cell lysis. Flavonoids are derivatives of phenolic compounds and act as protein coagulators. The toxicity of phenol compounds to bacteria adjusts the number of hydroxyl groups and the concentration given. Phenol can bind to proteins through hydrogen bonds resulting in damage to the protein structure. [23]

Terpenoids have antibacterial activity by reacting with porin (a transmembrane protein) on the outer membrane of the bacterial cell wall to form strong polymer bonds that cause damage to porin. Porin is the entrance and exit of compounds so that when porin is damaged it will reduce the permeability of bacterial cell walls. This cell wall permeability will interfere with the entry of nutrients and other compounds, thus inhibiting bacterial growth and can even cause bacterial death. Luo stated that red dragon fruit peel contains β-amirin (15.87%) and α-amirin (13.90%) which are the terpenoid group.[22] Other than terpenoids, the antibacterial activity of dragon fruit peel yogurt is also caused by alkaloids. Alkaloids carry out the antibacterial activity by disrupting the peptidoglycan that makes up bacterial cells so that the cell wall layer is not formed completely and causes cell death. Another bioactive substance that is believed to have an antibacterial role in red dragon fruit peel yogurt is flavonoids. Mardiana's research stated that red dragon fruit peel yogurt using Lactobacillus bulgaricus and Streptococcus thermophilus bacterial isolates had higher levels of flavonoids compared to dragon fruit peel yogurt using commercial starters.[12] Flavonoids are known to have antibacterial activity due to their complex formation process with bacterial cell walls. Flavonoids work by damaging bacterial cell membranes in the phospholipid portion to further reduce membrane permeability. The decrease in membrane phospholipid permeability due to the activity of phenolic components causes lysis of bacterial cell membranes. Saponin activity mechanism as an antibacterial is possible because saponins can cause proteins and enzymes leakage in cells. Tannins also play a role in inhibiting bacterial growth by damaging bacterial cell proteins.[20] Other alkaloid compounds found in the peel of red dragon fruit are betacyanins (betanin and isobetanin). The fermentation process in making yogurt of red dragon fruit peel causes an increase in the content of betacyanin.[24] Alkaloids act as antibacterial through an inhibitory mechanism of DNA synthesis. Alkaloid compounds have alkaline groups that contain nitrogen which will react with amino acid compounds that make up the bacterial cell walls and DNA. This reaction will result in changes in the structure and arrangement of amino acids, causing changes in the genetic balance of the DNA chain. This will cause bacterial cell lysis which will then cause cell death in bacteria. [25]Alkaloids can also inhibit bacterial growth through the action mechanism of the dihydrofolate reductase enzyme so that it inhibits nucleic acid synthesis. [26]

The limitations of this study are that the phytochemical (tannins, alkaloids, and terpenoids) tests on red dragon fruit peel yogurt were not carried out.

CONCLUSIONS

Red dragon fruit peel yogurt with a dose of 2.7 mL has the potential to inhibit the growth of Bacillus subtilis bacteria.

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REFERENCES

1. Waglechner, N. & Wright, G. D. Antibiotic resistance : it’s bad, but why isn't ’ t it worse ?

BMC Biology. 2017; 15(84): 1–8.
2. Davies, J. & Davies, D. Origins and Evolution of Antibiotic Resistance. Microbiol. Mol. Biol. Rev. 2010; 74 (3): 417–433.
3. Earl, A. M., Losick, R. & Kolter, R. Ecology and Genomics of Bacillus subtilis. Trends Microbiol. 2008; 16(6): 1–11.
4. Rahayu, W. P. & Nurwitri, C. Mikrobiologi Pangan. IPB Press; 2012.
5. Jaafar, R. A., Ridhwan, A., Mahmod, N. Z. C. & Vasudevan, R. Proximate Analysis of Dragon Fruit (Hylocereus polyhizus). Am. J. Appl. Sci. 2009; 6(7): 1341–1346.
6. Jaya, I. K. D. Morphology And Physiology Of Pitaya And It Future Prospects In Indonesia. Crop Agro 2010; 3 (1): 44–50.
7. Nurliyana, R., Zahir, S., Suleiman, M., Aisyah, M. & Rahim, K. Antioxidant study of pulps and peels of dragon fruits : a comparative study. International Food Research Journal. 2010; 17: 367–375.
8. Amalia, S., Wahdaningsih, S. & Untari, E. K. Antibacterial Activity Testing Of N-Hexane Fraction Of Red Dragon (Hylocereus polyrhizus Britton & Rose) Fruit Peel On Staphylococcus aureus ATCC 25923. Traditional Medicine Journal 2014; 19(2): 89–94.
9. Fitratullah, A., Maruddin, F., Yuliati, F., Prahesti, K. & Taufik, M. Addition of red dragon fruit (Hylocereus polyrhizus) on yogurt : Effect on lactic acid content, pH, and the inhibition of Escherichia coli growth. in *IOP Conf. Series: Earth and Environmental Science 2019; 343:* 1–7 (IOP Publishing). doi:10.1088/1755-1315/343/1/012034.
10. Maulana, I. *et al.* Antibacterial Test of Red Dragon Fruit Extract Peel (Hylocereus polyrhizus) Against Bacteria Salmonella pullorum. Jurnal Medika Veterinaria 2018; 12 (1): 9–14.
11. Septiana, W. C. & Ardiaria, M. Efek Pemberian Seduhan Kulit Buah Naga Merah (Hylocereus polyrhizus) terhadap Kadar Malondialdehyde (MDA) Tikus Sprague Dawley Dislipidemia. Journal of Nutrition College 2017; 5 (4): 344–352.
12. Mardiana, Budiono, I. & Putriningtyas, N. D. Comparison of organoleptic, protein, lipid and flavonoid content of commercial starter and isolated culture red dragon fruit peel yogurt. Food Research. 2020: 4(3): 920–925.
13. Ayala, F. R. *et al*. Microbial flora, probiotics, Bacillus subtilis and the search for a long and healthy human longevity. Microbial Cell 2017; 4 (4): 133–136.
14. Murase, T., Yamada, N. & Uchimura, H. Accumulation of intermediate-density lipoprotein in the plasma of cholesterol-fed hypothyroid rats. Metabolism 1983; 32 (2): 146–150.
15. Putriningtyas, N. D. & Wahyuningsih, S. Potensi yogurt kacang merah (Phaseolus vulgaris L) ditinjau dari sifat organoleptik, kandungan protein, lemak dan flavonoid. Jurnal Gizi Indonesia (The Indonesian Journal of Nutrition). 2017*;* 6 (1): 37–43.
16. Prayoga, E. Perbandingan Efek Ekstrak Daun Sirih Hijau (Piper betle L.) dengan Metode Difusi Disk dan Sumuran terhadap Pertumbuhan Bakteri Staphylococcus aureus. [Tesis]. Jakarta: Universitas Islam Negeri Syarif Hidayatullah; 2013. doi:10.1023/B:FOOP.0000019620.04821.a2.
17. Maruddin, F., Malaka, R. & Taufi, M. Characteristics and antimicrobial activity of dangke whey fermentation with sugar addition. Bulgarian Journal of Agricultural Science. 2019; 25 (2): 410–417.
18. Octaviani, M., Fadhli, H. & Yuneistya, E. Uji Aktivitas Antimikroba Ekstrak Etanol dari Kulit Bawang Merah (Allium cepa L.) dengan Metode Difusi Cakram. Pharm. Sci. Res. 2019*;* 6 (1): 62–68.
19. Fitriani, E. Uji Aktivitas Antibakteri Ekstrak Etanol Daun Sirsak (Annona muricata L.) terhadap Shigella flexneri Secara In Vitro. [Tesis]. Pontianak: Universitas Tanjungpura; 2014.
20. Ritarwan, K. & Nerdy, N. Antibacterial Activity of Red Dragon Fruit Leaves Extract and White Dragon Fruit Leaves Extract Against Meningitis Bacterial. Orient. J. Chem. 2018; 34 (5): 2534–2538.
21. Sah, B. N. P., Vasiljevic, T., Mckechnie, S. & Donkor, O. N. Antibacterial and antiproliferative peptides in synbiotic yogurt — Release and stability during refrigerated storage. J. Dairy Sci. 2016; 99: 4233–4242.
22. Luo, H., Cai, Y., Peng, Z., Liu, T. & Yang, S. Chemical composition and in vitro evaluation of the cytotoxic and antioxidant activities of supercritical carbon dioxide extracts of pitaya (dragon fruit) peel. Chemistry Central Journal. 2014; 8 (1): 1–7.
23. Karimi, S., Azizi, F., Nayeb-aghaee, M. & Mahmoodnia, L. The Antimicrobial Activity of Probiotic Bacteria Escherichia coli Isolated

from Different Natural Sources Against Hemorrhagic E. coli O157:H7. Electron. Physician 2018; 10 (3): 6548–6553.
24. Choo, K. Y., Kho C., Ong, Y. Y., Thoo, Y.Y., Lim, R.L. H., Tan, C.P., et al. Studies on the storage stability of fermented red dragon fruit (Hylocereus polyrhizus) drink. Food Sci. Biotechnol. 2018; 27: 1411–1417.
25. Wahdaningsih, S., Untari, E. K. & Fauziah, Y. Antibakteri Fraksi n-Heksana Kulit Hylocereus polyrhizus Terhadap Staphylococcus epidermidis dan Propionibacterium acnes. Pharm. Sci. Res. 2014; 1 (3): 180–193.
26. Astridwiyanti, A. A. B., Mahendra, A. N. & Dewi, N. W. S. Uji efektivitas ekstrak etanol kulit buah naga merah (Hylocereus polyrhizus) terhadap Staphylococcus aureus ATCC 25923 secara in vitro. Intisari Sains Medis 2019; 10 (3): 482–486.

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Differences in triglyceride levels before and after whey protein intervention in field workers exposed and unexposed to arsenic

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ABSTRACT

Background: A work environment that is exposed to heavy metals, such as a coal mining environment, can change fat metabolism in the body. Changes in fat metabolism will lead to cardiovascular disease. Consumption of dairy products, e.g. whey protein, can reduce the risk of metabolic disorders and cardiovascular disease.
Objectives: To test and analyze the differences in triglyceride levels before and after whey protein intervention to field workers with different working conditions.
Materials and Methods: This experimental research with pretest and posttest was conducted on field workers at PT Bukit Asam Tbk. Tarahan Port Unit, Lampung as an Arsenic exposed group and PTPN VII Way Berulu Business Unit as an Arsenic unexposed group. Both groups received 24 grams of whey protein daily for 28 days. The triglyceride levels before and after the intervention were measured by laboratory analysis using the enzymatic calorimetry method. The data were analyzed using independent t-test, Mann Whitney test, and Wilcoxon test.
Results: The average triglyceride levels increased by 50.48 ± 98.09 mg/dL in the exposed group and 16.78 ± 67.67 mg/dL in the unexposed group. There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups.
Conclusions: The whey protein intervention increased the triglyceride level in the exposed group and decreased it in the unexposed group.

Keywords: Triglycerides; Whey Protein; Arsenic (As).

BACKGROUND

With a good nutritional status and health status, the workforce will provide maximum results for the company. The work environment greatly influences their nutritional needs and health status. The work environment with exposure to chemical substances will affect the health status of the workers if it is not properly attended to by the manager.[1] Mining work environment, such as coal mining, is susceptible to the exposure of hazardous chemicals. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.[2,3] Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. It can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.[4]

Some epidemiological studies have shown that the consumption of dairy products can reduce the risk of metabolic disorders and cardiovascular disease.[5] Protein in cow's milk consists of 80% casein and 20% whey protein, both of which are rich in amino acids.[6] Whey protein is a food source that has high protein and essential amino acids, as well as branched-chain amino acids (BCAA) that play an important role in tissue growth and repair.[5] The results of the previous studies indicate that after the intervention of whey protein on 20 male subjects aged 30 to 50 years with relatively high serum cholesterol levels (≥ 200 mg/dL), their serum triglyceride levels decreased and their serum HDL levels increased after 8 weeks of intervention.[7]

MATERIALS AND METHODS

This research is an experimental study with pretest and posttest. The sample was assigned into two groups, Arsenic (As) exposed group (field workers of PT Bukit Asam Tbk. Tarahan Port Unit, Lampung, a coal mining product management company) and Arsenic (As) unexposed group (field workers of PT Perkebunan Nusantara VII Way Berulu Business Unit, a company engaged in the

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management of rubber plantations). Both groups received intervention in the form of taking whey protein for 28 days[8] with a dose of 24 grams per day dissolved in 300 ml of mineral water.[9] The variable measured was the subjects' triglyceride levels before and after the whey protein intervention.

This research was conducted after obtaining permission from the Health Research Ethics Committee of the Faculty of Medicine, Diponegoro University, Semarang, from January to March 2020. Whey protein intervention was carried out at the research site, PT Bukit Asam Tbk. Tarahan Port Unit in Lampung and PT Perkebunan Nusantara VII Way Berulu Business Unit. Subject characteristic data including name, age, and place of birth were collected through interviews using a questionnaire. The anthropometric data of the subjects consisted of body weight, measured using digital scales, and height, measured using a microtoise. Body mass index (BMI) was calculated using the BMI calculation formula based on measurements of weight and height. The physical activity data of the subjects were collected through interviews using the Physical Activity Level (PAL) questionnaire and then calculated with the PAL calculation formula. Food intake data were collected through interviews using a 24-hour food recall questionnaire 3 times each week during the study. The triglyceride levels before and after the intervention were measured by laboratory analysis using the enzymatic calorimetry method.

Data analysis was carried out through the SPSS program with the Shapiro-Wilk test to determine the normality of data distribution for each group, independent t-test, and Mann Whitney test to determine the average difference between the subject's characteristics and nutritional intake variables. Wilcoxon test was also used to determine differences in triglyceride levels before and after the intervention.

RESULTS

The data in this study are primary data taken from the research sample, field workers exposed to different work environments. The total sample consisted of 64 people (31 from the exposed group and 33 from the unexposed group).

Subject Characteristics

Subject characteristics data consisted of age, body weight (BW), body height (BH), body mass index (BMI), and physical activity level (PAL). They are shown in Table 1.

Table 1. Subject Characteristics in Both Groups

	Exposed (n = 31)		Unexposed (n = 33)		*p*
	Mean±SD	**minimum-maximum**	**Mean±SD**	**minimum-maximum**	
Age (year)	29.84±8.84	20-56	38.94±4.50	28-54	0.000[2]
Body weight (kg)	70.87±14.11	45-109	56.55±6.96	44.08-75.00	0.000[1]
Body height (cm)	168.03±5.89	154-179	162.01±4.84	153-171	0.000[1]
BMI (kg/m^2)	25.31±4.21	18.09-36.84	21.55±2.26	17.56-26.23	0.000[1]
PAL (unit)	1.57±0.13	1.41-1.87	1.99±0.24	1.44-2.78	0.000[2]

[1] difference test using Independent T-Test, [2] difference test using Mann Whitney

Table 1 shows significant differences in all variable subject characteristics ($p < 0.05$). The average weight, height, and BMI were higher in the exposed group compared to the unexposed group, whereas the average age and PAL were higher in the unexposed group.

Nutritional Intake

Nutritional intake data consists of total intake per day and intake adequacy. Nutritional intake data are presented in Table 2 and the data on the intake adequacy levels in both groups in Table 3.

Table 2. Overview of Total Nutritional Intake Per Day in Both Groups

	Exposed (n = 31)		Unexposed (n = 33)		*p*
	Mean±SD	**minimum-maximum**	**Mean±SD**	**minimum-maximum**	
Energy (kcal)	2018.72±452.88	1313.50-3314.90	2149.83±515.61	1213.20-3890.00	0.285[1]
Protein (g)	96.21±29.04	56.10-180.90	86.62±26.66	56.30-167.30	0.116[2]
Fat (g)	83.89±40.50	35.80-153.30	67.01±30.06	22.30-170.50	0.134[2]
Carbohydrate (g)	252.30±68.47	148.7-452.80	298.47±66.39	191.80-485.90	0.004[2]

[1] difference test using Independent T-Test, [2] difference test using Mann Whitney

Table 2 shows that the average energy and carbohydrate intake in the unexposed group was higher than those of the exposed group. The average protein and fat intake show a higher value in the exposed group. There was no significant difference in the average energy, protein, and fat intake, but there was a significant difference in the average carbohydrate intake (p = 0.004).

Table 3. Nutritional Intake Adequacy Level in Both Groups

Adequacy Level		Category	
		Exposed (n = 31)	Unexposed (n = 33)
Energy	Deficient	23 (74.2%)	26 (78.8%)
	Sufficient	8 (25.8%)	7 (21.2%)
Protein	Deficient	14 (45.2%)	21 (63.6%)
	Sufficient	17 (54.8%)	12 (36.4%)
Fat	Deficient	13 (41.9 %)	22 (66.7%)
	Sufficient	15 (48.4%)	9 (27.3%)
	Excessive	3 (9.7%)	2 (6.1%)
Carbohydrate	Deficient	28 (90.3%)	26 (78.8%)
	Sufficient	3 (9.7%)	7 (21.2%)

Table 3 shows that almost all of the adequacy levels of energy, protein, fat, and carbohydrate intakes in both groups were insufficient. Meanwhile, the fat adequacy of 9.7% of the total subjects in the exposed group and 6.1% in the unexposed group fell into the excessive category.

Triglyceride Levels Before and After Whey Protein Intervention

The triglyceride levels before and after whey protein intervention were tested for normality using the Shapiro Wilk test, resulting in the abnormal distribution of triglyceride levels with a p-value of < 0.05; thus, the Wilcoxon test was used to see the differences in triglyceride levels before and after the whey protein intervention.

Table 4. Triglyceride Levels Before and After Whey Protein Intervention in Both Groups

Triglyceride Levels (mg/dL)	Exposed (n = 31)		Unexposed (n = 33)		*p*
	Mean±SD	minimum-maximum	Mean±SD	min-max	
Before	125.32±68.97	35.00-310.00	157.18±104.82	38.00-562.00	0.202[1]
After	175.80±127.99	48.00-590.00	140.39±80.78	43.00-425.00	0.344[1]
Difference/delta (Δ)	50.48±98.09	414.00(-68.00)	-16.78±67.67	184.00(-137.00)	0.000[1]
p	0.003[2]		0.038[2]		

[1] difference test using Mann Whitney, [2] difference test using Wilcoxon

Table 4 shows that the average triglyceride level before the intervention in the unexposed group was higher than in the exposed group, and the average triglyceride level after the intervention in the exposed group was higher. The average triglyceride level in the exposed group increased by 50.48 ± 98.09 mg/dL and in the unexposed group decreased by 16.78 ± 67.67 mg/dL. The Wilcoxon test results showed differences in triglyceride levels in both groups (p < 0.05).

DISCUSSION

This study explores the differences in triglyceride levels in field workers with different work environment exposures before and after the whey protein intervention. The groups in this study consisted of field workers in a coal mining environment with exposure to Arsenic, an obesogenic substance, and field workers in a rubber plantation environment. Whey protein intervention was carried out for 28 days with a dose of 24 grams per person per day.

The results showed that the intervention could reduce triglyceride levels in the obesogen unexposed group. Meanwhile, in the exposed group, there was an increase in triglyceride levels after the intervention. The decrease in triglyceride levels in the unexposed group was due to several factors, such as lower BMI value, higher PAL value, and lower fat intake of the unexposed group compared to those of the exposed group.

On the other hand, the triglyceride level in the exposed group was found to increase after the whey protein intervention. This is due to several factors, including higher BMI value, lower PAL, and higher

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fat intake, as well as Arsenic exposure that occurs every day in the work environment of the exposed group.

The BMI value of the unexposed group was lower than that of the exposed group. Previous research has suggested that there is a positive correlation between BMI and triglyceride levels. High BMI is directly proportional to triglyceride levels in the body.[10] The linear relationship between BMI and triglyceride levels is seen in individuals with higher BMI between 18 to 35 kg/m^2, posing a 5.1% greater risk of having high triglyceride levels.[11] The PAL value of the exposed group was lower than that of the unexposed group. Previous research found that moderate to heavy PAL could increase the average HDL level between 0.89 to 1.71 mg/dL and reduce the average triglyceride level between 0.93 to 0.98 mg/dL.[12] Besides, fat intake also greatly affects triglyceride levels in the body. Based on the results of the 3×24 hour recall in the study, fat intake in the exposed group was higher than that in the non-exposed group. The results of statistical tests also indicate the same, where the average fat intake of the exposed group is higher than that of the unexposed group. Previous research has shown that an increase in fat by 10% of daily needs will lead to increased triglyceride levels by 0.06 mmol/L.[13]

Arsenic exposure that occurs every day in the work environment of the exposed group also influences changes in lipid metabolism in the body. Based on a study conducted on ApoE mice exposed to moderate Arsenic, it was found that mice with a normal diet and received 200 ppb-Arsenic exposure for 8 weeks had a significant increase in plasma triglyceride levels and triglyceride levels. The average triglyceride levels of mice exposed to 0 ppb, 200 ppb, and 1,000 ppb of Arsenic for 8 weeks were 138 ± 18 mg/dL, 233 ± 29 mg/dL, and 178 ± 26 mg/dL, respectively. This implies that chronic Arsenic exposure will increase triglyceride levels in the body.[14]

Triglycerides or triacylglycerols are formed from three fatty acids and mono glycerol. Triglycerides function as energy substances. As cells need energy, the lipase enzyme in fat cells breaks down triglycerides into glycerol and fatty acids to be released into the blood vessels.[15] Triglyceride synthesis is influenced by Growth Hormone (GH). GH increases the activity of Hormone Sensitive Lipase (HSL) which can break down triglycerides into free fatty acids in fat tissue.[15] Triglyceride synthesis occurs when the energy source from carbohydrates is sufficient, and then the fatty acids will experience esterification, forming esters with glycerol and triglycerides as long-term energy reserves. Once there is no energy reserve from carbohydrates, the fatty acids and triglyceride reserves in the tissues will be broken down through the lipolysis process. Triglyceride levels in the body are influenced by several factors including genes, age, gender, food intake, obesity, physical activity, and smoking habits. The accumulation of excess triglyceride levels in the body will increase the risk of dyslipidemia. Dyslipidemia is a condition of an abnormal lipid profile in the body that increases levels of total cholesterol, triglycerides, and LDL and decreases HDL levels in the blood.[16]

Milk consists of two protein contents, 80% casein and 20% whey. When casein is separated from whole milk, the whey will remain and contain 68% protein.[5] Whey protein is a liquid byproduct of cheese making. Whey protein consists of several biological components, including BCAA which are easily absorbed and used by the body, lactoferrin as a non-enzymatic antioxidant, immunoglobulin as an antibody, β-Lactoglobulin as BCAA that plays a role in retinol-binding and can modulate the lymphatic response in humans, α-Lactalbumin which can increase antibody response to systematic antigen stimulation and suppress T cell response in lymphocytes, lactoperoxidase which can remove harmful bacteria for the human body, glycomacropeptide, and serum albumin.[17]

The high protein and amino acid content in whey protein make it an alternative food source of protein and amino acids with many health benefits. Increased protein intake can lead to changes in circulating lipids and lipoprotein levels, such as decreased LDL, decreased serum triglycerides, and increased HDL concentrations.[18] Research on female Wistar rats showed that whey protein concentrate supplementation could reduce cholesterol levels and serum triglyceride concentrations after consumption for 8 weeks compared to casein supplementation.[19] The decreased lipid profile levels with whey protein consumption occur through the role of BCAA in whey which accelerates digestion and absorption rates in intestines. β-Lactoglobulin content helps the absorption of cholesterol in the intestine, inhibits the expression of genes in charge of the absorption and synthesis of intestinal fatty acids and cholesterol, and increases the excretion of fecal steroids.[20]

Global competition demands a productive workforce for companies. A productive workforce will be created through good nutritional and health status. Some epidemiological studies have shown that nutrient intake is positively associated with labor productivity.[1] The work environment condition is one factor that influences the health and nutritional status of the workforce. A work environment with exposure

to hazardous chemicals such as a mining environment can affect the health status of workers. Coal, one of the largest natural resources in Indonesia, creates many job opportunities in the coal mining environment. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.[2,3] Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. Arsen can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.[4]

LIMITATION

This study did not analyze the levels of Arsenic (As) in the body of the research subjects, so the exact amount of exposure that affects triglyceride levels in the body remains unknown. Further research on this topic still needs to be conducted.

CONCLUSIONS

There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups. Whey protein intervention led to an increase in triglyceride levels in the exposed group and a decrease in triglyceride levels in the unexposed group although only a slight decrease was produced with the whey protein intervention for 28 days.

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REFERENCES

1. Suma'mur. Higiene Perusahaan dan Kesehatan Kerja (Hiperkes). Jakarta: Sagung Seto; 2013.
2. Radi, F. Hasni, M.J. Obesogens As An Environmental Risk Factor For Obesity. Malays J Public Health Med. 2014;14 (3):63–70.
3. Baillie-Hamilton P. Chemical Toxins: A Hypothesis To Explain The Global Obesity Epidemic. J Altern Complement Med. 2002;8:185–92.
4. Ceja-Galicia ZA, Daniel A, Salazar AM, Panico P, Ostrosky-Wegman P, Diaz-Villasenor A. Effects of arsenic on adipocyte metabolism: Is arsenic an obesogen? Elsevier Mol Cell Endocrinol Xxx. 2017;1-8.
5. Marshall K. Therapeutic applications of whey protein. Altern Med Rev. 2004;9:136–156.
6. Furst P, Stehle P. What Are The Essential Elements Needed For The Determination of Amino Acid Requirements in Humans ? J Nutr. 2004;134:1558–65.
7. Kawase M, Hashimoto H, Hosoda M, Morita A, Hasono A. Effect of Administration of Fermented Milk Containing Whey Protein Concentrate to Rats and Healthy Men on Serum Lipids and Blood Pressure. J Diet Sci. 2000; Vol. 83, No. 2.
8. Bhathena S, Velasquez M. Beneficial Role Of Dietary Phytoestrogens In Obesity And Diabetes. Am J Clin Nutr. 2002;76:1191–1201.
9. Vatani, DS, Golzar, FAK. Changes in Antioxidant Status and Cardiovascular Risk Factors of Overweight Young Men After Six Weeks Supplementation of Whey Protein Isolate and Resistance Training. Elsevier Ltd. 2012;59(Appetite):673–8.
10. Shamai L, Lurix E, Shen M, Novaro GM, Szomstein S, Rosenthal R, et al. Association of Body Mass Index and Lipid Profiles: Evaluation of a Broad Spectrum of Body Mass Index Patients Including the Morbidly Obese. 2010;21 (1)(Obesity Surgery):42–47.
11. Hansen SEJ, Madsen CM, Varbo A, Nordestgaard BG. Body mass index, triglycerides and risk of acute pancreatitis: A population-based study of 118,000 individuals. 2019;(The Journal of Clinical Endocrinology&Metabolism.).
12. Silva Rc, Diniz M, Alvim S, Vidigal P, Fedeli L, Barreto S. Physical Activity and Lipid Profile in the ELSA-Brasil Study. 2016;107(1):10–9.
13. Kdekian A, Alssema M, Van Der Beek EM, Greyling A, Vermeer MA, Mela DJ, et al. Impact of isocaloric exchanges of carbohydrate for fat on postprandial glucose, insulin, triglycerides, and free fatty acid responses-a systematic review and meta-analysis. Eur J Clin Nutr. 2019.

14. Lemaire M, Lemarié CA, Flores Molina M, Schiffrin EL, Lehoux S, Mann KK. Exposure to Moderate Arsenic Concentrations Increases Atherosclerosis in ApoE−/− Mouse Model. Oxf Univ Press. 2011;122 (1)(Toxicological Sciences):211–221.

15. Karyadi E. Hidup Bersama Penyakit Hipertensi, Asam Urat, Jantung Koroner. Jakarta: PT. Intisari Mediatama; 2006.

16. Lichtenstein AH. A. H. Diet and Lifestyle Recommendations Revision 2006: A Scientific Statement From the American Heart Association Nutrition Committee. Dalam 2006. Page 82–96. (Circulation; vol. 114 (1)).

17. Anthony JC, Anthony TG, Kimball SR, Jefferson LS. Signaling Pathways Involved In Translational Control Of Protein Synthesis In Skeletal Muscle By Leucine. J Nutr. 2001;131:856S-860S.

18. Pal S, Radavelli-Bagatini S. The Effects Of Whey Protein On Cardiometabolic Risk Factors. Obes Rev. April 2012.

19. Zhang, X., Beynen, A. C. Lowering effect of dietary milk-whey protein v. casein on plasma and liver cholesterol concentrations in rats. 1993;70(British Journal of Nutrition):139.

20. Pal S, Ellis V, Dhaiwal S. Effects of Whey Protein Isolate on Body Composition, Lipids, Insulin and Glucose in Overweight and Obese Individuals. Br J Nutr. 2010;104:716-723.

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The effect of additional protein, phosphatidylcholine, phosphatidylserine, and inulin on S100β levels of acute ischemic stroke patients at Dr. Kariadi Central Hospital, Semarang

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ABSTRACT

Background: The brain releases biochemical substrates, such as S100β protein, into circulation in response to ischemic conditions as a sign of damage in nerve cells and disruption of the blood-brain barrier's integrity. Thrombolytic therapy has led to the development of many neuroprotective therapies such as protein, phosphatidylcholine, phosphatidylserine, and inulin, which can be added to food products. Protein, phospholipids, and inulin, have a neuroprotective impact on nerve cells in the brain and blood-brain barrier.
Objective: To prove the effect of protein, phosphatidylcholine, phosphatidylserine, and inulin on S100β levels and clinical outcomes in patients with acute ischemic stroke.
Materials and Methods: This study was done in a single-blind RCT. Eighteen ischemic stroke patients were randomly divided into nine subjects for the intervention group and nine subjects for the control group. The Control group received 250 ml conventional formula milk (11.8 g protein) 3 times/day. The intervention group received 250 mL commercial milk 3 rimes/day which contained 15 g protein with 128 mg phosphatidylcholine, 32 mg phosphatidylserine, and 3 g inulin. All of the groups were given hospital-standard therapy for ischemic stroke. S100β levels were measured at pre and post-intervention.
Results: Pre and post S100β levels in intervention and the control group did not show any statistically difference (p = 0.777 and p = 0.096), but there was a trend of decreasing levels of S100β in the intervention group (-24.6 ± 252.0 pg/mL) versus control group (135.8 ± 216.2 pg/mL).
Conclusions: The addition of protein, phosphatidylcholine, phosphatidylserine, and inulin did not have a significant effect on S100β levels.

Keyword: Protein; Phosphatidylcholine; Phosphatidylserine; Inulin; S100β; Stroke

BACKGROUND

Ischemic stroke is the most common stroke that occurs when blood vessels in the brain are clogged by plaque/embolism. Clogged blood vessels interfere with blood flow going to the brain, reducing the supply of oxygen and glucose to the brain. This condition could lead to the death of nerve cells in the brain (apoptosis).[1,2] Damaged and dead nerve cells will cause some biochemical substrates to leave the brain into the circulatory system in response to ischemic conditions. This clog could also result in reduced proteins that play a role in maintaining the integrity of the blood-brain barrier membrane, thus the membrane increases its permeability. This, in turn, causes several biochemical substrates that should stay in the components of the brain to move into the circulatory system.[1–6]

Various biochemical substrates released in response to ischemic conditions play important roles in triggering brain tissue damages[6], one of them is S100β protein. This protein helps to regulate intracellular calcium levels[3], where excessive intracellular Ca^{2+} levels will lead to apoptosis (death of nerve cells).[2,5] Excessive intracellular Ca^{2+} caused by the decrease in the supply of blood that carries oxygen and nutrients to the brain, so the brain will lack energy (ATP) to be function normally. The Ca^{2+} ATPase pump which normally picks up Ca^{2+} into the

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organelle becomes inactive due to lack of ATP in the brain, resulting in the accumulation of intracellular Ca^{2+}. Accumulation of intracellular Ca^{2+} will removes Mg^{2+} ions in charge to keep N-methyl-D-aspartate (NMDA) receptors from being active. Activation of the NMDA receptor will cause stimulation of the post-synaptic membrane, where this membrane is 5 times higher in permeability for Ca^{2+}, so the intracellular Ca2+ ion becomes excessive, which will lead to nerve cell apoptosis. Apoptosis will cause the release of S100β protein in brain components to move into the circulation. S100β protein secretion increases along with the response of glial cells due to metabolic disorders such as head injury, damaged blood-brain barrier, and ischemia.[6]

Damage to cells in the brain will also trigger the activation of catabolic hormones that the patient will experience hypercatabolism.[7] Hypercatabolism, if not treated promptly, will worsen the patient's nutritional status.[7] Also, stroke patients also experience intestinal dysbiosis through immunological pathways.[8–11] Dysbiosis is a condition in which qualitative and quantitative changes occur in the composition, distribution, and metabolic activity of microbes in the intestines, causing adverse effects and worsening the patient's clinical outcome.

Appropriate therapy is the key in ischemic stroke patients.[12,13] One of the pharmacological therapies given is thrombolytic therapy (rTPa).[14] This therapy cannot be given to all acute ischemic stroke patients due to very strict indication criteria, especially in terms of duration (time window). The best timeframe for the administration of this therapy, which can provide the benefit of brain functional improvement and can reduce mortality, is <3 hours and ranged between 3-4.5 hours after symptom onset.[15,16] If the therapy is given not according to the guidelines, it can cause side effects such as the risk of bleeding in the brain and gastrointestinal tract, and angioedema.[14]

This situation has led to the development of many neuroprotective therapies, namely the addition of phospholipids which can be given directly in the form of supplements (cytolin) or fortified in food products such as milk. Phospholipids are a type of fat found in many nerve cell membranes. There are several types of phospholipids, the common ones are phosphatidylcholine and phosphatidylserine. Phosphatidylcholine increases the biosynthesis of membrane phospholipids which is degraded by an increase in free radicals during brain ischemia (neuroprotection).[12,17] Phosphatidylcholine also inhibits the activation of enzymes that trigger apoptosis of nerve cells,[12,17] thus affecting S100β levels. Also, the protein contained in milk will increase protein intake, muscle mass, and possibly increase body mass index (BMI). Therefore, it can improve the patient's motoric function and prevent the patient from experiencing a decrease in nutritional status.[18–20]

One of the nutrients that need to be given to ischemic stroke patients is inulin. Studies regarding the effect of inulin in improving ischemic conditions are still very limited in ischemic stroke patients and there are no studies about the direct effect of inulin on blood biomarkers of brain damage such as S100β protein. So far research has proven that Short-Chain Fatty Acid (SCFAs) acts as neuroprotective agents in the nerve cells. Inulin will be quickly fermented by *Bifidobacteria* and *Lactobacilli* (probiotic bacteria) and will produce SCFA in the form of acetic acid, propionic, butyrate, L-lactate, CO2, and hydrogen as fermentation products.[21,22] SCFA can be used as the source of energy (ATP) by the central nervous system because long-chain fatty acids cannot cross the blood-brain barrier membrane. SCFA has been proved to play a neuroprotective role, synthesizing neurotransmitters, and modulating the immune system. Thus, SCFA can improve dysbiotic conditions due to ischemic stroke.[23–25]

Dr. Kariadi Central Hospital is an "A-Accredited" Hospital that has a Stroke Unit with an average of two new stroke patients every day. The number of stroke patients in this hospital continues to increase.[26] This research aimed to prove the effect of additional protein, phosphatidylcholine, phosphatidylserine, and inulin on S100β levels and clinical outcomes of acute ischemic stroke patients at Dr. Kariadi Central Hospital, Semarang.

MATERIALS AND METHODS

A randomized control trial with single-blind was used as the design of this research. The effect of additional protein, phosphatidylcholine, phosphatidylserine, and inulin was seen through S100β levels. The research subjects were acute ischemic stroke patients in the inpatient room of Rajawali 1A of Dr. Kariadi Central Hospital Semarang amounted to 18 people (control = 9, intervention = 9), 10 subjects were male and 8 subjects were female. The research lasted for 4 months (February-May 2020). Acute ischemic stroke patients were selected to participate if their attack onsets were <72 hours and aged > 18 years.

Also, the researchers had randomized the subjects through Consecutive Sampling and provided informed consent to the patient/patient's family. During the implementation of the research, 7 patients dropped out or lost to follow-up with the following details: 4 patients not domiciled in Semarang, 2 patients died during the intervention, and 1 patient was forced to move out from hospital because of cost limitation.

The physical and neurological examinations had been performed by the doctor in charge of treating those patients. Supporting data, such as weight, blood pressure, blood glucose, lipid profile, smoking history, rTPa therapy status, and history of recurrent stroke had been taken from the patients' medical records once when the patient entered the stroke ward only to determine the risk factor data such as obesity, hypertension, diabetes mellitus, dyslipidemia, and smoking history. The researchers assessed the subject's dietary intake of energy, carbohydrate, protein, fat, phosphatidylcholine, phosphatidylserine, and inulin every day using the 24 hours recall method. The intake data consumed by the subjects will be compared with the patient's requirement to obtain the adequacy of nutritional intake which is expressed in percentage (%) with the following formula:

$$\text{Adequacy intake:} \frac{nutrient\ intake}{nutrient\ need} \text{x } 100\%$$

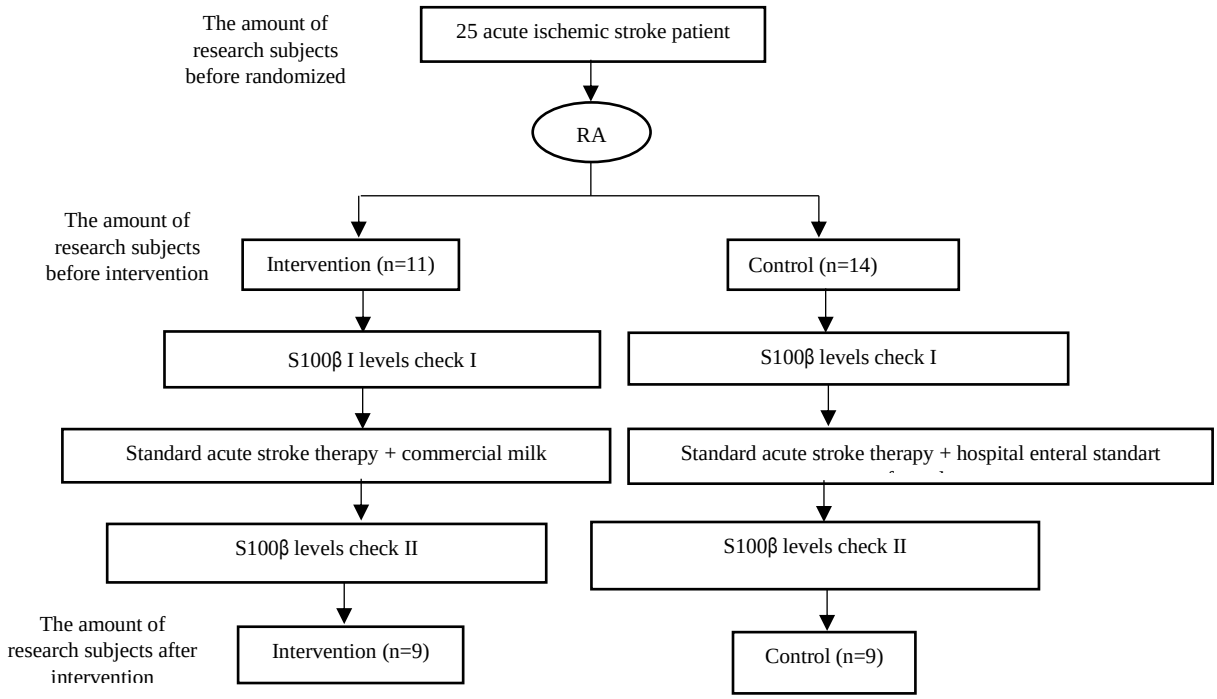

Figure 1. The Flow Chart of the Amount of Research Subjects (Start to Finish)

The nutritional requirement has been adjusted to the nutritional requirement for ischemic stroke patients: 30 kcal/kg BW for energy, 60% TEE for carbohydrate, 20% TEE for protein, 20% TEE for fat, 1000 mg/day for phosphatidylcholine, 100 mg/day for phosphatidylserine, and 12 mg/day for inulin. All data collection has been recorded on the research sheets that had been prepared by researcher.

The control group was given milk based on the enteral standard formula from the hospital that contains 11.8 g protein (from hospital database) without the addition of phosphatidylcholine, phosphatidylserine, and inulin as much as 250 ml.

The intervention group was given 69 g of commercial milk powder which contains higher protein than the control group (15 g) with the addition of 128 mg phosphatidylcholine, 32 mg phosphatidylcholine, and 3 g inulin. The commercial milk given to the intervention group contains a moderate glycemic index, therefore, it is necessary to adjust the dose and the duration of the intervention for subjects with diabetes. In the intervention group, subjects with diabetes mellitus received 34.5 g of commercial milk powder (half the original dose) so the duration of the treatment becomes longer (14 days). Researchers cannot exclude subjects with

diabetes because of the limited number of study subjects. All the treatment was given 6 times a day during the acute phase (1-3 days according to the patient's swallowing ability) by enteral tube and then given orally 3 times a day during the recovery phase until the end of treatment (day 7). During the administration of the intervention, the researchers observed functional and biochemical changes as well as possible side effects.

Whole blood vein samples were collected from the mediana cubiti vein. A sampling of 6 mL was carried out using a red *vacutainer* (blood clot without the addition of EDTA). The duration between blood sampling to storing should not exceed 8 hours. Blood samples were sent to the central laboratory of Dr. Kariadi Central Hospital Semarang for centrifugation and stored in a deep freezer (-80ºC) until the number of research, samples were met. After meeting the sample size, blood serums were sent to the GAKI Laboratory of the RSND-FK UNDIP. Serum S100β levels were checked using the Human S100β ELISA (Enzyme-Linked Immunosorbent Assay) kit Elabscience with catalog number E-EL-H1297. The inspection method was following the manual found in the kit. The analysis of serum S100β levels has been carried out twice; when the patients were admitted to Dr. Kariadi Hospital Semarang before given intervention (day 1) and after given intervention (day 7). This research has received Ethical Clearance approval by the Medical Research Ethics Commission of FK UNDIP / RSDK with registration No. 479 / EC / KEPK-RSDK / 2020.

Statistical analysis was performed using SPSS version 22.0 for windows. Data with categorical scales were expressed in the distribution of frequencies and proportions. The numerical scale data [levels of S100β and changes (Δ) of S100β] were tested for normality and then tested using Paired T-test and Independent T-Test. Also, different levels of S100β and changes (Δ) of S100β in the DM and Non-DM subgroups have been carried out using Paired T-Test/ Independent T-test for normally distributed data and Willcoxon/ Mann Whitney for data that were not normally distributed.

RESULTS

Research Subject Characteristics

There was no significant difference in the category of age (p = 0.347), gender (p = 0.637), mean BMI (p = 0.468), obesity status (p = 1.000), hypertension (p = 0.576), diabetes mellitus (p = 1.000), dyslipidemia (p = 1.000), smoking status (p = 1.000), rt-PA therapy (p = 1.000), inpatient duration (p = 0.637), intervention duration (p = 0.206), and history of recurrent stroke (p = 1.000).

Adequacy Level of Energy, Protein, Fat, and Carbohydrate Intake

The adequacy level was obtained from the total intake per day compared to the actual needs and was presented in percentage. Table 2 shows that there were significant differences in the mean level of adequacy of energy intake (p = 0.008), protein (p = 0.002), carbohydrate (p = 0.002), phosphatidylcholine (p=0.000), phosphatidylserine (p=0.000), and inulin (p=0.000) however there was no significant difference in the mean level of adequacy of fat intake between groups (p = 0.912). For the control group, all adequate levels of nutrient intake were deficits, whereas in the intervention group only adequate levels of intake of fat and phosphatidylcholine were deficits.

Serum S100β Levels Distribution Before and After Interventions

The results of statistical tests showed that there was no significant difference in the distribution of serum S100β levels both before (p = 1.000) and after the intervention (p = 0.576) between the intervention group and the control group. The serum S100β level category <236.7 pg/mL indicates that the patients tended to have a good improvement in clinical outcome. Serum S100β levels> 236.7 pg/mL are associated with poor clinical outcomes and even death. Based on the aforementioned results, one subject in the intervention group before treatment had a high-risk S100β level category, but it went down to the low-risk category after the treatment compared to one subject in the control group who had a low-risk S100β level category before treatment and went up to being categorized as high risk after the treatment. This shows that there was a trend of improvement in S100β levels in the intervention group compared to the control group.

Differences in S100β levels and changes (Δ) of S100β levels

The mean serum S100β levels in baseline of the control and intervention groups (358.8 $\pm$ 215.3 and 512.3 $\pm$ 343.9 pg / mL, respectively) were not statistically significant (p = 0.273). This illustrates that the condition of the two groups before the intervention was homogeneous. In table 4, it is shown that there was a trend of decreasing mean serum S100β levels in the intervention group from 512.3 $\pm$ 343.9 pg / mL to 487.7 $\pm$ 366.8 pg / mL, while in the

control group there was an increasing trend in the mean serum S100β level from 358.8 ± 215.3 pg / mL to 494.6 ± 296.4 pg / mL. Also, the pre and post S100β levels in both the intervention and control groups were not statistically significant (p = 0.777 and p = 0.096, respectively). There was no significant difference in the mean change (Δ) of serum S100β levels in the intervention group (-24.6 ± 252.0 pg / mL) and control group (135.8 ± 216.2 pg / mL), but in the intervention group, the tendency of decline was better than the control group although it was not statistically significant (p = 0.166).

Table 1. The Characteristics of Research Subjects in Research Groups

Characteristic	Group		*p*
	Intervention (n=9)	Control (n=9)	
Gender			
- Male	4 (44.4%)	6 (66.7%)	0.637[b]
- Female	5 (55.6%)	3 (33.3%)	
Mean age (year)	50.6 ± 14.8	62.6 ± 8.8	0.053[a]
Age category			
- >60	3(33.3%)	6(66.7%)	0.347[b]
- <60	6(66.7%)	3(33.3%)	
Mean BMI (kg/m^2)	25.3 ± 4.1	24.1 ± 2.9	0.468[a]
Obesity Status			
- Yes (BMI ≥ 25 kg/m^2)	4 (44.4%)	3 (33.3%)	1.000[b]
- No (BMI < 25 kg/m^2)	5 (55.6%)	6 (66.7%)	
Hipertension			
- Yes (BP ≥ 130/80 mmHg)	8 (88.9%)	6 (66.7%)	0.576[b]
- No (BP < 130/80 mmHg)	1 (11.1%)	3 (33.3%)	
Diabetes Mellitus			
- Yes	3 (33.3%)	4 (44.4%)	1.000[b]
- No	6 (66.7%)	5 (55.6%)	
Dislipidemia			
- Yes	7 (77.8%)	8 (88.9%)	1.000[b]
- No	2 (22.2%)	1 (11.1%)	
Smoking Status			
- Yes	5 (55.6%)	6 (66.7%)	1.000[b]
- No	4 (44.4%)	3 (55.6%)	
rt-PA Therapy			
- Yes	2(22.2%)	1(11.1%)	1.000[b]
- No	7(77.8%)	8(88.9%)	
Inpatient Duration			
- <7 days	6(66.7%)	4(44.4%)	0.637[b]
- ≥7 days	3(33.3%)	5(55.6%)	
Intervention Duration			
- 7 days	6(66.7%)	9(100%)	0.206[b]
- 14 days	3(33.3%)	0(0%)	
History of Recurrent Stroke			
- Yes	3(33.3%)	3(33.3%)	1.000[b]
- No	6(66.7%)	6(66.7%)	

[a] Independent T-Test; [b] Fisher-Exact Test
The values in table are: mean ± SD; n (%) Percentage shown in columns

Table 2. The Mean Adequacy Intake of Energy, Protein, Fat, and Carbohydrate between Groups

Adequacy Levels	Intervention Mean±SD	Control Mean±SD	*p*
Energy (%)	95.1±18.4	73.8±10.1	0.008[a]
Carbohydrate (%)	107.0±20.6	76.6±12.2	0.002[a]
Protein (%)	94.2±17.6	68.5±10.9	0.002[a]
Fat (%)	74.3±21.2	73.4±11.2	0.912[a]
Fosfatidilkolin (%)	72.7±8.1	25.3±3.4	0.000[a]
Fosfatidilserin (%)	112.4±12.3	48.7±10.3	0.000[a]
Inulin (%)	92.4±8.0	24.4±7.3	0.000[a]

[a] Independent T-Test

Table 3. The Distribution of S100β Levels before Intervention in Research Groups (n=18)

	Category of Serum S100β Levels (pg/mL)	Group		*p*
		Intervention (n=9)	Control (n=9)	
Before Intervention	Low risk (<236.7)	2(22.2%)	2(22.2%)	1.000[b]
	High risk (≥236.7)	7(77.8%)	7(77.8%)	
After Intervention	Low risk (<236.7)	3(33.3%)	1(11.1%)	0.576[b]
	High risk (≥236.7)	6(66.7%)	8(88.9%)	

[b]Fisher Exact Test

Table 4. The Difference in Serum S100β Levels (pg/mL)

Group	n	Before Mean±SD	After Mean±SD	*P*	Δ Mean±SD
Intervention	9	512.3±343.9	487.7±366.8	0.777[c]	-24.6±252.0
Control	9	358.8±215.3	494.6±296.4	0.096[c]	135.8±216.2
p		0.273[a]	0.965[a]		0.166[a]

[a]Independent T-Test; [c] *Paired T-Test*

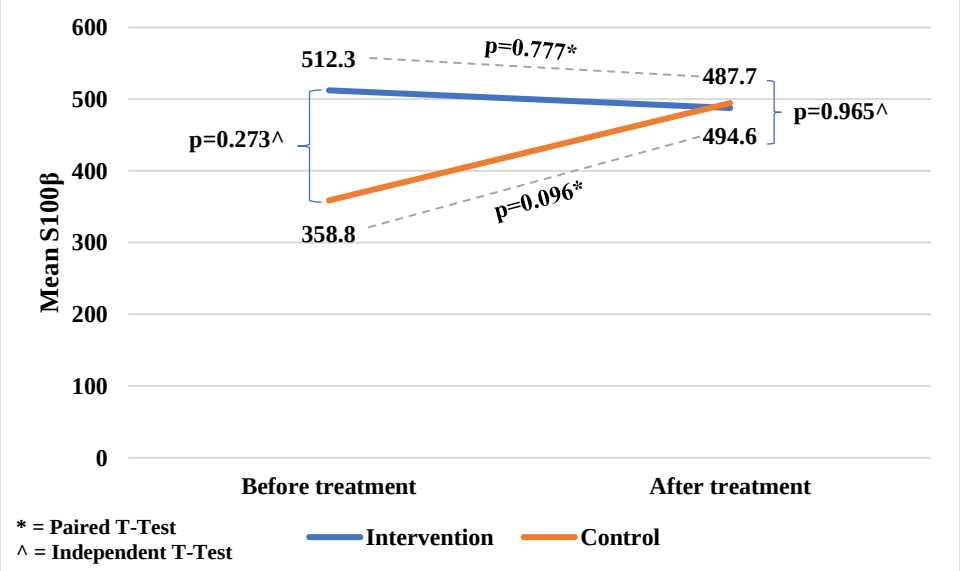

Figure 2. The Serum S100β Levels before and after Intervention

Sub-group analysis of S100β levels

In DM and non-DM sub-groups, the median serum S100β levels before treatment between the intervention and control groups were not statistically significant (p = 0.480 and p = 0.251, respectively). This illustrates that the condition of the two sub-groups before the intervention was homogeneous. There was no significant difference in the median

levels of S100β before and after the intervention in both DM (p intervention = 0.285; p control = 0.272) and non-DM sub-group (p intervention = 0.484; p control = 0.308).

In DM and Non-DM sub-groups, there was a trend of decreasing S100β levels in a better direction for the intervention group and the decrease (Δ) in the DM sub-group was bigger than Non-DM sub-group compared to the control group where there was an increasing trend (Δ) in S100β levels in DM and Non-DM sub-group and the increase (Δ) in DM sub-group was bigger than Non-DM sub-group.

DISCUSSION

The data of research subjects showed no significant difference in mean BMI, mean age, age category, gender, obesity, hypertension, diabetes mellitus, dyslipidemia, smoking status, rt-PA therapy, inpatient duration, intervention duration, and history of recurrent stroke (p>0.05).

Table 5. The *Sub Group* Analysis of S100β Levels between Groups

Sub Grup	Kelompok	n	Sebelum Median(Min-Max)	Sesudah Median(Min-Max)	*p*	Δ Median(Min-Max)
DM	Intervention	3	971.9(115.5-977.9)	521.8(146.6-771.8)	0.285[c]	-200.1(-456.1 – 31.1)
	Control	4	428.8(252.1-794.2)	641.0(253.4-1015.9)	0.272[d]	117.8(-83.1 – 494.9)
	P		0.480[e]	0.589[a]		0.102[a]
Non-DM	Intervention	6	475.1(28.5-660.7)	392.7(25.6-1004.8)	0.484[c]	-39.9(-117.5 – 344.5)
	Control	5	255.9(107.1-426.5)	417.8(187.7-610.9)	0.308[c]	75.0(-178.8 – 355.0)
	P		0.251[a]	0.593[a]		0.584[e]

[a] Independent T-Test; [c] Paired T-Test; [d] Willcoxon Test; [e] Mann Whitney Test

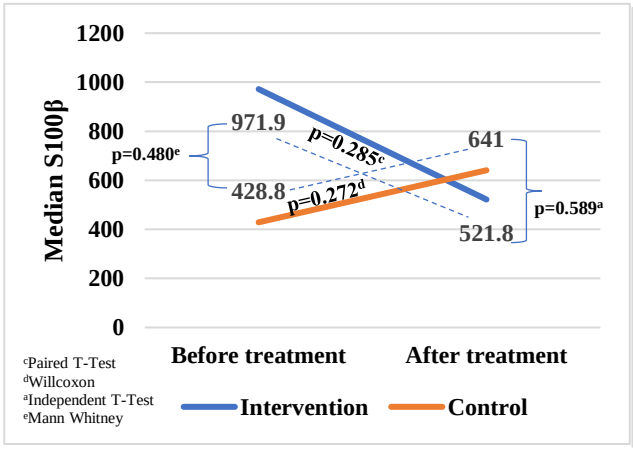

Figure 3. The S100β Levels before and after intervention in DM Sub Group

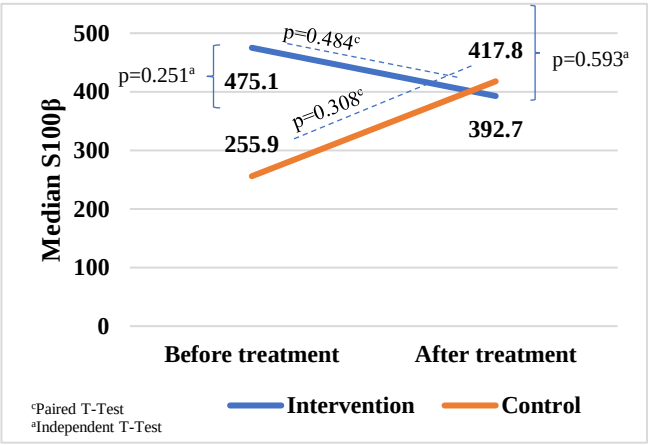

Figure 4. The S100β Levels before and after intervention in Non-DM Sub Group

Stroke does not only occur in the elderly, but also occurs in those at productive age under 45 years, and there are some cases where the sufferers are under 30 years old.[27,28] In this study, 2 subjects were under 30 years old. The habit of consuming food containing high fat and smoking has been shown to influence the incidence of ischemic stroke at an age less than 45 years.[29] People who consume high-fat foods ≥3 times/week have a 3.744 times more risk of suffering from ischemic stroke compared to people who consume high-fat foods <3 times/week.[29] High-fat food sources when consumed excessively can increase cholesterol levels in the blood. If the cholesterol level in the blood is abnormal (≥ 200 mg / dL) it will increase the accumulation of fat on the walls of blood vessels, causing narrowing of the blood vessels and disrupting the blood supply to the brain.[30] Cholesterol can form blood clots and plaques that clog the arteries and eventually cut off the blood flow to the heart and brain. This can lead to heart attacks and strokes.[31] Smoking is associated with a hypercoagulable state. It is characterized by an increase in thromboxane release which causes increased platelet activation and degeneration of the vascular endothelium, which promotes the formation of thrombus-producing plaque. Nicotine has no direct

effect on this mechanism, but it is possible that the burning of cigarette smoke causes thromboembolic formation, leading to ischemic stroke.[32]

The data on the adequacy of nutritional intake showed that there were significant differences in levels of the adequacy of energy, carbohydrate, and protein between groups ($p < 0.05$). There was no difference in the level of adequacy of fat between groups ($p > 0.05$). The nutritional content of commercial milk given to the intervention group had higher energy, carbohydrate, and protein content (Energy = 289.8 kcal, Protein = 15.18 grams, KH = 44.16 grams per 250 mL) compared to milk given to the control group (Energy = 222.2 - 246.4 kcal, Protein = 11 - 11.8 grams, CH = 28 - 29.2 grams per 250 mL), while milk given to the control group had higher fat content (9 gram / 250 mL) compared to the milk given to the intervention group (5.52 g / 250 mL). This was also influenced by nutritional requirement acceptance of each respondent, method of administration (enteral tube for acute phase and then orally for recovery phase), the role of medical team, especially nurses, and family members in motivating patients to eat, as well as the length of hospitalization of patients.

Also, there were six subjects (66.7%) in the intervention group and four subjects (44.4%) in the control group who returned home before the 7th day. Patients in both control and intervention groups who returned home before the 7th day still had poor appetite, but patients in the intervention group have received the commercial milk to be consumed at home while the control group did not. This led to a higher level of energy, protein, and carbohydrate intake in the intervention group even though the patient did not have a good appetite. Also, patients who returned home before the 7^{th} day tended to have an increase in fat intake based on the results of the recall of intake by telephone. They tended to eat food that contains high fat rather than at the hospital, where the menu has been adjusted with the requirements and conditions of each respondent.

The results of statistical tests showed that there was no significant difference in the mean serum S100β levels between before and after the intervention or between groups. The causes of this insignificant difference were multifactorial, one of which was that the subjects in this research have different comorbidities such as hypertension, diabetes mellitus, obesity, and dyslipidemia. According to research, S100β levels in subjects who have comorbid like diabetes mellitus, hypertension, central obesity, and dyslipidemia will tend to be increased.[33,34] Two subjects in the intervention group and one subject in the control group in this research received rt-PA therapy. Some research proved the effectiveness of citicoline as a neuroprotective therapy, showing that rt-PA could improve blood flow in the penumbra area. This leads to becoming soluble and having difficulty improving its performance due to the effects of thrombolysis. Also, the higher the severity of the stroke will affect the performance of citicoline because the penumbra areas that are damaged are not fully reached by citicoline.[12,35] The results of the difference test (Δ) S100β levels in this research were not statistically significant ($p = 0.166$) between groups. This shows that rt-PA therapy does not affect the intervention given. Another research also stated that rt-PA did not significantly affect S100β levels.[36] The reason was that the response of each individual to rt-PA therapy was different. rt-PA therapy can exert a significant neuroprotective effect only when thrombolysis and reperfusion are achieved earlier, whereas in cases where reperfusion is not immediately achieved there may be side effects such as blood-brain barrier (BBB) disruption and hemorrhagic transformation.[36]

A study shows that there is a positive and significant correlation between age and S100β levels in study subjects with mild TBI (Traumatic Brain Injury). The study reported that the subjects aged >65 years had higher levels of S100β (> 0.10 μg / L) compared to subjects aged <65 years. This is because older subjects usually have chronic diseases or neurological diseases such as Alzheimer's and Parkinson's, but further research is needed to explain this association.[37] In this study, the mean age of subjects in the control group was older (62.6 ± 8.8) than in the intervention group (50.6 ± 14.8). This is one of the possible causes of S100β levels in the control group which tended to increase compared to the intervention group.

Research on the effect of giving protein, neuroprotective substances (phosphatidylcholine and phosphatidylserine), and inulin on brain damage markers, especially S100β, is still very limited. In this research, the interventions were administered orally in the form of powdered milk containing neuroprotectants (phosphatidylcholine and phosphatidylserine) and prebiotic inulin. In oral administration, phosphatidylcholine and phosphatidylserine are almost completely absorbed efficiently. Once absorbed, phosphatidylcholine and phosphatidylserine spread widely throughout the body through the blood-brain barrier and reach the central nervous system (CNS).[38,39] Inulin is soluble in

water and difficult to be hydrolyzed by enzymes in the digestive tract, thus it can reach the large intestine mostly intact. In the large intestine, inulin will be fermented by probiotic bacteria into short-chain fatty acids (SCFA) and some specific microflora that produces lactic acid.[40]

The results of different tests for S100β levels were not statistically significant, but in the intervention group, there was a trend of decreasing S100β levels compared to the control group. According to research, a high-protein diet given from the beginning provides regulation at the molecular level, which helps to increase SOD expression, suppresses MDA expression, and restores iNOS expression.[41] This suggests that a high-protein diet given from the beginning can inhibit oxidative stress, thereby helping the body to eliminate free radicals that play a role in the pathophysiological process of stroke.[41]

Vitamin B supplementation (folic acid, riboflavin, vitamin B12) and choline are effective in improving stroke recovery by suppressing the transcription factor p53 mechanism which causes apoptosis (death of nerve cells) where p53 activation is triggered by an increase in homocysteine. Also, there is an increase in neuroplasticity through increased BDNF (Brain-derived neurotrophic factor) expression and activation of anti-oxidative mechanisms characterized by increased levels of the transcription factor Nrf2 (nuclear factor erythroid 2-related factor 2) and SOD2 (Superoxide dismutase 2) enzyme. This means an increase in the flexibility of cells to manage oxidative damage.[42]

Several kinds of research on the effects of phosphatidylserine supplementation in experimental animals and humans have been carried out on the improvement of cognitive and memory functions by largely unexplainable mechanisms. In essence, phosphatidylserine plays a role in the activation of signaling proteins and receptors that are important for neuron survival, differentiation, and regulation of neurotransmitters in the signaling process.[39]

Inulin is an example of a prebiotic. Prebiotics are components of food ingredients that the digestive tract cannot enzymatically digest. Thus, they will be fermented by microbiota in the large intestine. Inulin acts as a substrate to increase the diversity of beneficial microbiota in the intestine, increasing the production of SCFA (acetate, propionate, and butyrate) as a result of anaerobic fermentation.[43] SCFA can affect the gut-brain axis and brain function directly or indirectly. SCFAs are absorbed by colonic cells via H^+-dependent monocarboxylate transporter (MCT) or Na^+-dependent monocarboxylate transporter (SMCTs). SCFA then binds to G protein-coupled receptors (GPCRs) such as FFAR2, FFAR3, GPR109a / HCAR2 (hydrocarboxylic acid receptors), and GPR164 by inhibiting histone deacetylation that will affect intestinal mucosal immunity and intestinal barrier integrity. SCFA interaction with some of these receptors on enteroendocrine cells will cause indirect signaling to the brain through the systemic circulation by inducing the secretion of intestinal hormones such as glucagon-like peptide 1 (GLP1), peptide YY (PYY), γ-aminobutyric acid (GABA), and serotonin (5-HT). Colon-generated SCFAs reaches the systemic circulation and other tissues leading to activation of brown adipose tissue, regulation of mitochondrial function in the liver, increased insulin secretion by β-pancreatic cells, and energy homeostasis throughout the body. Systemically, SCFAs influence systemic inflammation by inducing differentiation of T regulatory (Treg) cells and regulating interleukin secretion. SCFA can cross the blood-brain barrier via monocarboxylate transporter (MCT) located on endothelial cells and improve the integrity of the blood-brain barrier by increasing the expression of tight junction proteins. Furthermore, in the central nervous system (CNS), SCFA fixes inflammation in the nerves by improving glial cell function and morphology, modulating levels of neurotrophic factors, increasing neurogenesis, contributing to serotonin biosynthesis, and increasing the function and homeostasis of nerve cells. SCFA interactions with the gut-brain axis can directly or indirectly affect the emotion, cognition, and pathophysiology of brain disorders.[44]

Since researchers can not exclude subjects with diabetes mellitus, then subgroup analysis was done. The results of DM and Non-DM sub-groups analysis showed that in DM and Non-DM sub-groups there was a trend of decreasing S100β levels in a better direction for the intervention group and a decrease (Δ) in the DM subgroup was bigger than Non-DM subgroup compared to a control group where there were a trend of increasing (Δ) levels of S100β in the DM and Non-DM subgroups and an increase (Δ) in DM subgroup was bigger than Non-DM subgroup. This reason was that in the DM subgroup the intervention duration was different from the Non-DM subgroup, especially in the intervention group. Milk given to the intervention group had a rather high glycemic index, so it was necessary to adjust the daily dose in patients with comorbid DM. This resulted in the intervention duration that was originally given for 7 days stretched to 14 days.

According to research, the pattern of S100β levels will increase from 0-2 hours of onset to 24-48 hours of onset (peak level) then S100β levels will decrease up to 2-3 weeks after onset.[45] This causes a decrease in (Δ) levels of S100β bigger in the DM subgroup compared to the Non-DM sub-group.

The limitation of this research was the number of samples which was 18 subjects (intervention = 9; control = 9). Dr. Kariadi Central Hospital is a referral hospital for COVID-19 patients, so the hospital limits the number of patients at the polyclinic every day. Also, many people are afraid of going to the hospital. Another reason might be the dose and duration of intervention. In some research, the minimum duration of 14 days[38,41,43,46,47] and a minimum dose that can provide a better clinical outcome is 1000 mg/day for phosphatidylcholine.[48,49] In this study, the mean phosphatidylcholine intake in the intervention group is still a deficit. The subjects receive the treatment by enteral tube in the acute phase (1-3 days after stroke onset) followed by oral administration. For administration through the enteral tube, it will be easier to ensure that the treatment is completed, whereas when given orally, some patients do not cooperate due to several factors such as the imperfect swallowing ability and the level of consciousness is still weak. Also, there were subjects in both the control and intervention group who returned home before the 7th day of treatment because some patients ask to continue the recovery phase at home after they have passed the acute period. This has to lead the researchers to recall those subjects regarding their intake by telephone. As a result, patients tend to forget about what they have consumed, also the (household size used by those subjects was different so that the recall results could be underestimating/overestimate. In the hospital, portion standards are already specified for each type of diet.

CONCLUSIONS

The addition of 15 g protein, 128 mg phosphatidylcholine, 32 mg phosphatidylserine, and 3 g inulin, three times a day, for seven days, in patients with acute ischemic stroke does not have a significant effect on S100β levels statistically, however, there is a decreasing trend of S100β levels for the intervention group.

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REFERENCES

1. Xing C, Arai K, Lo EH, Hommel M. Pathophysiologic cascades in ischemic stroke. Int J Stroke. 2012;7(5):378–85.
2. Guo Y, Li P, Guo Q, Shang K, Yan D, Du S, et al. Pathophysiology and biomarkers in acute ischemic stroke - A review. Trop J Pharm Res. 2013;12(6):1097–105.
3. Rezaei O, Pakdaman H, Gharehgozli K, Simani L, Vahedian-Azimi A, Asaadi S, et al. S100 B: A new concept in neurocritical care. Iran J Neurol. 2017;16(2):83–9.
4. Secades JJ. Citicoline: pharmacological and clinical review, 2016 update. Rev Neurol. 2016;63(S03): S1–73.
5. Álvarez-Sabín J, Román GC. The role of citicoline in neuroprotection and neuro repair in ischemic stroke. Brain Sci. 2013;3:1395–414.
6. Kadry H, Noorani B, Cucullo L. A blood-brain barrier overview on structure, function, impairment, and biomarkers of integrity. Fluids Barriers CNS [Internet]. 2020;17(1):1–24. Available from: https://doi.org/10.1186/s12987-020-00230-3
7. Kasim VN, Pateda SM, Hadju V, Jafar N. Suplementasi ekstrak albumin ikan gabus terhadap status gizi dan imunitas pasien stroke. J Gizi Klin Indones. 2017;13(3):91.
8. Singh V, Roth S, Llovera G, Sadler R, Garzetti D, Stecher B, et al. Microbiota dysbiosis controls the neuroinflammatory response after stroke. J Neurosci. 2016;36(28):7428–40.
9. Hennezel E, Abubucker S, Murphy LO, Cullen TW. Total Lipopolysaccharide from the. MBio. 2017;2(6):1–12.
10. Hotamisligil GS. Inflammation and metabolic disorders. 2019;7121(December):1–17.
11. Li H, Li H, Zhang X, Zhang X, Pan D, Pan D, et al. Dysbiosis characteristics of gut microbiota in cerebral infarction patients. Transl Neurosci. 2020;11(1):124–33.
12. Overgaard K. The effects of citicoline on acute ischemic stroke: A review. J Stroke Cerebrovasc Dis. 2014;23(7):1764–9.
13. Adibhatla RM. Citicoline in stroke and TBI clinical trials. Nat Rev Neurol. 2013;9(3):173.
14. Presley B. Penatalaksanaan Farmakologi Stroke Iskemik Akut. Buletin Rasional [Internet]. 2013;12(1):6–8. Available from:

http://repository.ubaya.ac.id/21378/1/Rasional Vol 12 No 1.pdf
15. Lansberg MG, O'Donnell MJ, Khatri P, Lang ES, Nguyen-Huynh MN, Schwartz NE, et al. Antithrombotic and thrombolytic therapy for ischemic stroke: Antithrombotic Therapy and Prevention of Thrombosis, 9th ed: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines. Chest. 2012;141(2 Suppl):e601S-e636S.
16. Jauch EC, Saver JL, Adams HP, Bruno A, Connors JJB, Demaerschalk BM, et al. Guidelines for the early management of patients with acute ischemic stroke: A guideline for healthcare professionals from the American Heart Association/American Stroke Association. Stroke. 2013;44(3):870–947.
17. Sahota P, Savitz SI. Investigational Therapies for Ischemic Stroke: Neuroprotection and Neuro recovery. Neurotherapeutics. 2011;8(3):434–51.
18. Zanini B, Simonetto A, Zubani M, Castellano M, Gilioli G. The effects of cow-milk protein supplementation in the elderly population: Systematic review and narrative synthesis. Nutrients. 2020;12(9):1–26.
19. Patel S. Functional food relevance of whey protein: A review of recent findings and scopes ahead. J Funct Foods [Internet]. 2015;19:308–19. Available from: http://dx.doi.org/10.1016/j.jff.2015.09.040
20. Mitchell CJ, McGregor RA, D'Souza RF, Thorstensen EB, Markworth JF, Fanning AC, et al. Consumption of milk protein or whey protein results in a similar increase in muscle protein synthesis in middle-aged men. Nutrients. 2015;7(10):8685–99.
21. Birkeland E, Gharagozlian S, Birkeland KI, Valeur J, Måge I, Rud I, et al. Prebiotic effect of inulin-type fructans on faecal microbiota and short-chain fatty acids in type 2 diabetes: a randomized controlled trial. Eur J Nutr [Internet]. 2020;59(7):3325–38. Available from: https://doi.org/10.1007/s00394-020-02282-5
22. Shoaib M, Shehzad A, Omar M, Rakha A, Raza H, Sharif HR, et al. Inulin: Properties, health benefits, and food applications. Carbohydr Polym [Internet]. 2016;147(October 2017):444–54. Available from: http://dx.doi.org/10.1016/j.carbpol.2016.04.020
23. Sherwin E, Sandhu K V., Dinan TG, Cryan JF. May the Force Be With You: The Light and Dark Sides of the Microbiota–Gut-Brain Axis in Neuropsychiatry. CNS Drugs. 2016;30(11):1019–41.
24. Braniste V, Al-Asmakh M, Kowal C, Anuar F, Abbaspour A, Tóth M, et al. The gut microbiota influences blood-brain barrier permeability in mice. Sci Transl Med. 2014;6(263):1–12.
25. Erny D, De Angelis ALH, Jaitin D, Wieghofer P, Staszewski O, David E, et al. Host microbiota constantly control maturation and function of microglia in the CNS. Nat Neurosci. 2015;18(7):965–77.
26. Arbayanto DN. Penyembuhan Stroke Perlu Kecepatan Waktu [Internet]. Ruang Publik. 2018. Available from: rri.co.id
27. Ghani L, Mihardja LK, Delima. Faktor Risiko Dominan Penderita Stroke di Indonesia. Bul Penelit Kesehat. 2016;44(1):49–58.
28. Laily SR. Hubungan Karakteristik Penderita dan Hipertensi dengan Kejadian Stroke Iskemik. J Berk Epidemologi. 2017;5(1):95–106.
29. Iskandar A, Hadisaputro S, Pudjonarko D, Suhartono, Pramukarso DT. Gaya Hidup Yang Berpengaruh Terhadap Kejadian Stroke Iskemik Pada Usia Kurang Dari 45 Tahun (Studi Di BLUD RSUD Cut Nyak Dhien Meulaboh Kabupaten Aceh Barat Provinsi Aceh). J Epidemiol Kesehat Komunitas. 2018;3(2):54–62.
30. Junaidi I. Stroke : Waspadai Ancamannya. Yogyakarta: Andi; 2011.
31. Mohr J, Wolf P, Moskowitz M, Mayberg M, Kummer R Von. Stroke: Pathophysiology, Diagnosis, and Management. 5th ed. Grotta J, editor. Philadelphia: Elsevier Saunders; 2011. 198–218 p.
32. Matsuo R, Ago T, Kiyuna F, Sato N, Nakamura K, Kuroda J, et al. Smoking status and functional outcomes after acute ischemic stroke. Stroke. 2020;846–52.
33. Lotosh NG, Savel'Eva EK, Selishcheva AA, Savel'Ev S V. Autoantibodies to neuron-specific proteins S100, GFAP, MBP, and NGF in the serum of rats with streptozotocin-induced diabetes. Bull Exp Biol Med. 2013;155(1):48–51.
34. Kheirouri S, Ebrahimi E, Alizadeh M. Association of S100B Serum Levels with

Metabolic Syndrome and its Components. 2018;31(4):201–6.
35. Dávalos A, Alvarez-Sabín J, Castillo J, Díez-Tejedor E, Ferro J, Martínez-Vila E, et al. Citicoline in the treatment of acute ischaemic stroke: An international, randomised, multicentre, placebo-controlled study (ICTUS trial). Lancet [Internet]. 2012;380(9839):349–57. Available from: http://dx.doi.org/10.1016/S0140-6736(12)60813-7
36. Woods SD, Flores R, Roberson PK, Lowery JD, Skinner RD, Culp WC. Decreased Serum Levels of S-100B Protein Reflect Successful Treatment Effects in a Rabbit Model of Acute Ischemic Stroke. Open Neurol J. 2011;5(1):55–7.
37. Calcagnile O, Holmén A, Chew M, Undén J. S100B levels are affected by older age but not by alcohol intoxication following mild traumatic brain injury. Scand J Trauma Resusc Emerg Med. 2013;21(1):2–7.
38. Wahyudi R, Hasmono D, Fitrina R, Armal K. Injected Citicoline Improves Impairment and Disability During Acute Phase Treatment in Ischemic Stroke Patients. Folia Medica Indones. 2016;51(4):245.
39. Glade MJ, Smith K. Phosphatidylserine and the human brain. Nutrition [Internet]. 2015;31(6):781–6. Available from: http://dx.doi.org/10.1016/j.nut.2014.10.014
40. Setiarto R, Widhyastuti N, Saskiawan I, Safitri R. Pengaruh Variasi Konsentrasi Inulin Pada Proses Fermentasi Oleh L. Acidophilus, L. Bulgaricus Dan S. Thermophillus. Biopropal Ind. 2017;8(1):1–17.
41. Ji M, Li S, Dong Q, Hu W. Impact of an early high-protein diet on neurofunctional recovery in rats with ischemic stroke. Med Sci Monit. 2018;24:2235–43.
42. Jadavji NM, Emmerson JT, MacFarlane AJ, Willmore WG, Smith PD. B-vitamin and choline supplementation increases neuroplasticity and recovery after stroke. Neurobiol Dis. 2017;103:89–100.
43. Hoffman JD, Yanckello LM, Chlipala G, Hammond TC, McCulloch SD, Parikh I, et al. Dietary inulin alters the gut microbiome, enhances systemic metabolism, and reduces neuroinflammation in an APOE4 mouse model. PLoS One. 2019;14(8):1–22.
44. Silva YP, Bernardi A, Frozza RL. The Role of Short-Chain Fatty Acids From Gut Microbiota in Gut-Brain Communication. Front Endocrinol (Lausanne). 2020;11:1–14.
45. Thelin EP, Nelson DW, Bellander BM. A review of the clinical utility of serum S100B protein levels in the assessment of traumatic brain injury. Acta Neurochir (Wien) [Internet]. 2017;159(2):209–25. Available from: http://dx.doi.org/10.1007/s00701-016-3046-3
46. Aquilani R, Sessarego P, Iadarola P, Barbieri A, Boschi F. Nutrition for brain recovery after ischemic stroke: An added value to rehabilitation. Nutr Clin Pract. 2011;26(3):339–45.
47. Richter Y, Herzog Y, Lifshitz Y, Hayun R, Zchut S. The effect of soybean-derived phosphatidylserine on cognitive performance in elderly with subjective memory complaints: A pilot study. Clin Interv Aging. 2013;8:557–63.
48. Martynov MY, Boiko AN, Kamchatnov PR, Kabanov AA, Yasamanova AN, Shchukin IA, et al. Neuroprotective therapy with citicoline (Ceraxon) in patients with ischemic stroke. Neurosci Behav Physiol. 2013;43(6):706–11.
49. Kato-Kataoka A, Sakai M, Ebina R, Nonaka C, Asano T, Miyamori T. Soybean-derived phosphatidylserine improves memory function of the elderly Japanese subjects with memory complaints. J Clin Biochem Nutr. 2010;47(3):246–55.

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The effect of addition protein, phosphatidylcholine, phosphatidylserine, and inulin on GFAP levels of acute ischemic stroke patients at Dr. Kariadi Hospital, Semarang

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ABSTRACT

Background: The occurrence of ischemia causes a loss of energy to switch to anaerobic processes resulting in acidosis due to reduced Adenosina Triphosphate (ATP). This condition makes neuron cells apoptotic. Apoptotic of several biochemical substrates in the brain, such as Glial Fibrillary Acidic Protein (GFAP) exit into the circulatory system which is associated with dysbiosis through immunological pathways.
Objectives: To determine the effect of giving enteral formula containing protein, phosphatidylcholine, phosphatidylserine, and inulin on GFAP levels in patients with acute ischemic stroke Dr. Kariadi Hospital.
Materials and Methods: This study was done in a single-blind RCT. Eighteen ischemic stroke patients were randomly divided into intervention (9 subjects) and control groups (9 subjects). The intervention group received 69 g of the powdered enteral formula three times a day for seven days. The formula contained protein (15 g), phosphatidylcholine (128 mg), phosphatidylserine (32 mg), and inulin (3 g). The subject who had diabetes mellitus received for 14 days at a dose of 34.5 g per day (7.5 g protein with additions 64mg phosphatidylcholine, 16mg phosphatidylserine, 1.5 g inulin). The control group received the standard enteral formula from the hospital, which contains (11.8 g protein without additions protein, phosphatidylcholine, phosphatidylserine, and inulin). GFAP levels by ELISA method (Enzyme-linked immunosorbent Assay) at pre and post-intervention.
Results: There was a trend of decreasing GFAP levels before and after in the intervention group towards a better direction from 8.37±4.25 to 8.30±4.9 compared with the control group which experienced an increasing trend from 5.4±1.8 to 7.5±4. There was no significant difference in GFAP levels after intervention between groups (p = 0.7).
Conclusions: The addition of protein, phosphatidylcholine, phosphatidylserine, and inulin had no significant effect on GFAP levels.

Keyword: GFAP; Protein; Phosphatidylcholine; Phosphatidylserine; Inulin

BACKGROUND

Ischemic stroke occurs when blood supply to the brain is interrupted or reduced due to a blockage. The occurrence of ischemia in cells and reduced glucose supply causes an increased glutamate expression. Glucose metabolism later switches to anaerobic processes so that it becomes acidosis due to reduced ATP. This situation prompts apoptosis or the death of nerve cells in the brain. In the event of apoptosis, some biochemical substrates may leave the brain and leak off into the circulatory system. In addition to apoptosis, there could be a reduction of proteins that play a role in maintaining the integrity of the blood-brain barrier membrane; hence membrane will encounter permeability. This results in GFAP which should be in the brain structure going out into the circulatory system.[1–4]

Glial Fibrillary Acidic Protein (GFAP) is one of the intermediate protein families, including vimentin, nestin, and is known as an intermediate filament (IF) which is a group of cell types found in the central nervous system and is responsible for maintaining the structure and migration of astroglia cells. GFAP plays a role in the mitotic process by adjusting the filamentous tissue in

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cells. Besides, GFAP also plays a role in neuron astrocyte interactions and communication between cells. Under physiological conditions, GFAP is not actively secreted by cells and is generally undetectable in the blood of healthy individuals. In a moment of brain injury, astroglia responds by producing more GFAP, which in turn will be released into the Central Nervous System (CNS) fluid and blood. GFAP is exclusively produced by astrocytes so that this protein is particularly observed in the brain. Examinations have shown that GFAP is released rapidly in cases of acute ischemic stroke, wherein levels peak within 12 hours of onset and are maintained for up to 72 hours of onset.[5]

A study by Dvorak et al which stated that serum GFAP has taken within 2-6 hours after stroke onset was significant in differentiating hemorrhagic stroke from ischemic stroke. GFAP reaches its highest level within 12 hours of stroke onset.[6] Supported by research by Puspitasari et al which states there is a significant correlation between GFAP serum levels with stroke severity scale after 1 month of stroke onset.[7] This is different from the study conducted by Neila et al, which aims to determine the difference between blood sampling time and serum GFAP levels within 24 hours of onset in patients with ischemic stroke and hemorrhagic stroke, which shows that there was no significant difference in GFAP levels in samples <6 hours, 6-12 hours, and 12-24 hours in ischemic strokes and ICH strokes.[8]

Factors that can influence GFAP include some cellular processes. In general, GFAP has an important role in the regulation between astrocytes and neurons. Changes in GFAP expression can affect synaptic function, interfere with glutamate metabolism, increase the volume of astrocytes that can expand to the surface area of neurons in contact with the astrocyte membrane, and hypoxia.[4]

Thrombolysis or *recombinant tissue plasminogen activator (*rTPA*)* may be one of his definitive therapies. However, this therapy cannot be given to all ischemic stroke patients. Given the strictness of criteria such as the time window, the time for rTPA therapy was <3 hours and a range of 3-4.5 hours after symptom onset. If the therapy is not performed according to established guidelines, one may experience a more severe risk, such as bleeding in angioedema, intracranial and gastrointestinal tract.[9] The limitations of the requirement for rTPA have made many pharmacological therapies emerged, such as neuroprotective therapies in which the addition of phospholipids given in supplement form (citicoline). Some of the most abundant types of phospholipids are phosphatidylcholine and phosphatidylserine. Phosphatidylcholine can improve the biosynthesis of membrane phospholipids which are deteriorated by an increment in free radicals during brain ischemia (neuroprotection). Besides that, the activation of the enzymes that trigger apoptosis in nerve cells will be inhibited by phosphatidylcholine so that it will affect GFAP levels.[10] Research on the effect of giving protein, neuroprotective substances (phosphatidylcholine and phosphatidylserine), and inulin on markers of brain damage, especially GFAP, is still very limited.

Animal studies with the administration of the drug cytoline have the potential to reduce acute brain damage and improve functional recovery, even when given several hours after an ischemic event because the phosphatidylcholine in cytoline can increase the biosynthesis of membrane phospholipids degraded by increased free radicals during brain ischemia (neuroprotection). Besides, cytoline has been shown to restore mitochondrial ATPase and Na1 / K1 ATPase activity to inhibit phospholipase A2 activation which triggers apoptosis of nerve cells.[11,12] so that will affect the levels of GFAP.

A Japanese study was conducted to investigate the effect of phosphatidylserine extracted from soybeans (Soy-PS) on the cognitive function of an elderly group with memory complaints. Oral administration of Soy-PS improves memory function, especially in the elderly with memory complaints. These effects were similarly observed at low doses (100 mg/day) and high doses (300 mg/day).[13]

Fulfillment of protein intake in stroke patients is often given enteral formula with high calories high protein diet which operates to preserve adequate nutritional intake to speed up the recovery period as the enteral formula contains high protein. Dysphagia, unconsciousness (coma), or what is known as enteral feeding is one alternative to meet the nutritional needs of stroke patients by providing nutrients in the form of enteral formula. The protein content in the enteral formula can improve body metabolism through appetite regulation and/or other mechanisms that can control body weight and body composition.[14] Several studies have proven the benefits of enteral formula protein in increasing protein consumption, muscle mass and possibly

increasing body mass index so that it can improve the motor function abilities of patients.[15–17]

Prebiotics is one of the nutrients needed in stroke patients in addition to phospholipids and protein. One type of which is named inulin. Prebiotic bacteria such as Bifidobacteria and Lactobacilli will ferment inulin to produce Short-Chain Fatty Acid (SCFA) and its fermented products in the form of propionate, butyrate, acetic acid, L-lactate, CO2, and hydrogen. Directly or indirectly, the gut-brain axis is influenced by SCFA. The blood-brain barrier can be penetrated by SCFA mediated by monocarboxylate transporter (MCT) and improves the integrity of the blood-brain barrier by increasing the expression of tight junction proteins. SCFA will improve inflammation in nerves by modulating levels of neurotrophic factors, improving morphology and glial cell function, and strengthening neurogenesis.[18] Previous research conducted by Liu et al explained that administering 10% inulin for 4 weeks can change the composition of the gut microbiota and improve gut function in obese mice.[19] The same thing was done by Chuncai et al which stated that the administration of NAC, inulin, and a combined therapy improved cognition in castrated rats. RX-fed rats treated with either the vehicle or testosterone had a significantly increased number of GFAP positive cells. This increment was attenuated by treatment with NAC, inulin, or the combined therapy.[20]

Enteral nutrition has been shown to reduce infectious complications and improve outcomes in patients with severe TBI. Increasing immune cell activity and the integrity of the intestinal barrier, most likely preventing immune dysfunction and bacterial translocation, are the benefits of early feeding, respectively. The nervous system and endocrine changes following enteral feeding can help reduce massive inflammation through vagus nerve-mediated mechanisms. In a journal published by Luyer in Bansal et al, it is shown that enteral feeding, especially dietary fat, stimulates the release of cholecystokinin and binds to cholecystokinin receptors which ultimately leads to activation of the efferent vagal system. This activation triggers an increase in acetylcholine at the gut synaptic level and decreases TNF-α production by way of acetylcholine binding to α-7 nicotinic receptors on macrophages and other immune cells. Therefore, the positive immunologic benefits of early enteral feeding after TBI may also be a result of decreased inflammatory cytokine production by way of intraluminal feeds inducing vagus nerve activity which can thus affect the effectiveness of GFAP levels.[21]

This study aimed to determine the effect of giving enteral formula containing protein, phosphatidylcholine, phosphatidylserine, and inulin on GFAP levels in ischemic stroke patients. The intervention group received 69 g of the powdered enteral formula containing 15 g of protein, 128 mg of phosphatidylcholine, 32 mg of phosphatidylserine, and 3 g of inulin dissolved in 250 ml of warm water, given three times a day for seven days.

Giving 69 g of powdered enteral formula because it contains high protein [22 g per 100 g in solid form][22]. The intake of phosphatidylcholine in the enteral formula also covers 60-90% of the daily requirement for phosphatidylcholine for adults and 3x1 enteral formula feeding for 7 days is following the therapeutic dosage for cytolin [23] where the administration of cytolin 2000 mg has a restorative effect.[24] According to the results of the study, it was shown that the administration of enteral nutrition within 7 days could reduce the mortality rate by 5.8% and survive more than the group who started giving nutrition late.[25] Feeding 69 g of enteral formula has been consistent with the therapeutic dose of phosphatidylserine, whereas administration of 100 mg/day of phosphatidylserine has provided improvement.[26] A dose of 0.5 g has been shown to increase the diversity of probiotic bacteria in the gut[27] which will produce short-chain fatty acids, namely SCFA. Inulin in the enteral formula has met the inulin dose for adults, which is 5-15 g /day and 0.5 g of inulin has been shown to increase the number of probiotic bacteria in the intestine which will produce short-chain fatty acids such as SCFA. In subjects suffering from diabetes, the treatment was carried out for 14 days at a dose of 34.5 g per day. The control group was given standard enteral formula from the hospital (11.8 g protein).

MATERIALS AND METHODS

This research is experimental research with a randomized control trial in a single-blind manner where the research subjects do not know what therapy is being obtained. The research subjects were 18 ischemic stroke patients aged >18 years, 9 men and 9 women who were divided into the intervention group (9) and the control group (9)

by randomization. This research was conducted for four months (February-May). The sample size was calculated by taking into account the minimum sample size by using the experimental sample size formula for clinical trials with the following formula:*****

$$n_1 = n_2 = 2\left(\frac{(Z_\alpha + Z_\beta)\ SB}{x1\text{-}x2}\right)^2$$

The results of previous studies showed that the level of GFAP in patients with ischemic stroke was 9.46 ng / mL (mean) and the standard deviation was 4.6 ng / mL.[28] The minimum difference considered significant (x1-x2) was 5.13 ng / mL.[28] If the error of type I is set at 15% (α=0.15) then Zα=1.440. The amount of type II error is set at 20% (β=0.20) then Zβ=0.842 and research power is 80%, the sample size calculation is as follows:

$$n = 2\left(\frac{(1.440 + 0.842)\ 4.6}{5.13}\right)^2$$
$$= 8.3 \approx 9 \text{ ischemic stroke patients/group}$$

When the sample was selected with the inclusion criteria of ischemic stroke patients with attack onset, not more than 72 hours as evidenced by an MSCT head scan and aged >18 years, and the exclusion criteria for ischemic stroke patients who had a head injury in the last 3 months, subarachnoid hemorrhage, brain tumor, infection of the central nervous system, psychiatric disorders, complications of acute or chronic renal failure, a history of reactions to enteral formula, hematemesis and / hematemesis + melena and those receiving citicoline. The researcher randomized the subjects by dividing the research subjects into the control group or the intervention group. Researchers provided an informed consent sheet for consent to be the study sample to the patient's family.

GFAP levels were checked at the beginning and the end of treatment. Examination of clinical characteristics is carried out by physical examination and a doctor examines neurological examination. The MSCT scan was performed to determine whether the patient had a head injury in the last 3 months, subarachnoid hemorrhage, brain tumor, infection of the central nervous system, psychiatric disorders, patient medical records to determine blood laboratory results (routine hematology, blood sugar, lipid profile) and complications. acute or chronic renal failure, history of reactions to enteral formula, hematemesis and / hematemesis + melena, and those receiving citicoline. During the study, researchers assessed the daily intake of food consumed by patients using the 24-hour recall method for 7 days. How to determine the patient's nutritional adequacy level through the average intake divided by the patient's requirement then multiplied by 100. The control group was given enteral formula based on the standard RS conventional enteral formula according to the character/comorbid of the ischemic patient (11.8 protein without the addition of phosphatidylcholine, phosphatidylserine, and inulin). The intervention group was given enteral formula containing 15 g of protein, 128 mg of phosphatidylcholine, 32 mg of phosphatidylserine, 3 g of 69 g of powdered enteral formula inulin, each dissolved in 250 ml of warm water and given three times a day for seven days. In the intervention group suffering from diabetes, the treatment was conducted for 14 days at a dose of 34.5 g per day. This provision is differentiated because the enteral formula given contains a high enough glycemic index which will have side effects in diabetes mellitus patients, so the administration will be reduced but it is carried out for a longer period. To avoid any side effects during the intervention, the researchers observed the patients every day.

The blood samples used were whole blood vein samples taken from the median cubital vein. The 6 ml blood sample was taken using a red vacutainer. The blood was then exported to the central laboratory of Dr. Kariadi for centrifuge and stored at –20ºC until testing. After the serum was successfully collected, the serum was sent to the Faculty of Medicine Undip GAKI laboratory for testing using the ELISA GFAP E-EL-H1888 kit.

This study has received Ethical Clearance approval by the Medical Research Ethics Commission FK UNDIP / RSDK with Number 503 / EC / KEPK-RSDK / 2020.

Statistical analysis was performed on a computer using the SPSS for windows version 20.0 program. Bivariate analysis was performed with the $\chi 2$ test or with the Fisher-Exact test to determine the change (Δ) in serum GFAP levels and the characteristics of the categorized subjects. The result is significant if $p < 0.05$. Multivariate analysis on confounding variables (age, sex, hypertension, diabetes mellitus, obesity status, dyslipidemia, history of stroke, rTPA therapy,

duration of intervention, and length of treatment) was carried out if any variables in the bivariate test were found to be different, with p <0.05 using General Linear Model.

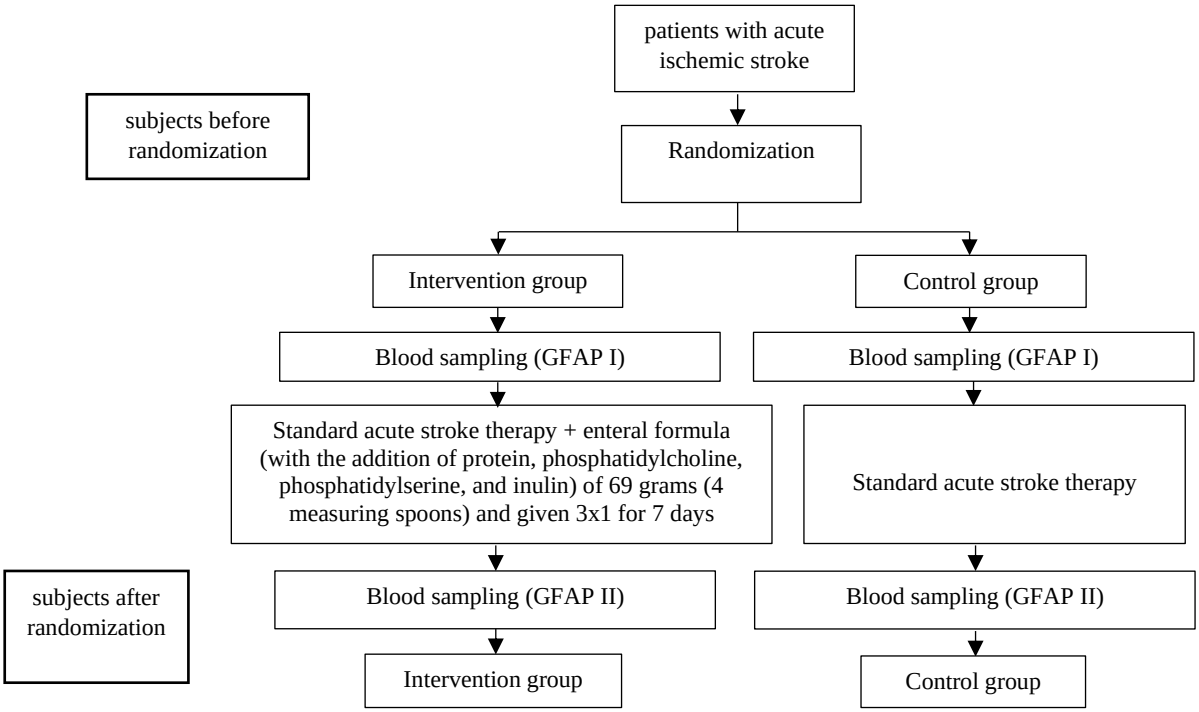

Figure 1. Flow chart of the research from the beginning to the end of the study

RESULTS

Characteristics of Research Subjects

In the implementation of the study, 8 out of 26 patients were found to have dropped out or lost to follow-up. Some of the things that caused the drop-out included patients who were not willing to continue the study (5) and died (3).

A total of 18 acute ischemic stroke patients were the subjects in the study. There were no significant differences in the categories of gender, age, BMI, obesity status, hypertension, diabetes mellitus, dyslipidemia, smoking, rTPA therapy, history of stroke, duration of intervention, and length of stay (p> 0.05). However, there were differences in the mean age category (p = 0.01).

Average Adequacy Level of Energy, Protein, Fat, Carbohydrates, Phosphatidylcholine, Phosphatidylserine, and Inulin Intake

Table 2 reveals that there is a significant difference in the adequacy of energy, protein, carbohydrate, phosphatidylcholine, phosphatidylserine, and inulin intake (p<0.05). There was no significant difference in the adequacy of fat intake between groups (p>0.05).

Difference of Mean and Change (Δ) GFAP levels

There was no difference in the mean GFAP level before the study between intervention group 8.37±4.25 and the control group 5.4±1.8 with a value (p = 0.08). These results designate that the conditions of the two groups before the intervention are corresponding (homogeneous). In the intervention group, the average GFAP level managed to decrease from 8.37±4.25 to 8.30±4.9, whereas in the control group, there was an increment from 5.4±1.8 to 7.5±4. This disparity shows that the intervention given affects even though this difference is not statistically significant (p = 0.9). Changes in Δ levels of GFAP in the intervention group also tended to decrease -0.07±2.26 compared to the control group which tended to increase 2.13±2.84 but not statistically significant (p> 0.05).

Table 1. Characteristics of Research Subjects in the Two Research Groups

Characteristics	Group		*p*
	Intervention (n=9)	Control (n=9)	
Gender			
a. Male	4 (44.4%)	6 (66.7%)	0.6[a]
b. Women	5 (55.6%)	3 (44.4%)	
Average age (years)	50.5 ± 14.8	65.5 ± 8.2	0.01[b]
Age Category (years)			
a. > 60	3 (33.3%)	7 (77.8%)	0.1[a]
b. <60	6 (66.7%)	2 (22.2%)	
Average BMI	25.2 ± 4	23.8 ± 2.6	0.3[b]
Obesity (BMI> 25)			
a. Yes	4 (44.4%)	2 (22.2%)	0.6[a]
b. Not	5 (55.6%)	7 (77.8%)	
Hypertension			
a. Yes	8 (88.9%)	7 (77.8%)	1[a]
b. Not	1 (11.1%)	2 (22.2%)	
Diabetes mellitus			
a. There is	3 (33.3%)	5 (55.6%)	0.6[a]
b. Not	6 (66.7%)	4 (44.4%)	
Dyslipidemia			
a. Yes	7 (77.8%)	8 (88.9%)	1[a]
b. Not	2 (22.2%)	1 (11.1%)	
Smoke			
a. Yes	4 (44.4%)	6 (66.7%)	0.6[a]
b. Not	5 (55.6%)	3 (33.3%)	
RTPA therapy			
a. Yes	2 (22.2%)	2 (22.2%)	1[a]
b. Not	7 (77.8%)	7 (77.8%)	
Stroke History			
a. Yes	3 (33.3%)	3 (33.3%)	1 [a]
b. Not	6 (66.7%)	6 (66.7%)	
Duration of Intervention			
a. 7 days	6 (66.7%)	9 (100%)	0.2 [a]
b. 14 days	3 (33.3%)	0 (0%)	
Length of Hospitalization			
a. <7 days	6 (66.7%)	3 (33.3%)	0.3 [a]
b. ≥ 7 days	3 (33.3%)	6 (66.7%)	

[a] *Fisher-Exact*; [b] *Independent T-Test*

Table 2. Adequacy Level of Patient Intake at Research

Variable (%)	Intervention	Control	*p*
	Mean ± SD	Mean ± SD	
Energy Adequacy Level	95 ± 18.4	75.7±11.9	0.018[b]
Protein Adequacy Level	94.1 ± 17.6	69.1±11.7	0.003[b]
Adequacy Level of Fat	74.3 ± 21.2	75.9±14.8	0.9[b]
Adequacy Level of Carbohydrates	106.9 ± 20.5	78.8±14.4	0.004[b]
Adequacy Level of Phosphatidylcholine	72.7±8	25.2±3.4	0.0[b]
Adequacy Level of Phosphatidylserine	112.3±12.3	49±10.2	0.0[b]
Adequacy Level of Inulin	92.4±7.9	24.1±7.6	0.0[b]

[b]*Independent T-Test*

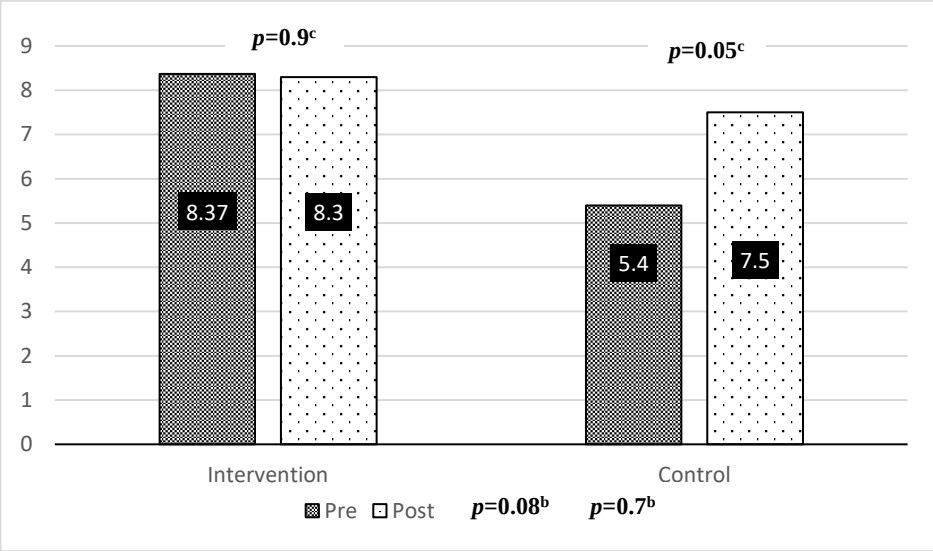

Figure 2. Average GFAP Levels Between Groups

[b] *Independent T-Test*; [c] *Paired T-Test*

Sub-Group Analysis of Inter-Group GFAP Levels

Median GFAP levels before the study in the intervention group DM subgroup 11.3 (2.4 – 12.4) with the control group 6 (3.9 – 8.1) was not statistically significant (p = 0.3). These results indicated that the state of the two groups before the intervention was in identical condition. In the DM sub-group in the intervention group, the median GFAP level increased from 11.3 (2.4 – 12.4) to 12.6 (2.1 – 12.8), it was the same as the control group which also increased from 6 (3.9 – 8.1) to 7 (3.4 – 15.6) and statistically, this difference was not significant (p>0.05). In the DM subgroup, the change in GFAP levels in the intervention group also decreased from 0.4 (-0.3 – 1.3) the same was the case with the control group 1 (-0.5 – 7.9) nonetheless, this difference was not statistically significant.

Median GFAP levels before the study in the non-DM subgroup of the intervention group 9.6 (1.1-12.3) with the control group 4.1 (2.8-6.7) were not statistically significant (p = 0.1). These results indicated that the condition of the two groups before the intervention is in the same condition. In the non-DM subgroup the intervention group median GFAP level there was a tendency to decrease from 9.6 (1.1-12.3) to 7.3 (0.8-14.2), in contrast to the control group which there was an increase from 4.1 (2.8-6.7) to 5.9 (3.8-8.2), and statistically, this difference was not significant (p>0.05). In the non-DM subgroup, the change in Δ GFAP levels in the intervention group also tended to decrease -0.2 (-3.5 – 3.4) is different the case with the control group which there was an increase of 0.8 (-0.5 – 4.9) however, this difference was not statistically significant.

Table 3. Sub-group analysis of GFAP levels between groups

Sub Group	Kelompok	n	Before	After	p	Δ
			Median (minimum-maximum)	Median (minimum-maximum)		Median (minimum-maximum)
DM	Intervention	3	11.3 (2.4 – 12.4)	12.6 (2.1 – 12.8)	0.2[d]	0.4 (-0.3 – 1.3)
	Control	5	6 (3.9 – 8.1)	7 (3.4 – 15.6)	0.1[c]	1 (-0.5 – 7.9)
	p		0.3[b]	0.8[e]		0.3[b]
Non DM	Intervention	5	9.6 (1.1-12.3)	7.3 (0.8-14.2)	0.7[c]	-0.2(-3.5 – 3.4)
	Control	5	4.1 (2.8-6.7)	5.9 (3.8-8.2)	0.2[c]	0.8 (-0.5 – 4.9)
	p		0.1[b]	0.4[b]		0.2[b]

[b] *Independent T-Test* ; [c] *Paired T-Test*; [d] *Wilcoxon*; [e] *Mann-Whitney*

GLM (General Linear Model)

Table 4 shows that there is no significant difference in GFAP levels controlled by the age variable (p = 0.3)

Table 4. General Linear Model

Variable	Covariat	*p*	R^2	Lower	Upper
(Δ) GFAP levels	Age	0.5	0.1	-0.14	0.079

DISCUSSION

Subject characteristics data revealed that there were no significant differences in the categories of gender, age, BMI, obesity status, hypertension, diabetes mellitus, dyslipidemia, smoking, rTPA therapy, history of stroke, duration of intervention, and length of stay (p>0.05). However, there were differences in the mean age category (p = 0.01). Stroke is often referred to as a disease suffered by older people. Currently, there was a trend that young people suffer from a stroke. The rising number of stroke cases at a young age can be prompted by a not-healthy lifestyle, such as smoking, alcohol consumption, frequent eating low-fiber foods such as fast food or junk food, and lack of exercise which will lead to hypertension. The habit is usually worsened by people in Central Java who are accustomed to consuming sweet foods or high glucose levels and foods containing coconut enteral formula which can trigger atherosclerosis and thrombosis, which results in reduced oxygen supply to the brain and can lead to stroke. The outcomes of the different test results for the adequacy of energy intake, carbohydrate intake, and fat intake between the intervention group and the control group showed significant results (p <0.05) but not for fat intake (p> 0.05). The exception is because one of the interventions given to the control group was standard enteral formula from the hospital, where before the 7th day and the patient had gone home, the researchers could not bring the enteral formula to continue therapy at home for one reason or another. Unlike the case with the intervention group before the 7th day; however, the patient had gone home, the researcher could provide the enteral formula to continue therapy at home. It could also be caused by monitoring food consumption before the study was completed. Researchers carry out a recall via cellular media such as WhatsApp because when the study took place coincided with the COVID-19 pandemic, which caused the patient to be unwilling to be visited by the researcher. If the patient during the study was still in the hospital, there was a standard portion for each type of diet. However, it is different from patients who have not returned home before the 7th day. There is also another possibility, such as when researchers do a recall, patients tend to overestimate or underestimate the food they eat. The size of the household measurement also varies, so it can affect recall results. There is also another possibility, such as when researchers do a recall, patients tend to overestimate or underestimate the food they eat. The size of the household measurement also varies, so it can affect recall results. There is also another possibility, such as when researchers do a recall, the patient tends to overestimate or underestimate the food consumed. The size of the household measurement also varies, so it can affect recall results.

The distribution of serum GFAP levels at the start and end of the study found that all patients were at abnormal levels (>0.11 ng/ml).[7] This could result in the presence of high-fat content or other anomalies in the sample that could prompt a matrix effect, which would interfere with detection. The hemolyzed blood sample contains the HRP analog (Horseradish Peroxidase), which can make a false-positive result. The hemolyzed sample releases a substance from the red blood cells, which can be mistaken for HRP. HRP is the critical reagent used for ELISA. Therefore, once the sample is hemolyzed, detection will be affected. This is one of the factors that can influence GFAP.

Other factors that can affect GFAP occur in several cellular processes. The drastic decrease in oxygen in the brain causes the brain to undergo anaerobic metabolism so that the availability of ATP is low. Hypoxic conditions cause neuron cell death in minutes because the brain is a part that is very sensitive to hypoxic states. Hypoxia triggers a series of pathways that lead to decreased energy and the extracellular release of glutamate. Neurons and glial cells can die through various causes such as excitotoxicity, cellular edema, oxidative stress, and inflammation. Hypoxia increases the formation of reactive oxygen species (ROS) which results in oxidative stress on cells. Increased levels of ROS are a major cause of brain tissue damage after hypoxia. ROS damage tissue directly through the modification of cellular proteins, lipids, and DNA. ROS indirectly interferes with cellular signaling and the regulation of gene expression. Astrocytes play an

especially important role in the communication between astrocytes-neurons through the release of several neurotrophic factors to maintain central nervous system homeostasis. Astrocytes also play a role in maintaining homeostasis in the environment around neurons. Recent research has shown that astrocytes are immunocompetent in the brain. If there is a disturbance in the brain that causes damage, the astrocytes will be activated and move quickly to the site of damage (astrogliosis). On excessive activation, it turns out that astrocytes increase the production of several substances, one of which is a glial fibrillary acidic protein (GFAP).[29]

Mean baseline GFAP serum levels in the intervention group 8.37±4.25 and control group 5.4± 1.8 with a value (p = 0.08). The final study serum GFAP levels in the intervention group were 8.30±4.9 and in the control group was 7.5±4. These numbers showed that the intervention given still affects even though the difference is not statistically significant (p = 0.9). In Figure 2, the intervention group declined -0.07±2.26 compared to the control group, which increased 2.45±2.86. This difference was statistically significant (p = 0.03).

The difference is partly because the patients in the study sample were patients who had repeated strokes (had a history of stroke), which according to the study of Ren *et al.* (2016) which examined GFAP levels in 13 subjects with hemorrhagic strokes, 23 subjects with ischemic strokes and 7 subjects with recurrent stroke reported that in patients with ischemic stroke, the mean serum GFAP level was significantly lower in subjects without a history of stroke compared with patients who had a previous stroke (p = 0.004). This increment occurred due to persistent pathophysiological processes including vascular disorders, long-term changes in BBB permeability, and damage to the brain parenchyma itself triggered by an initial stroke.[30] In this study, patients with recurrent strokes found five patients in the control group, three patients in the treated group.

This can then be caused by comorbid patient diseases such as diabetes mellitus. Research conducted by Lotosh *et al.* (2013) on experimental animals by inducing streptozotocin (STZ) showed that autoantibodies against neuron proteins, especially GFAP increased sharply on day five after being induced by streptozotocin. In this study, it can be concluded that STZ produces a toxic effect on astrocytes which causes the release of GFAP protein into the bloodstream and causes the formation of AAb (autoantibody) which is specific to GFAP protein.[31]

The COVID-19 pandemic can also cause another thing during the research, which resulted in a lack of research samples. Where in a study conducted by Zang *et al.* (2019) which examined the relationship between giving citicoline injection to 102 patients with hemorrhagic stroke and GFAP levels showed a significant relationship (p = 0.000).[28]

Another thing is made possible by the dose of enteral formula that is too small, the duration of administration, and the route of administration which can be the cause of the research results. The doses in this study were 45 g protein/day, 384 mg/day phosphatidylcholine, 96 g/day phosphatidylserines, and 9 g/day inulin. In some studies, the minimum duration of intervention given was 14 days and obtained significant results. [26,32–35]

The results obtained by the DM sub-group analysis of GFAP levels in the intervention group showed an increasing trend of 0.4 (-0.3 – 1.3) compared to the control group of 1 (-0.5 – 7.9) and statistically not significantly different (p>0.05). Unlike the case with the non-DM subgroup in the intervention group which showed a decreasing trend of -0.2(-3.5 – 3.4) compared to the control group where there was an increasing trend of 0.8 (-0.5 – 4.9) and not statistically significant (p> 0.05). This occurred because, in patients with hyperglycemia, it can provoke nerve degeneration through oxidative stress. Metabolic and oxidative factors often cause rapid changes in glial cells. A key indicator of this response was an increased synthesis of GFAP, which is an astrocytic marker. The event was evidenced by the study of Baydas *et al.* (2003) who conducted a study on 40 experimental animals divided into three groups. The first group was injected with STZ, the second group was injected with STZ + melatonin, and the last group was the control group. The results showed that the STZ-induced group caused glial reactivity, while the STZ + melatonin-injected group decreased the GFAP recativity.[36]

Research on the effect of giving protein, phosphatidylcholine, phosphatidylserine, and inulin on markers of brain damage such as GFAP is still minimal. In this study, the intervention was administered orally or in the form of powdered enteral formula with protein,

phosphatidylcholine, phosphatidylserine, and inulin. Efficiently, phosphatidylcholine and phosphatidylserine are almost entirely absorbed, which then spreads throughout the body across the blood-brain barrier and into the central nervous system. Inulin is easily dissolved in water and difficult to hydrolyze by enzymes in the digestive tract so that it can reach the large intestine as a whole. Probiotic bacteria will ferment inulin in the intestines and become short-chain fatty acids called SCFA.[10,23,27]

In this study, the intervention gave still affected even though it was not statistically significant (p> 0.05), according to research from Ji *et al.* (2018), a high protein diet given earlier can improve motor coordination. Currently, most of the hospitals have implemented facilities such as flexor and elbow extensor training, physiotherapy, for example, teaching good movement patterns and posture. However, this facility will not be optimal if there are patients with severe disabilities and a lack of cooperation between patients and hospital staff. Increasing SOD expression, restoring iNOS expression, and suppressing MDA expression are among the advantages of the early high protein diet. This suggests that oxidative stress can be inhibited if a high-protein diet is given early on so that free radicals and inflammation in the body can be reduced and can improve post-stroke clinical outcomes.[32]

Supported by research by Aquilani *et al*. (2008) which provided supplementation with hyperproteic nutritional formulas (10% protein = 40 grams of protein) in 20 patients for 21 days, it showed that the National Institutes Stroke Scale (NIHSS) scores of patients with protein administration increased on average 4.4 compared to a control group average of 3.0. However, there was an increase in motor recovery of the paretic arms and legs which were found to be better able to withstand gravity for 10 seconds (with legs outstretched) in the intervention group. The improvement in the performance of the extremities against gravity was less seen in the control group patients.[33] In this study, it has a ratio of 1: 2 related to protein administration with research conducted by Aquilani.

In a study conducted by Tykhomyrov *et al.* (2019) who examined the effect of giving CDP-choline or citicoline injections to 33 ischemic stroke patients with GFAP levels who gave citicoline injection of 1000 mg for 14 days, they decreased GFAP levels by 61% with (p = <0.01). CDP-choline can protect astrocytes and neurons, as well as increase angiogenic capacity through downregulation of angiostatin in post-ischemic patients with atrial fibrillation after acute ischemic stroke. Also, CDP-choline can increase cell proliferation, vasculogenesis, and synaptophysin levels and reduce GFAP levels in the peri-infarct area in ischemic stroke. In this study, enteral nutrition was given with a content of 384 phosphatidylcholine/day.[34]

In a study conducted by Kataoka *et al*. (2010) which examined the effect of Soy-PS administration on 78 elderly people with mild cognitive impairment (50-69 years) were randomly selected to consume Soy-PS (100 mg or 300 mg/day) or placebo for 6 months. Administration of Soy-PS orally for 6 months improved memory function, especially delayed memory in the elderly with memory complaints. Administration of Soy-PS was able to improve memory function, especially delayed memory in the elderly with memory complaints. Increasing the memory score is also one of the benefits of giving Soy-PS. The activation of signaling proteins and receptors that are essential for neuronal survival is influenced by phosphatidylserine. Phosphatidylserine also plays a role in signaling processes such as regulating neurotransmitters. In this study, the intervention was given with enteral nutrition containing 96 phosphatidylserines/day for 7 days.[26]

The administration of inulin in Hofman's study was 8% by injection of inulin in mice which were equivalent to 40 grams of inulin in humans per day. Hoffman's research shows that 8% inulin can increase cecal content, produce more SCFA, increase the number of bacterial enzymes in the cecum, increase systemic metabolism by modulating the gut microbiome, and can reduce the characteristics of brain inflammation in early AD (Alzheimer's Disease) compared to 4% inulin. Hoffman's research also states that 8% is considered the maximum tolerable and beneficial amount for the human organism. In this study, 9 grams of inulin per day were given.[35]

Inulin is a part of prebiotics. Prebiotics is a component of food that cannot be digested by the digestive tract enzymatically, which will then fermented by microbiota in the intestine. Inulin is a substrate that progresses the diversity of beneficial microbiota in the intestine so that the production of SCFAs such as acetate, propionate, and butyrate will increase as a result of anaerobic fermentation. The gut-brain axis is influenced by

SCFA either directly or indirectly. SCFA binds to various receptors to further inhibit histone deacetylation and affect the integrity of the intestinal barrier and intestinal mucosal immunity. The interaction performed by the SCFA on various receptors can induce indirect signaling to the brain by inducing the secretion of intestinal hormones. Systemic inflammation is affected by SCFA by regulating leukin secretion and inducing differentiation of Tregs or T regulatory cells. The blood-brain barrier can be penetrated by the SCFA through the MCT or monocarboxylate transporter and improves the integrity of the blood-brain barrier. SCFA can enhance neurogenesis, function, and homeostasis of nerve cells and improve the morphology and function of glial cells found in the central nervous system. [35,37]

RTPA therapy also appears to contribute to study results. In the study by Davalos *et al.* (2002), 47% of patients were treated with rTPA and given citicoline therapy 1000 mg per 12 hours intravenously for three days, then orally were given 2x500 mg tablets per 12 hours for a total of six weeks. rTPA can rebuild blood flow in the penumbra; therefore it is difficult for citicoline to enhance the performance caused by the effects of thrombolysis. The severity of most stroke patients in Davalos *et al.*'s study was also high so that no penumbra region was susceptible to being saved by citicoline. Consequently, in the subgroup without rTPA, the efficacy of citicoline exhibited better results than the group with rTPA.[24] In this study, 4 patients received rTPA therapy.

Furthermore, it can be caused by patients who have gone home before the 7th day. Although given enteral formula to continue therapy at home, this can cause researchers to be unable to control whether or not the intervention is complete. The drugs received and the presence of comorbidities that concern the improvement of clinical outcome in ischemic stroke patients, because of these factors, a significant reduction in GFAP levels required.

A multicentre study conducted on 4023 ischemic stroke patients who were given a protein-energy supplement of 62.5 g / day for 6-7 months in 125 hospitals and 15 countries showed that routine oral protein energy supplementation of a usual hospital diet did not improve outcomes in patients admitted with recent stroke.[38] A study by Sabin *et al.* (2013) giving citicoline to 347 ischemic stroke subjects (1g / day for 12 months) showed no significant difference (p = 0.186).[39] Supported by Clark's study that gave citicoline to 453 ischemic stroke subjects (1000 mg 2x / day for 12 weeks) showed no significant difference (p>0.05).[40] A study conducted by Manor *et al.* (2013) regarding the administration of phosphatidylserine in 30 elderly with memory complaints (300 mg/day for 12 weeks) showed no significant difference in focused attention, sustained attention, visuospatial learning, spatial short-term memory (p>0.05).[41] The study by Tuncay *et al.* (2018) who performed Enteral Formula with Probiotic Content (EFPC) intervention in 23 neuro-critical care patients for 21 days was associated with more frequent dosing of subjects showing no significant difference.[42]

The results of this study emphasized that enteral formula containing protein, phosphatidylcholine, phosphatidylserine, and inulin had a good effect on GFAP levels, as evidenced by changes (Δ) in serum GFAP levels in the intervention group which decreased -0.07±2.26 compared to the control group, which increased 2.13±2.84 although it was not statistically significant (p>0.05).

In General Linear Model (GLM), it shows that there is no significant difference in (Δ) GFAP levels were controlled by age variable (p>0,05). This may be due to the lack of research samples. During the study, there was a COVID-19 pandemic which made patients tend to hesitate or fear to have themselves examined at Dr. Kariadi. Remembering Dr. Kariadi is a referral hospital in Semarang.

The limitations of this study were the shortage of sample, length of time, and dose of administration. The advantage of this study is to provide enteral nutrition therapy to stroke patients by examining brain damage markers (GFAP) because this study is still very limited, so it can add to the literature for other researchers.

CONCLUSIONS

The addition of 15 g protein, 128 mg phosphatidylcholine, 32 mg phosphatidylserine, and 9 g inulin given for seven days had no significant effect on GFAP levels.

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REFERENCES

1. Liu G, Geng J. Glial fibrillary acidic protein as a prognostic marker of acute ischemic stroke. Hum Exp Toxicol. 2018;37(10):1048–53.
2. Surjawan Y, As'ad S, Ranakusuma TAS, Wijaya A. The different patterns of blood S100b protein and GFAP concentrations in ischemic stroke. Med J Indones. 2013;22(4):215–20.
3. Petzold A. Glial fibrillary acidic protein is a body fluid biomarker for glial pathology in human disease. Brain Res [Internet]. 2015;1600:17–31. Available from: http://dx.doi.org/10.1016/j.brainres.2014.12.027
4. Middeldorp J, Hol EM. GFAP in health and disease. Prog Neurobiol [Internet]. 2011;93(3):421–43. Available from: http://dx.doi.org/10.1016/j.pneurobio.2011.01.005
5. Žurek J, Fedora M. The usefulness of S100B, NSE, GFAP, NF-H, secretagogin and Hsp70 as a predictive biomarker of outcome in children with traumatic brain injury. Acta Neurochir (Wien). 2012;154(1):93–103.
6. Dvorak F, Haberer I, Sitzer M, Foerch C. Characterisation of the diagnostic window of serum glial fibrillary acidic protein for the differentiation of intracerebral hemorrhage and ischaemic stroke. Cerebrovasc Dis. 2009;27(1):37–41.
7. Puspitasari V, Gunawan PY, Wiradarma HD, Hartoyo V. Glial fibrillary acidic protein serum level as a predictor of clinical outcome in ischemic stroke. Open Access Maced J Med Sci. 2019;7(9):1471–4.
8. Raisa N, Sujuti H, Purnomo H, Rahayu M, Dalhar M. the Influence of Sampling Time To Serum Glial Fibrillary Acidic Protein (Gfap) Levels in Acute Stroke. MNJ (Malang Neurol Journal). 2019;5(1):30–3.
9. Presley B. Penatalaksanaan Farmakologi Stroke Iskemik Akut. Bul Rasional [Internet]. 2013;12(1):6–8. Available from: http://repository.ubaya.ac.id/21378/1/Rasional Vol 12 No 1.pdf
10. Glade MJ, Smith K. Phosphatidylserine and the human brain. 2015;31(6):781–6. Available from: http://dx.doi.org/10.1016/j.nut.2014.10.014
11. Overgaard K. The effects of citicoline on acute ischemic stroke: A review. J Stroke Cerebrovasc Dis [Internet]. 2014;23(7):1764–9. Available from: http://dx.doi.org/10.1016/j.jstrokecerebrovasdis.2014.01.020
12. Sahota P, Savitz SI. Investigational Therapies for Ischemic Stroke: Neuroprotection and Neuro recovery. Neurotherapeutics. 2011;8(3):434–51.
13. Kato-Kataoka A, Sakai M, Ebina R, Nonaka C, Asano T, Miyamori T. Soybean-derived phosphatidylserine improves memory function of the elderly Japanese subjects with memory complaints. J Clin Biochem Nutr. 2010;47(3):246–55.
14. Anderson H, Luhovyy B, Akhavan T, Panahi S. Enteral formula proteins in the regulation of body weight, satiety, food intake, and glycemia. Nestle Nutr Work Ser Pediatr Progr. 2011;67:147–59.
15. Lien DTK, Nhung BT, Khan NC, Hop LT, Nga NTQ, Hung NT, et al. Impact of enteral formula consumption on performance and health of primary school children in rural Vietnam. Asia Pac J Clin Nutr. 2009;18(3):326–34.
16. Patel S. Functional food relevance of whey protein: A review of recent findings and scopes ahead. J Funct Foods. 2015;19:308–19.
17. Mitchell CJ, McGregor RA, D'Souza RF, Thorstensen EB, Markworth JF, Fanning AC, et al. Consumption of enteral formula protein or whey protein results in a similar increase in muscle protein synthesis in middle-aged men. Nutrients. 2015;7(10):8685–99.
18. Colonic T, Homeostasis C, Smith PM, Howitt MR, Panikov N, Michaud M, et al. Reports 1000. 2013;(August):569–74.
19. Liu TW, Cephas KD, Holscher HD, Kerr KR, Mangian HF, Tappenden KA, et al. Nondigestible fructans alter gastrointestinal barrier function, gene expression, histomorphology, and the microbiota profiles of diet-induced obese C57BL/6J Mice. J Nutr. 2016;146(5):949–56.
20. Chunchai T, Keawtep P, Arinno A, Saiyasit N, Prus D, Apaijai N, et al. N-

acetyl cysteine, inulin and the two as a combined therapy ameliorate cognitive decline in testosterone-deprived rats. Aging (Albany NY). 2019;11(11):3445–62.
21. Bansal V, Costantini T, Ryu SY, Peterson C, Loomis W, Putnam J, et al. Stimulating the central nervous system to prevent intestinal dysfunction after traumatic brain injury. J Trauma - Inj Infect Crit Care. 2010;68(5):1059–63.
22. BPOM RI. Peraturan Kepala Badan Pengawas Obat dan Makanan Republik Indonesia Nomor 9 Tahun 2016 tentang Acuan Label Gizi. Badan Pengawas Obat dan Makanan Republik Indones. 2016;1–28.
23. Wahyudi R, Hasmono D, Fitrina R, Armal K. Injected Citicoline Improves Impairment and Disability During Acute Phase Treatment in Ischemic Stroke Patients. Folia Medica Indones. 2016;51(4):245.
24. Dávalos A, Castillo J, Álvarez-Sabín J, Secades JJ, Mercadal J, López S, et al. Oral citicoline in acute ischemic stroke: An individual patient data pooling analysis of clinical trials. Stroke. 2002;33(12):2850–7.
25. Wirth R, Smoliner C, Jäger M, Warnecke T, Leischker AH, Dziewas R, et al. Guideline clinical nutrition in patients with stroke. Exp Transl Stroke Med. 2013;5(1):1–11.
26. A. K-K, M. S, R. E, C. N, T. A, T. M. Soybean-derived phosphatidylserine improves memory function of the elderly Japanese subjects with memory complaints. J Clin Biochem Nutr [Internet]. 2010;47(3):246–55. Available from: http://linker.worldcat.org/?rft.institution_id=129635&spage=246&pkgName=UKPMC&issn=0912-0009&linkclass=to_article&jKey=897&issue=3&provider=NLM&date=2010-11&aulast=Kato-Kataoka+A.%3B+Sakai+M.%3B+Ebina+R.%3B+Nonaka+C.%3B+Asano+T.%3B+Miyamori+T.&atitle=Soyb
27. Setiarto RHB, Widhyastuti N, Saskiawan I, Safitri RM. Pengaruh Variasi Konsentrasi Inulin Pada Proses Fermentasi Oleh Lactobacillus acidophilus, Lactobacillus bulgaricus Dan Streptococcus thermophilus. Biopropal Ind. 2016;8(June):1–17.
28. Zang J, Liu AX, Qi L. The cytological mechanism and effects of hypertensive cerebral hemorrhage treatment by citicoline on serum GFAP and copeptin level. Eur J Inflamm. 2019;17.
29. Andriani A, Prijanti AR, Mudjihartini N, Jusman SWA. Dampak Hipoksia Sistemik terhadap Malondialdehida, Glial Fibrillary Acidic Protein dan Aktivitas Asetilkolin Esterase Otak Tikus. eJournal Kedokt Indones. 2016;4(2).
30. Ren C, Kobeissy F, Alawieh A, Li N, Li N, Zibara K, et al. Assessment of serum UCH-L1 and GFAP in acute stroke patients. Sci Rep. 2016;14(6):1–9.
31. Lotosh NG, Savel'Eva EK, Selishcheva AA, Savel'Ev S V. Autoantibodies to neuron-specific proteins S100, GFAP, MBP, and NGF in the serum of rats with streptozotocin-induced diabetes. Bull Exp Biol Med. 2013;155(1):48–51.
32. Ji M, Li S, Dong Q, Hu W. Impact of an early high-protein diet on neurofunctional recovery in rats with ischemic stroke. Med Sci Monit. 2018;24:2235–43.
33. Aquilani R, Scocchi M, Iadarola P, Franciscone P, Verri M, Boschi F, et al. Protein supplementation may enhance the spontaneous recovery of neurological alterations in patients with ischaemic stroke. Clin Rehabil. 2008;22(12):1042–50.
34. Tykhomyrov AA, Kushnir YS, Nedzvetsky VS, Grinenko T V., Kuryata O V. Citicoline affects serum angiostatin and neuro specific protein levels in patients with atrial fibrillation and ischemic stroke. Ukr Biochem J. 2019;91(5):34–45.
35. Hoffman JD, Yanckello LM, Chlipala G, Hammond TC, McCulloch SD, Parikh I, et al. Dietary inulin alters the gut microbiome, enhances systemic metabolism, and reduces neuroinflammation in an APOE4 mouse model. PLoS One. 2019;14(8):1–22.
36. Baydas G, Reiter RJ, Yasar A, Tuzcu M, Akdemir I, Nedzvetskii VS. Melatonin reduces glial reactivity in the hippocampus, cortex, and cerebellum of streptozotocin-induced diabetic rats. Free

Radic Biol Med. 2003;35(7):797–804.
37. Silva YP, Bernardi A, Frozza RL. The Role of Short-Chain Fatty Acids From Gut Microbiota in Gut-Brain Communication. Front Endocrinol (Lausanne). 2020;11(January):1–14.
38. Dennis M. Routine oral nutritional supplementation for stroke patients in hospital (FOOD): A multicentre randomized controlled trial. Lancet. 2005;365(9461):755–63.
39. Alvarez-Sabín J, Ortega G, Jacas C, Santamarina E, Maisterra O, Ribo M, et al. Long-term treatment with citicoline may improve poststroke vascular cognitive impairment. Cerebrovasc Dis. 2013;35(2):146–54.
40. Clark WM, Wechsler LR, Sabounjian LA, Schwiderski UE. A phase III randomized efficacy trial of 2000 mg citicoline in acute ischemic stroke patients. Neurology. 2001;57(9):1595–602.
41. Manor I, Magen A, Keidar D, Rosen S, Tasker H, Cohen T, et al. Safety of phosphatidylserine containing omega3 fatty acids in ADHD children: A double-blind placebo-controlled trial followed by an open-label extension. Eur Psychiatry. 2013;28(6):386–91.
42. Tuncay P, Arpaci F, Doganay M, Erdem D, Sahna A, Ergun H, et al. Use of standard enteral formula versus enteric formula with prebiotic content in nutrition therapy: A randomized controlled study among neuro-critical care patients. Clin Nutr ESPEN [Internet]. 2018;25:26–36. Available from: https://doi.org/10.1016/j.clnesp.2018.03.123

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Collaborative model as a training for increasing village health worker competency about complementary feeding

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ABSTRACT

Background: A Kader refers to a Village Health Worker (VHW), a volunteer, which becomes one of the sources of community reference. Commonly, they sustain a mother's knowledge regarding adequate complementary feeding. However, there are still some VHW who have not possessed a health education background nor been able to be independent.

Objectives: This study aimed to analyze the impact of collaborative models on VHW's competence as in knowledge, self-efficacy, attitudes, and counseling skill on the topic of complementary feeding.

Materials and Methods: The design of this research is a quasi-experiment control group pretest-postest, with the retrieval of subjects using purposive sampling. Subjects were selected based on the location of the posyandu. The population in this research was VHWs in the Lembang district were 40 VHWs in each group. The treatment group was given training intervention for 1 month with a collaborative model, combining several methods into a series. The main topic was counseling and complementary feeding. The control group was given booklets and leaflets. VHW's competence was measured using questionnaires. This research was conducted in January-April 2020.

Results: Statistical test results before the treatment of both groups showed no difference (p>0,05) in each variable. Two months after the intervention, there were significant differensces in the mean score of knowledge (p=0,001), attitude (p=0,001), and VHWs self-efficacy (p=0,000) in both groups. VHW counseling skills (p=0,149) until the first month there was not a significant difference. Unexpectedly, in the second month, the VHW counseling skills could not be observed due to the global COVID-19 pandemic. The mean score of each group increased significantly, but the score of the treatment group was higher than the control group.

Conclusions: A collaborative model is effective when compared to only providing booklets and leaflets in increasing VHW's knowledge, attitudes, and self-efficacy but not effective yet for VHW counseling skills.

Keywords: Collaborative model; Competence; Complementary feeding; VHW

BACKGROUND

The double burden of malnutrition is a health problem that still needs to be seriously addressed. The double burden of malnutrition is indicated by malnutrition problems (stunting and wasting) along with problems with overnutrition (obesity).[1] Based on Basic Health Research (*RISKESDAS or Riset Kesehatan Dasar in Bahasa Indonesia*), the prevalence of malnutrition and malnutrition under five in Indonesia is still high. The progress from 2007 to 2013 has shown improvements, although the process for correcting all forms of malnutrition is still quite slow.[2]

The prevalence of severe malnutrition and malnutrition (weight-for-age) in Indonesia in 2018 was 3.9% and 13.8%. At the provincial level, malnutrition and undernutrition for children under five in West Java were 2.6% and 10.6% respectively. The prevalence of very short and stunted children (weight-for-age) in Indonesia in 2018 is 11.5% and 19.3%. At the provincial level, very short and short children under five in West Java reveals a figure of 11.7%, and 19.4%, which is slightly higher than the national figure. In 2018 the prevalence of very thin and underweight (weight-for-height) in Indonesia was 3.5% and 6.7%. Additionally, very thin and underweight toddlers in West Java expose a figure of 3.2% and 5.2%, which is slightly lower than the national figure.[2] The most serious malnutrition problem in West Java is in West Bandung Regency with a percentage of 22.4%.[3] This high prevalence rate indicates that Indonesia is still

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experiencing serious nutritional problems that need to be prevented and addressed.

One of the factors provoking malnutrition is the inadequate provision of complementary foods and early weaning. The attitudes and understanding of mothers about how to provide complementary foods, how to maintain nutritional intake, and manage food contribute to cases of malnutrition in infants and toddlers.[4,5]

A health *kader* refers to Village Health Worker (VHW) who is a volunteer selected by the community and tasked with developing the community, in this case, VHWs are also summoned as health promoters or promoters.[6] VHWs` contributions to improving access to care and health knowledge, behaviors, and outcomes are well documented, notably for conditions such as asthma, hypertension, diabetes, and HIV/AIDS; for procedures such as cancer screening and immunizations; and for maternal and child health in general.[7–11] One of the duties of a VHW is to provide health education, especially in *Posyandu*. *Posyandu* is Integrated Services Post (SIP) in Indonesian. *Posyandu* is a platform where VHW is linked to the problems currently being faced by society. In other words, VHWs are reference persons who maintain a close relationship with the community.[12,13] The existence of *Posyandu* and VHWs is needed in a promotional and preventive approach, especially concerning improving child nutrition and maternal and child health.[14] In this case, they are expected to gain expertise and proficiency in providing counseling and giving assistance on exclusive breastfeeding and appropriate complementary foods. Based on interviews with several VHWs in Lembang Subdistrict, many VHWs were not skilled in serving and building excellent and pleasant communication. For instance, some VHWs used regional languages that sounded harsh or played high pitch while communicating with the patients.

Other factors that influence the performance of VHWs apart from knowledge include self-efficacy.[15] A person's level of confidence in his ability is expressed as self-efficacy.[16] A person who has high self-efficacy will be able to think quickly and have stable self-confidence in managing his duties when situations requiring high levels of stress.[17] In this manner, a VHW is expected to develop positivity and self-efficacy, one of which can be acquired through the learning process or training.

The local government has provided various sessions of training to VHWs, especially regarding the issues of mothers and toddlers.[18] Based on preceding research, in this Lembang sub-district, training for VHWs had been held. The methods used in the previous training were lectures and workshops, which intended to broaden the knowledge, upgrade skills of VHWs in various matters, as well as giving practical solutions in providing VHWs competence.[19,20] The method used by the Lembang District government was the conventional method. The weakness of the conventional method is that the VHWs get bored quickly, which has made it challenging to absorb the information. Based on the interview, the VHWs wanted training with a modified method because they thought that the substance presented was too heavy to be conveyed by the conventional system.

Learning methods that are considered effective for community-based health education are active and independent learning processes. One of which is the collaborative learning method.[21] Collaborative learning is an educational approach to teaching and learning that involves groups of learners working together to solve a problem, complete a task, or create a product.[22] The learning model is a framework of the learning approach, learning strategies, learning methods, learning media, and learning techniques.[23] Collaborative methods are commonly used in formal education, however, this method is rarely used for community-based health education or training.[24] In this case, this refers to arranging a learning concept that incorporates government and community participation. In the collaborative learning method, implementation with two or more parties involved is preferable.[25]

One research that implemented collaborative training in the cattle breeder community conducted by Bank Indonesia (BI) representatives of West Sumatra, BPTUHPT Padang Mengatas, Faculty of Animal Husbandry, Andalas University, Department of Animal Husbandry and Animal Health, SKPD of Animal Husbandry services in West Pasaman Regency and private business institutions designated that participants encountered changes (improvements) in cognitive aspects of (43.53%).[26]Through a collaborative method of training, VHWs are expected to improve their performance. In regards to the gap of VHW`s proficiency and their main work, it is necessary to research the effect of collaborative learning models on knowledge, self-efficacy, attitudes, and skills of VHWs in counseling regarding complementary feeding.

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MATERIALS AND METHODS

This study employed a quasi-experimental non-randomized control group pretest-postest design by taking the number of subjects using purposive sampling. The population in this study was 161 VHW in Lembang District, West Bandung Regency. Respondents in this study were 80 active VHWs who met the inclusion criteria. All respondents were divided into two groups, namely the group provided with training facilities using collaborative methods which included the provision of booklets and leaflets (the treatment group), and the group that was only given booklets and leaflets as learning facilities (control group). The treatment and control groups were determined by grouping by *Posyandu* area to anticipate interactions between groups during treatment. The subject size was calculated using the formula for the mean of two independent populations. The subjects obtained were 40 VHWs in each group.

The criteria for respondents in this study were taken based on inclusion and exclusion criteria. The inclusion criteria in this study comprise; VHWs are active with 2-5 years of experience, VHW attends the activities, and VHWs are willing to become respondents. VHWs who were unable to attend the event and not willing to be respondents are involved in the exclusion criteria.

This training was conducted in January - April 2020 in Lembang District, West Bandung Regency. The independent variable in this study is the provision of training with a collaborative learning model for VHWs, while the dependent variable is the knowledge, attitudes, self-efficacy, and counseling skills of VHWs. The variables of knowledge, attitudes, and VHW counseling skills were obtained by using a questionnaire that had been tested for validation and reliability testing. The self-efficacy variable uses the General Self-efficacy Scale (GSE) instrument developed by Schwarzel & Jarusalem (1995) in the adaptation of Indonesian which has also been tested for validation and reliability testing.[27]

The pre-test was carried out in the first to the third week of January. Training for the treatment group was carried out from the fourth week of January to the third week of February. Treatment is carried out once a week, on Saturdays for training and during the activities in Posyandu for facilitator assist. The first post-test was carried out in the first to the third week of March and the second post-test was carried out in the first to the third week of April. The training for the treatment group was guided by two facilitators. There were 2 facilitators, consisting of 1 person from the West Bandung District Health Office and 1 lecturer from the Indonesian Education University (Universitas Pendidikan Indonesia). Facilitators are selected based on recommendations from the health office and university. Researchers also discuss and explain the methods used in the training to be implemented. The researcher reiterates whether the facilitator can use an intervention method that combined seminar, role-playing, group discussions, and technical guidance in a series or not. Then decide whom the facilitator will fill in the training. The distribution of booklets and leaflets for VHWs (control group) was carried out at the same time as the first week of training for the treatment group at different locations.

Booklet and leaflet containing discussion of balanced nutrition for infants around 6-24 months. An infant's need for energy and nutrients starts to exceed what is provided by breast milk, and complementary foods. A detailed discussion about what to how recommends that infants start receiving complementary foods. The booklet also contains an explanation of basic communication and counseling for VHWs. Meanwhile, the leaflet only contains infographics regarding complementary feeding, which is made short and easy to understand. This leaflet is given to be a material that VHWs can use in counseling.

Implementation of Training

The training for the treatment group was carried out within 1 month. In the first and second weeks of meetings, training was directed at one of the selected *Posyandu*. In the first week, the VHWs studied a set of materials on complementary foods. The training was carried out with the guidance of two facilitators. At first, VHWs who had been gathered in one class were given a set of booklets and leaflets, succeeded by the presentation by the facilitator. The facilitator presented the materials verbally with slide tools. In presenting the material, the facilitator also explains using props and occasionally explains in role plays.

Subsequently, VHWs were asked to discuss how to form groups of 5-6 people. Each VHW was demanded to make a question. The facilitator exchanged the questions, and later each group was urged to solve the question through discussion. The facilitator went around monitoring the discussion activities and helped out once in a while. Each group presented the results of the discussion, and the other groups presented their responses. The facilitator helped by explaining the remaining unanswered questions.

In the second week, the material presented is about counseling activities. The facilitator also carried out the training activities like the first week. The activities were more or less like the prior week. Two facilitators also conducted the material for the second week. In the third week, the facilitator assisted VHWs during the activities in *Posyandu*. The facilitator reaffirmed the core points to be conveyed to each of the VHWs.

To conduct a statistical analysis of the characteristics of the respondents, we employed descriptive and frequency distribution tables. Analysis of the mean difference in confounding variables used the independent t-test for normally distributed data, the Mann Whitney test for abnormally distributed data, and the Chi-square test for nominal data types. The difference in the mean of each dependent variable in the two groups before and after the intervention was carried out using a paired t-test because the data were normally distributed. To find the mean difference test for each dependent variable before and after the intervention between groups, we utilized the Mann-Whitney test because the data were not normally distributed. The multivariate test conducted was a general linear model, repeated measure against each variable after the intervention in both groups. The statistical test in this study used a 95% confidence level. This research has passed the research ethics review by the Health Research Ethics Commission of the Faculty of Medicine, Diponegoro University.

RESULTS

Respondent Characteristics

Table 1 explicates that most of the age of VHWs in the control and treatment groups were included in the age group 36-55 years. According to the inclusion criteria, the VHWs possessed 2-5 years of experience. In the control group, there were two male VHW members, while in the treatment group, all were women. Most of VHWs had graduated from high school. Most of VHWs had attended previous training and during the research process also received other information regarding research material from other sources. Based on the results of statistical tests (p> 0.05), the characteristics of the control and treatment groups respondents did not have a significant difference or were in the same condition. Equal conditions in the two groups before the treatment was given, are expected to describe the comparison of how far the treatment results in the two groups in this study. later these results can be generalized to a larger subject with the same conditions. Equal conditions also affect the causal relationship between the independent variable and the dependent variable.[28,29]

Table 1. Characteristics of VHW

Characteristics of VHW	Control			Treatment			*p*
	Mean ± SD	Min	Max	Mean ± SD	Min	Max	
Age (years)	43.40 ± 9.48	22	71	43.23 ± 9.37	23	59	0.934[1]
Length of time as a VHW (years)	3.48 ± 1.15	2	5	3.52 ± 1.11	2	5	0.842[2]
Characteristics of VHW		**n**	**%**		**n**	**%**	
Gender							0.247[3]
Male		2	5		0	0	
Female		38	95		40	100	
Education							0.213[2]
SD		8	20		5	12.5	
Junior High		11	27.5		10	25	
High school		19	47.5		20	50	
D3 / S1		2	5		5	12.5	
Training							0.500[3]
Yes		36	90		35	87.5	
Not		4	10		5	12.5	
Other sources of information							0.644[3]
Yes		36	90		36	90	
Not		4	10		4	10	

[1] Independent sample T-Test
[2] Mann-Whitney
[3] Chi-square
* Significant

VHW Competence before Treatment

Before VHWs were given training and facilities to learn, VHWs were first given a pre-test to see the initial scores in the two groups. The results of this study indicated that the knowledge, attitudes, self-efficacy, and VHW skills of counseling before being treated statistically have no differences between groups (p> 0.05). The absence of this significant difference indicates that the two groups possessed identical initial conditions.

VHW Competence 1 Month after Treatment

The data collection for the post-test 1 was carried out one month following the training completion, namely in the 4th week of February to the 3rd week of March 2020. Based on the results in Table 2, the statistical test showed a difference in the mean score of each variable (p <0.05) between the control group and the treatment group after 1 month of training, except for the variable of counseling skills. The difference in mean scores stated that the collaborative model training carried out for the treatment group exhibited a higher and significant increase in scores compared to the control group which was only given booklets and leaflets as learning facilities. However, in the counseling skills variable of VHWs, there were no significant differences between groups, but the change in scores in the treatment group was greater.

VHW Competence 2 Months after Treatment

Data collection for post-test 2 was carried out two months after the training was completed, namely in the 4th week of March to the 3rd week of April 2020. For the variable of VHWs counseling skills in post-test 2, observations could not be made when the VHWs conducted counseling during the *Posyandu* activities. Observations could not be executed because of the pandemic, which resulted in the elimination of *Posyandu* activities in April.

Based on the results in Table 2, the statistical test showed a difference in the mean score of each variable (p <0.05) between the control group and the treatment group after two months of training, except for the variable counseling skills. In post-test 2, the mean score for each variable also showed that the treatment group had a significantly higher score. Just like in post-test 1, this attested that training with collaborative methods provides more optimal results when compared to only providing booklets and leaflets as learning materials for VHWs. The optimal result shows that the collaborative method/learning process is one of the suitable methods for VHW learning. Collaborative learning is an educational approach to teaching and learning that involves groups of learners working together to solve a problem, complete a task, or create a product. The main characteristics of collaborative learning are a common task or activity in small group learning, cooperative behavior, interdependence, and individual responsibility and accountability.[30,31]

Table 2. **Average Competency Score of VHWs in Each Group**

VHW **Competence**	**Pretest**						
	Control			**Treatment**			
	Mean ± SD	**Min**	**Max**	**Mean ± SD**	**Min**	**Max**	***p***
Knowledge	68.38±10.7	45	85	68.0±10.73	40	85	0.876[1]
Attitude	71.95±4.94	62	82	71.87±4.76	62	82	0.945[1]
Self-efficacy	34.08±2.53	29	39	34.23±2.57	29	39	0.793[1]
Counselling Skills	70.38±12.31	45	90	70.13±11.12	50	90	0.924[1]
	Post Test 1						
Knowledge	71.88±11.41	50	95	79.87±11.17	60	100	0.002[1*]
Attitude	73.15±4.45	66	82	76.45±4.19	68	85	0.001[1*]
Self-efficacy	34.35±2.77	30	40	36.53±3.64	31	43	0.004[1*]
Counselling Skills	72.63±11.98	45	90	76.25±10.17	50	95	0.149[1]
	Post Test 2						
Knowledge	72.25±10.56	50	95	80.50±11.08	60	100	0.001[1*]
Attitude	73.45±4.26	66	82	76.78±4.1	69	85	0.001[1*]
Self-efficacy	34.65±2.87	30	41	37.30±3.52	31	44	<0.001[1*]
Counselling Skills	-	-	-	-	-	-	-

[1] Independent sample T-Test
*significant

Change in VHW Competence

Changes in the competence of VHWs in the control group and the treatment group were seen by comparing the mean values of pre-test, post-test 1, and post-test 2 in each group. The difference between each pair of data collection time (pre-test - post test1, post test1 - post-test 2, and pre-test - post-test 2) will also be seen in the comparison between the control group and the treatment group.

VHW Knowledge

The comparison of the mean scores of VHWs knowledge in pre-test - post-test 1 and pre-test - post-test 2 showed that there was a significant difference ($p < 0.05$) in the two groups, but there was no significant change ($p = 0.323$ and $p = 0.200$) in comparison of the mean score of post-test 1 to post-test 2. Likewise, based on the comparison of the delta score, the knowledge value of VHWs between groups ($p = 0.633$). The results of the different mean score test are in line with previous research which also showed a significant difference from pre-test to post-test 1 and from pre-test to post-test 2.[32]

Although both groups had a statistically significant mean score of knowledge, the mean final score in the treatment group was more notable than the control group. This result is also in line with other studies, although there were very significant differences in the two groups, the changes that occurred in the treatment group were much greater than in the control group.[33]

Attitude of VHWs

The comparison of the mean score of the attitude of the VHWs in the pre-test - post-test 1, post-test 1 - post-test 2, and pre-test – post-test 2 showed that there were significant differences in the two groups. Nonetheless, if you look at the mean score of the attitude of the VHWs in the post-test 1 and 2, the treatment group has a higher mean score than the control group. This event is in line with the results of previous research that the provision of nutrition education interventions for mothers of toddlers and *Posyandu* VHWs increased attitudes, the average nutritional attitude of VHWs was higher in the intervention group than in the control group.[34]

On the change in the mean score of the attitude of the VHWs, the comparison of the time pairs of data taking post-test 1– post-test 2 had insignificant results ($p = 0.662$). When viewed from the changes or deltas of each pair of data collection time, it can be seen that the most massive increment in value is in the difference between the pre-test and post-test 2 values in each group, especially the treatment group. This result is in line with other studies. From the pre-test and post-test, it can be seen that there is an increase in the mean attitude of *VHW*s in exclusive breastfeeding efforts in both groups.[33]

Table 3. Changes in Mean Score before and after Treatment

Change (delta)	Control	*p*	Treatment	*p*	*p*
			VHW Knowledge		
Pre – Post1	3.5 ± 3.6	<0.001[1*]	11.87 ± 5.15	<0.001[1*]	<0.001[3*]
Post1 – Post2	0.38 ± 2.37	0.323[1]	0.62 ± 3.04	0.200[1]	0.633[3]
Pre – Post2	3.88 ± 3.49	<0.001[1*]	12.5 ± 5.31	<0.001[1*]	<0.001[3*]
Pre – Post1 – Post2		<0.001[2*]		<0.001[2*]	
			Attitude of VHWs		
Pre – Post1	1.2 ± 1.36	<0.001[1*]	4.58 ± 2.01	<0.001[1*]	<0.001[3*]
Post1 – Post2	0.3 ± 0.61	0.003[1*]	0.33 ± 0.62	0.002[1*]	0.662[3]
Pre – Post2	1.5 ± 1.5	<0.001[1*]	4.9 ± 2.16	<0.001[1*]	<0.001[3*]
Pre – Post1 – Post2		<0.001[2*]		<0.001[2*]	
			VHW Self Efficacy		
Pre – Post1	0.28 ± 0.75	<0.001[1*]	2.3 ± 1.45	<0.001[1*]	<0.001[3*]
Post1 – Post2	0.3 ± 0.61	0.003[1*]	0.78 ± 0.89	<0.001[1*]	0.006[3*]
Pre – Post2	0.58 ± 0.98	0.001[1*]	3.08 ± 1.69	<0.001[1*]	<0.001[3*]
Pre – Post1 – Post2		<0.001[2*]		<0.001[2*]	
			Counselling Skills		
Pre – Post1	2.25 ± 2.52	<0.001[1*]	6.13 ± 3.84	<0.001[1*]	<0.001[3*]
Post1 – Post2	-	-	-	-	-
Pre – Post2	-	-	-	-	-
Pre – Post1 – Post2		-		-	

[1] paired samples t-test
[2] repeated measures
[3] Mann-Whitney
*significant

VHW Self Efficacy

The comparison of the mean scores of the VHWs self-efficacy in the pre-test - post-test 1. post-test 1 - post-test 2. and pre-test - post-test 2 explicated that there were significant differences in the two groups. Based on the results of different tests. the mean score of the VHW's self-efficacy in the two groups had a significant change. These results are in line with other studies that show a significant difference in self-efficacy scores after attending the training. However. if you look at the mean scores of the VHWs self-efficacy in the post-test 1 and 2, the treatment group had a higher mean score than the control group.[35]

Counseling Skills

The results of the comparison of the mean score of VHWs counseling skills in the pre-test and post-test 1 showed that there were significant differences in the two groups. The comparison of the mean score of VHWs' counseling skills at the pre-test or post-test 1 with the post-test 2 in the two groups was not calculated because the variables were not taken at that time.

Based on the different test results, the mean score of counseling skills of VHWs in the two groups had a significant change. If we see that the mean score of the post-test 1 treatment group has a higher mean score than the control group. But if you look back at table 1, the comparison of the mean score of the counseling skills of VHW between each group on the post-test 1 was not significant.

DISCUSSION

Collaborative Model and VHW Competence

VHW Knowledge

VHWs in the treatment group or those who received training using the collaborative method coupled with the provision of booklets and leaflets experienced a higher increase in knowledge compared to the control group who were only given booklets and leaflets. Based on the compared mean pre-post1-post2 score with statistical tests, it can be seen that there is a significant difference ($p < 0.001$) in the increase in the mean score of VHW knowledge of the two groups over time. Other research on collaborative models also shows that the condition of knowledge and understanding of learning citizens after participating in the application of collaborative learning models shows a significant increase. This condition is an indication that shows that the application of the collaborative learning model is quite powerful in increasing the knowledge and understanding of citizens in achieving optimal learning outcomes. The name social constructivism flows from the belief that learners construct their networks of knowledge by collaborating with others as they connect new information to their present knowledge and interests. Because each person is different, students come away from the same activity or lesson with different individual representations of the ideas studied.[36] Active learning like the collaborative model, providing students with opportunities to interact with people from a wide range of social, cultural, and ethnic backgrounds. It is often directed towards priority health needs and the redistribution of resources to specific populations and requires a synthesis of clinical skills, knowledge, capabilities, and attitudes.[21]

As stated in theory, health education in the short term can result in changes and increases in knowledge of individuals, groups, and society.[15] Likewise, the results of other studies stated that the lecture method training accompanied by discussions, simulations, and practices increased student knowledge in weighing toddlers at *Posyandu*.

Attitude of VHWs

Based on the compared mean pre-post1-post2 score with statistical tests, it can be seen that there is a significant difference ($p < 0.001$) in the increase in the mean score of an attitude of the VHW of the two groups over time. However, the increase in the mean value of the treatment group was greater than the control group. The significant increase in scores from pre-test to post-test 2 in the treatment group indicated that training using collaborative methods was optimal in improving VHWs' attitudes.

The increase in the attitude of VHWs in the treatment group was allegedly due to a stimulus in the form of support from the training facilitators (the University and West Bandung District Health Office), and village officials because of the cooperation. This statement is following the theory which asserts that attitude formation is determined by several elements including personal experience, the influence of others who are considered essential, cultural influences, mass media, educational institutions, and religious institutions as well as emotional influences.[37] External factors that are deliberately given can change human attitudes, so that consciously or not, the individual concerned will adopt a certain attitude.[38]

Learning methods that are considered effective for community-based health education are active and independent learning processes.[18] Collaborative learning is an educational approach to

teaching and learning that involves groups of learners working together to solve a problem, complete a task, or create a product.[22] This research also combines discussion methods, role modeling, and technical guidance in the field during the training process as a stimulus to get an energetic atmosphere when VHWs receive information unlike the case with the control group, which only received material in the form of booklets and leaflets.

VHW's Self Efficacy

Based on the results of statistical tests, the mean score of the VHWs' self-efficacy in the two groups had a significant change after treatment. Also, based on the compared mean pre-post1-post2 score, it can be seen that there is a significant difference ($p < 0.001$) in the increase in the mean score of VHWs' self-efficacy of the two groups over time. This is in line with the results of previous studies, that there was a change in scores in the two groups that were significantly different. The significant results up to post-test 2 indicate that the training effect can still be felt by the experimental group when the measurement is carried out.[35]

However, if you look at the mean score of the VHWs' self-efficacy in post-test 1 and 2, the treatment group has a higher mean score than the control group. A study affirmed that training and experience have a direct effect on knowledge, self-efficacy, and skills.[39] This statement is consistent with that given to the treatment group. During the training, the facilitator provided an overview of examples of counseling in *Posyandu* activities; it presented experience for VHWs and has made it easy for them to understand and retain information.

In this case, optimal learning suggests that the ability of the learning community after they receive the learning experience changes. Learning to realize that knowledge is not something fixed or finished, but always requires further elaboration and depending on the perspective taken. This condition means that there had been an increase from the previous one or what is usually occurred, due to the innovative learning model.[40] This point can be reached by developing a learning model that can increase the learning outcomes of citizens, namely through collaborative learning models.

Counseling Skills

Based on the compared mean score of counseling skills of the control group (72.63 ± 11.98) and the treatment group (76.25 ± 10.17) in Table 1, it is found that there is no significant difference ($p = 0.149$) between these groups. Thus, although the mean score of VHW skills in the treatment group was greater, this collaborative model was not yet effective in improving VHWs' counseling skills until the first month. Although based on the compared mean pretest - post-test 1 between the control and the treatment group there was a significant difference ($p < 0.001$). This significant increase in the delta is not sufficient to describe a significant increase in VHW skills in the final results of the comparison between groups. Skills are procedural matters that require repeated practice so that they can become new reflex habits that are not easily lost.[41] The direct practice method is one method that can be used to implement skills.[42]

CONCLUSION

Collaborative methods by providing booklets and leaflets to the treatment group increased the knowledge, attitudes, and self-efficacy of VHWs more than the control group, which was only given booklets and leaflets. Collaborative methods are more effective in increasing VHWs competence so that collaborative methods can be applied as an alternative training model for VHWs. However, collaborative methods have not been able to significantly improve the counseling skills of VHWs until the first month.

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REFERENCES

1. WHO. The double burden of malnutrition: Policy Brief. WHO. 2016;
2. Kementerian Kesehatan RI. Riset Kesehatan Dasar: Riskesdas 2013. Kementerian Kesehatan RI. 2013;
3. Dinas Kesehatan Provinsi Jawa Barat. Profil Kesehatan di Jawa Barat Tahun 2016. Dinas Kesehatan Jawa Barat. 2017;
4. Hijra H, Fatimah-Muis S, Kartasurya MI. Inappropriate complementary feeding practice increases the risk of stunting in children aged 12-24 months. Universa Medicina. 2016;

5. Pokharel P, Adhikari A, Lamsal P, Adhikari R. Effect of Complementary Feeding Practices and Nutritional Status of Children (6-23 months) in Tamang Community, Ambhanjyang VDC of Makwanpur. Janaki Medical College Journal of Medical Science. 2017;
6. Ismawati C. Posyandu dan Desa Siaga. Yogyakarta: Nuha Medika; 2010.
7. Rosenthal EL, de Heer H, Rush CH, Holderby LR. Focus on the future: a community health worker research agenda by and for the field. Progress in community health partnerships : research, education, and action. 2008;
8. Swider SM. Outcome effectiveness of community health workers: An integrative literature review. Public Health Nursing. 2002.
9. Brownstein JN, Chowdhury FM, Norris SL, Horsley T, Jack L, Zhang X, et al. Effectiveness of Community Health Workers in the Care of People with Hypertension. American Journal of Preventive Medicine. 2007.
10. Brownstein JN, Bone LR, Dennison CR, Hill MN, Kim MT, Levine DM. Community health workers as interventionists in the prevention and control of heart disease and stroke. American Journal of Preventive Medicine. 2005.
11. Lewin S, Babigumira S, Bosch-Capblanch X, Aja G, van Wyk B, Glenton C, et al. Lay health workers in primary and community health care: A systematic review of trials. Lay Health Workers. 2006;
12. Pradana D. Pengaruh Pendidikan Kesehatan Oleh Kader Terhadap Praktek Ibu Dalam Pemberian Mp-Asi Di Wilayah Kerja Puskesmas Ledokombo Kabupaten Jember. Universitas Jember; 2012.
13. Vaughan JP, Walt G. Village health workers, and primary health care. Tropical Doctor. 1983;
14. Yumianti. Faktor-Faktor Yang Mempengaruhi Keaktifan Ibu Balita Berkunjung Ke Posyandu Delima Mekar 03 Di Desa Karangdadap Kec. Kalibagor, Kab. Banyumas. Universitas Muhammadiyah Purwokerto; 2014.
15. Notoatmodjo S. Pendidikan Dan Perilaku Kesehatan. Jakarta: Rineka Cipta; 2003.
16. Bandura A, Caprara GV, Barbaranelli C, Pastorelli C, Regalia C. Sociocognitive self-regulatory mechanisms governing transgressive behavior. Journal of Personality and Social Psychology. 2001;
17. Baranowski T, Cheryl P, Guy SP. How individual. environment and health behavior interact, social cognitive theory in Text Book Health Behavior and Health Education. 2nd ed. California: Jossey-Bass Inc; 1997.
18. Hariyanti T, Prabandari YS. Efektivitas Metode Kolaboratif Learning dan Kooperatif dalam Pendidikan Kesehatan Berbasis Komunitas. Jurnal Pendidikan Kedokteran Indonesia. 2014;3(1):9–17.
19. Rusmana A, F Boesoeri S, Khadijah ULS. Pendidikan Pola Hidup Sehat Melalui Literasi Kesehatan Di Kecamatan Lembang Kabupaten Bandung Barat. Dharmakarya: Jurnal Aplikasi Ipteks untuk Masyarakat. 2017;6(3):153–5.
20. Tanbrin NNI. Program Pelatihan Intervensi Dini Bersumberdaya Keluarga Bagi Kader Rehabilitasi Berbasis Masyarakat Di Desa Lembang Kabupaten Bandung Barat. Universitas Pendidikan Indonesia; 2020.
21. Mennin S, Petroni-Mennin R. Community-based medical education. Clinical Teacher. 2006;
22. Laal M, Laal M. Collaborative learning: What is it? In: Procedia - Social and Behavioral Sciences. 2012.
23. Risman Z. Pengaruh Model Pembelajaran Kolaboratif Terhadap Hasil Belajar IPA Peserta Didik Kelas V MIS DDI Bosalia KAB. Jeneponto. Universitas Islam Negeri (UIN) Alauddin Makassar; 2017.
24. Jones KA, Jones JL. Making Cooperative Learning Work in the College Classroom : An Application of the 'Five Pillars' of Cooperative Learning to Post-Secondary Instruction. 2008;8(2):61–76.
25. Beard C, John P. Experiential Learning: A Best Practice Handbook for Educators and Trainer. Kogan Page Publishers; 2006.
26. Madarisa F. Pelatihan Kolaboratif Untuk Perbaikan Kompetensi Peternak Sapi di Kabupaten Pasaman Barat. Universitas Andalas; 2016.
27. Schwarzer R, Jerusalem M. Generalized self-efficacy scale. Measures in health psychology: A user's portfolio. 1995;
28. Plutchik R. The nature of emotions: Human emotions have deep evolutionary roots, a fact

that may explain their complexity and provide tools for clinical practice. American Scientist. 2001;
29. Mumford MD, Hester KS, Robledo IC, Peterson DR, Day EA, Hougen DF, et al. Mental Models, and Creative Problem-Solving: The Relationship of Objective and Subjective Model Attributes. Creativity Research Journal. 2012;
30. Chandra R. Collaborative Learning for Educational Achievement. IOSR Journal of Research & Method in Education (IOSR-JRME). 2015;
31. Panitz T. Collaborative versus Cooperative Learning: A Comparison of the Two Concepts Which Will Help Us Understand the Underlying Nature of Interactive Learning. Cooperative Learning and College Teaching. 1997;
32. Sukiarko E. Pengaruh Pelatihan Dengan Metode Belajar Berdasarkan Masalah Terhadap Pengetahuan Dan Keterampilan Kader Gizi Dalam Kegiatan Posyandu Studi Di Kecamatan Tempuran Kabupaten Magelang. Universitas Diponegoro; 2007.
33. Jumiyati, Nugrahaeni S, Margawati A. Pengaruh Modul Terhadap Peningkatan Pengetahuan, Sikap Dan Praktek Kader Dalam Upaya Pemberian Asi Eksklusif. Gizi Indonesia. 2014;37(1):19–28.
34. Nikmawati E, C KM, Khomsan A, Sukandar D, Atmawikarta A. Intervensi pendidikan gizi bagi ibu balita dan kader Posyandu untuk meningkatkan PSK (Pengetahuan Sikap dan Keterampilan) serta status gizi balita. Jurnal Pendidikan Teknologi Kejuruan. 2009;5(15):407–17.
35. Ardiyanti D, Alsa A. Pelatihan " PLANS " untuk Meningkatkan Efikasi Diri dalam Pengambilan Keputusan Karir. Gadjah Mada Journal Of Propfesional Psychology. 2015;1(1).
36. Jacobs GM. Collaborative Learning or Cooperative Learning? Beyond Words. 2015;3(1).
37. Strachan DL, Källander K, Ten Asbroek AHA, Kirkwood B, Meek SR, Benton L, et al. Interventions to improve motivation and retention of community health workers delivering integrated community case management (iCCM): stakeholder perceptions and priorities. American Journal of Tropical Medicine and Hygiene. 2012;
38. Gisore P, Rono B, Marete I, Nekesa-Mangeni J, Tenge C, Shipala E, et al. Commonly cited incentives in the community implementation of the emergency maternal and newborn care study in western Kenya. African Health Sciences. 2013;
39. Partiprajak S, Thongpo P. Retention of basic life support knowledge, self-efficacy and chest compression performance in Thai undergraduate nursing students. Nurse Education in Practice. 2016;
40. Elbers E, Streefland L. Collaborative learning and the construction of common knowledge. European Journal of Psychology of Education. 2000;
41. Sugiarto I. Mengoptimalkan Daya Kerja Otak dengan Berpikir Holistik dan Kreatif. Jakarta: Gramedia Pustaka Utama; 2011.
42. Herijulianti E, Indriani T, Artini S. Pendidikan Kesehatan Gigi. Jakarta: EGC; 2001.