THE EFFECT OF ADDITIONAL DADIH ON LACTIC ACID BACTERIA AND NUTRITIONAL VALUE OF PUDDING AS A FOOD SUPPLEMENTATION

HELMIZAR^{1*}, AZRIMAIDALIZA¹, RESTU SAKINAH¹ and ARIEF YANDRI¹

¹Department of Nutrition, Faculty of Public Health, Andalas University St. Perintis Kemerdekaan No. 94, Padang, Indonesia *E-mail: helmizar@ph.unand.ac.id

Accepted 7 October 2022, Published online 31 October 2022

ABSTRACT

Dadih is a dairy product obtained from fermented buffalo milk in West Sumatra, Indonesia. It contains various nutrients and probiotics that are beneficial for prenatal supplementation and pregnancy outcomes. In this study, a pudding enriched with *dadih* was formulated. The study was conducted in November-December 2019 at the Nutrition Laboratory Andalas University. The design of this study was a True Experiment using a Completely Randomized Design (CRD) with two replications consisting of four formulas. They were F0, F1, F2, and F3 with the addition of *dadih* at 0, 80, 90, and 100 g, respectively. Analysis of nutritional value and the number of Lactic Acid Bacteria (LAB) was performed. Based on the nutritional value analysis, 100 g of *dadih* pudding contains 11.59% of carbohydrate, 1.24% of protein, 1.09% of fat, with a total of 61.13 calories for F0; 10.41% of carbohydrate, 2.29% of protein, 3.29% of fat, with a total of 80.41 calories for F1; 10.12% of carbohydrate, 4.74% of protein, 3.32% of fat, with a total 89.32 calories for F2; and 11.84 % of carbohydrate, 3.05% of protein, 3.13% of fat, with a total 87,73 calories for F3. The result of the LAB test of *dadih* pudding showed that: F0 at 3.1 × 10³ (CFU/g); F1 at 1.7 × 10° (CFU/g); F2 at 2.4 × 10° (CFU/g), and F3 at 2.4 × 10° (CFU/g). While the result of the Kruskall Wallis test showed that there was a significant difference between the hedonic test and the hedonic quality test of four *dadih* pudding formulas (p<0.000). The chosen formula was F3 with the highest nutrition value average of 87.73 kcaL total calories. *Dadih* pudding is recommended as a food supplementation because it is good for the digestive system and can increase the immune system of pregnant women.

Key words: Dadih pudding, lactic acid bacteria, nutritional value, pregnant women

INTRODUCTION

Nutrition is an important factor that determines the quality of human health and well-being. A person's nutritional status is counted as good when their physical and mental development is in balance. The optimal level of nutritional status will be achieved if optimal nutrient intake is fulfilled. The nutritional status of a person at a time is not only determined by the nutrient consumed but is more determined by the previous consumption (Wirayo, 2002). Poor nutritional status in pregnant women will cause incomplete fetal growth, low birth weight, and miscarriage. The optimal nutrients consumed during pregnancy greatly impact the development and fetal growth. The first 1000 days of a child's life, is a golden period for child growth and development (Bellieni, 2016). Many women go through their entire pregnancy without reaching the minimum intake of micronutrients (Aji & Yerizel, 2019). Therefore, one of the Government programs related to balanced nutrition is "The First 1000 Days of Life" to achieve

Related to the topic discussed in the above paragraph, it is strongly recommended that pregnant women should consume 3000 calories every day consisting of 55-75% of carbohydrates, 10-15% of protein, 20-30% of fat, 200 µg of folic acid, 250-300 mg of calcium and 35 mg of iron (Fikawati & Wahyuni, 2012). Commonly, poor nutrition during pregnancy leads to micronutrient deficiency status (Farias, 2020). Long-term undernutrition during pregnancy will cause the mother to experience a condition of Chronic Malnutrition (Nurrmadinisia, 2012). The fact showed that in 2018, there were 17.3% of pregnant women experienced chronic malnutrition in Indonesia. Meanwhile, in West Sumatra, there were 17% of pregnant women experienced the same disease (Litbangkes, 2018). To meet the nutritional needs of pregnant women, it is necessary to provide additional food during pregnancy, this strategy is used to overcome nutritional problems. The nutritional content in one pack of biscuits as additional food from the government (3 pieces/60 g) contains 6 g of protein, 12 g of fat, 11 kinds of vitamins, and 7 minerals, with

optimal growth and development of the fetus.

^{*} To whom correspondence should be addressed

a total of 270 calories.

West Sumatra has special foods that can be used as food supplementation. One of them is dadih. This traditional food is rich in probiotics so that it can be developed into functional food (Usmiati & Risfaheri, 2013). Though manufacturing is a traditional process and has not fulfilled any national or international standards for yogurt and fermented milk, dadih provided safety and health for people who consumed it (Kechagia et al., 2013). There are many studies and publications regarding the benefits of probiotics for human health, both in vivo studies in experimental animals and clinical studies in human groups. Nowadays, Asian and European people use probiotics as health food and pharmaceuticals, which subsequently increases the demand for these probiotic sources (Kasimin et al., 2020). The mechanism of action behind the health benefits is largely unknown, but several probiotic strains have been isolated from dadih, which may contribute to its health properties (Venema & Surono, 2018). Several studies had demonstrated that Dadih has beneficial effects on the intestinal health of mothers and children due to its nutritional value and its role as a probiotic (Balqis et al., 2018). For pregnant women, it can lower the abortus rate and can increase immunity during pregnancy (Purwati, 2018). In addition, dadih also contains good Lactic Acid Bacteria (LAB) which can convert carbohydrates (glucose) into lactic acid. Besides, LAB is also used in the food fermenting process, acting as a food preservative, improving texture, and adding to the taste of food (Helmizar et al., 2019). The nutritional content of dadih includes 9.96% of protein, 9.89 % of fat, 73.02% of water, and pH 5.02, with the amount of LAB 1.70×106 CFU/g. In addition, dadih also contains calcium and vitamin B complex which is from milk, including vitamins B and K which are formed during the fermentation process (Helmizar et al., 2019).

The objectives of this study are aimed at developing *dadih* into several formulations by adding it to pudding. The next step is to identify the nutritional value, of Lactic Acid Bacteria (LAB), and then conducted an organoleptic test to get the best pudding formula to be consumed as a food supplementation

for pregnant women.

MATERIALS AND METHODS

Materials

The design of this research is a true experiment, which applies a completely randomized design (CRD) with two replications consisting of 4 levels of F0, F1, F2, and F3. In this study, F0 functions as a standard formula which will be compared to F1, F2, and F3. In this stage, dadih is not added to F0, meanwhile, it is added to F1 (80 g), F2 (90 g), and F3 (100 g) as shown in Table 1. Furthermore, a proximate test or analysis of nutritional content was carried out to see the nutritional value (carbohydrates, protein, fat) in each formula. Afterward, the LAB test was carried out on the formulas (F0, F1, F2, & F3). The hedonic and hedonic quality tests were carried out to assess the quality and preference of each formula. In this test, the researchers selected 30 semi-trained panelists. The panelists are nutrition major students who had received theory and training regarding the organoleptic test. Afterward, based on the highest score on the hedonic test as a representative of the panelists' acceptance of dadih pudding, the selected formula is determined.

Dadih pudding was made by modifying the recipe made by Helmizar (2019). Thus, there was no starter used in making dadih pudding in this study. Pudding powder that has the ingredient of gelatin and vanilla flour was mixed with the water, and the mixture was heated and stirred until it boiled. The pudding was cooled until 60–70 °C and then mixed well with dadih. Dadih pudding was poured into the pudding mold and chilled in the refrigerator for it to be ready to serve (Taufiqa et al., 2021).

Hedonic tests and hedonic quality tests which were carried out on *dadih* pudding are based on color, aroma, taste, and texture. Meanwhile, the proximate test was carried out at Padang Industrial Baristand Laboratory, and the LAB test was carried out <u>in</u> the Pharmacy Laboratory of Dharma Andalas University. The hedonic test and hedonic quality of *dadih* pudding were carried out in the Nutrition Laboratory of Public Health Faculty, Andalas University.

Table 1. The material composition of the dadih pudding formula

Material type	Material weight in each formula				
Material type	F0	F1	F2	F3	
Dadih (g)	0	80	90	100	
Agar Flour (g)	5	5	5	5	
Cornstarch (mL)	15	15	15	15	
Sugar (g)	25	25	25	25	
Milk (mL)	150	150	150	150	
Water (mL)	150	70	60	50	

The nutrition analysis of dadih pudding in terms of proximate compositions was analyzed referring to the standard method. The protein content was determined by using the Kjedal method. The total lipid was extracted with hexane by using the Soxhlet method. Moisture value was evaluated by using the oven drying method, and the ash content was determined by using the furnace incineration method (Horwitz, The total carbohydrate was calculate by difference, whereby, % total carbohydrate = 100% - % (moisture + ash + crude protein + crude fat) (BeMiller & Low, 1998). The calories of *dadih* pudding were calculated according to the equation developed by Nielsen (1998). That is every 1 g carbohydrate is equivalent to 4 kcaL, 1 g protein equals 4 kcaL, and 1 g fat is equivalent to 9 kcaL. Kruskal Wallis test was used to know if there were significant differences between the hedonic and hedonic quality test of four *dadih* pudding formulas (p<0.05).

RESULTS AND DISCUSSION

Nutritional value of dadih pudding

The proximate test is one of the determining factors in product manufacture because the proximate test will show the nutritional value of a product, and will also affect the quality of a product. In the *dadih* pudding product, the test carried out were carbohydrates, protein, fat, and energy. This can be seen in Table 2.

The carbohydrate content of each formula is different, except for F0 as a standard formula. So, it should be noted that with the increase of dadih addition, the carbohydrate content also increases. In the standard formula (F0) the carbohydrate content was 11.59%, the carbohydrate came from cornstarch and sugar. In F1 the carbohydrate content was 10.41%, and the amount of cornstarch and sugar used was the same as in F0 but there was a decrease in carbohydrate content which should have increased because dadih also contains carbohydrates. The carbohydrate content of F2 was 10.12%. Compared to F1, the carbohydrate content was lower, it should be higher than F1 because more amount of dadih was added. This may be caused by inaccurate weight because the difference between dadih's amount was

only 10 g. This was because the scale used was the food scale, not the analytic scale. Another possibility was that the temperature and cooking time for the pudding was not the same. Consequently, the total weight of the pudding was also different, because the amount of water was reduced when boiled at different temperatures and times. Based on Table 2, in F3 there was an increase in carbohydrate content compared to F1 and F2, in which the carbohydrate content of F3 was 11.84%.

During pregnancy, an addition of 25-40 g of carbohydrates is required per day. Based on the results of the calculation of the organoleptic test score for *dadih* pudding, formula F3 was chosen as the best and most preferred formula. The carbohydrate analysis in the selected *dadih* pudding formula was 11.84% or equal to 11.84 per 100 g of *dadih* pudding. Thus, to meet the need for additional carbohydrates during pregnancy, it is recommended to consume as much as 300-400 g of *dadih* pudding/day.

The result of standard pudding protein content was 1.24%, F1 *dadih* pudding contains 2.29% protein, F2 4.74%, and F3 3.05%. The protein content in the *dadih* pudding formula between F0 and F1 was increased, in this case, it was probably a result of the *dadih* addition to the pudding formula, there was also an increase in protein content between F1 and F2 due to differences in the number of additional *dadih* while between F2 and F3 there was a decrease in protein content, presumably the result of the cooking process which causes protein damage and a reduction in protein content of *dadih* pudding. The protein content difference only comes from the different amounts of *dadih* added. Based on the theory, 10 g of *dadih* contains 0.7% protein.

During pregnancy, an addition of 10-20 g of protein per day is required (Ministry of Health 2019). Based on the results of the calculation of the organoleptic test score for *dadih* pudding, formula F3 obtained the chosen formula. The protein analysis in the selected *dadih* pudding formula is 3.05% or equal to 3.05 g per 100 g of *dadih* pudding. Thus, to meet the need for additional protein during pregnancy, it is recommended to consume as much as 300-400 g of *dadih* pudding/day.

The total fat content in the dadih pudding formula

Table 2. Nutritional value of dadih pudding per 100 g

Took Donous stone	Unit	Analysis results			
Test Parameters		F0	F1	F2	F3
Carbohydrate	%	11.59	10.41	10.12	11.84
Protein	%	1.24	2.29	4.74	3.05
Fat	%	1.09	3.29	3.32	3.13
Calories	kcal	61.13	80.41	89.32	87.73

Information: F0 = standard formula, F1 = pudding formula with the addition of *dadih* (80 g), F2 = pudding formula with the addition of *dadih* (90 g), and F3 = pudding formula with the addition of (100 g).

varies; the standard formula (F0) fat was 1.09%, F1 3.29%, F2 3.32% and F3 3.13%. The difference between F0 and F1 fat content comes from dadih, and the difference between F1 and F2 and F3 fat content comes from the amount of dadih added. Based on theory, 10 g of dadih contains 0.8% fat. Based on the results obtained, there was a decrease in fat content by 0.19% between F2 and F3. The fat content of F3 should be higher than F2 because the amount of dadih added to F3 was more than F2. This happened because the cooking process caused a decrease in fat. The fat content at F0 was 18.63%, at F1 was 26.55%, at F2 was 25.15%, and at F3 was 22.32% of what it should have been, which means that the average loss of the nutritional content of the ingredients for making dadih pudding was 75%.

According to the Indonesian recommended dietary allowance (RDA) for pregnant women, 6-10 g addition of fat is required per day (Ministry of Health, 2013). Based on the calculation of the organoleptic test score for *dadih* pudding, fat analysis in the selected *dadih* pudding formula (F3) was 3.13% or equal to 3.13 g per 100 g of *dadih* pudding. Thus, to meet the need for additional fat during pregnancy, it is recommended to consume as much as 300-400 g of *dadih* pudding/day.

Based on Ministry of Health Indonesia 2017 data, the nutritional value of food supplementation for pregnant women contains a minimum of 270 calories, a minimum of 13 g of carbohydrates, 6 g of protein, and a minimum of 12 g of fat. From the calculation of the nutritional value of dadih pudding, to be compared with the food supplementation for pregnant women from the government (layer biscuits), dadih pudding produces 300 g of final weight or as much as 2 cups of pudding. During the 1st trimester of pregnancy, a pregnant woman requires an additional 180 calories of energy, 25 g of carbohydrates, 20 g of protein, and 6 g of fat (Ministry of Health, 2013). In the 2nd and 3rd trimesters, pregnant women need approximately additional energy of 300 calories, 40 g of carbohydrates, 20 g of protein, and 10 g of fat. To meet the additional needs of a pregnant woman in the 1st trimester, it is recommended to consume 200

g of *dadih* pudding per day (160 calories) and in the 2nd and 3rd trimesters it is recommended to consume 400 g of *dadih* pudding per day, or a pregnant woman should consume one to two cups of *dadih* pudding.

Lactic Acid Bacteria (LAB)

The number of lactic acid bacteria was determined by using *deMannRogosa Sharpe* Agar (MRSA) media. In the LAB test, the number of lactic acid bacteria colonies was calculated in each formula, the results of the LAB test in this study can be seen in Table 3.

Table 3. lactic acid bacteria test of dadih pudding

Formula	Number of Colonies (CFU/g)
F0	3.1 × 10 ³
F1	1.7 × 10°
F2	2.4 × 10°
F3	2.4 × 10 ⁹

Dadih has the amount of LAB in correspondence to SNI 281: 2009 that the minimum amount of LAB in fermented milk is 1 × 10⁷ CFU/g. In previous research, the LAB content of 100 g of dadih fermented for 2 × 24 h was 1.70 × 10⁶ CFU/g (Daswati, Hidayati, & Elfawati, 2009). Based on the LAB analysis on the dadih pudding formula, as shown in Figure 1, F0 as the standard formula with no additional dadih, contains 3.1 × 10³ CFU/g, F1 contains 1.7 × 10⁹ CFU/g, F2 contains 2.4 × 10⁹ CFU/g, and F3 contains 2.4 × 10⁹ CFU/g. However, the amount of viable LAB fulfilled the concentration of probiotics needed to obtain a clinical effect is often quoted as 10⁶ CFU/mL in the small bowel and 10⁸ CFU/g in the colon (Hill et al., 2014).

The result of a similar study found that total LAB in the original dadih (6.4 × 10° CFU/mL) was almost the same as those in $Mango\ Dadiah$ (MD) pudding (6.1 × 10° CFU/mL), whereas they were reduced (2.4 × 10° CFU/mL) in Chocolate Dadiah (CD) pudding. A decrease of 62.5% of total LAB in CD as compared to the original dadiah may be due to the chocolate powder used as the additional ingredient in the pudding (Taufiqa $et\ al.$, 2021).



Fig. 1. Isolated colonies of LAB from samples cultured in de mann, rogosa, sharpe (mrs) agar medium after incubation for 48 h at 37 °C.

The organoleptic test

The organoleptic test is one of the important parameters to determine the panelists' acceptance of a food product. For this reason, organoleptic tests were carried out, including the hedonic test (preference) and the hedonic quality test. The results of the hedonic test are rated on a scale of 1 to 7, with the levels of really dislike, dislike, somewhat dislike, ordinary, somewhat like, like, and like.

The results of the data analysis of the panelists' preference for *dadih* pudding in terms of taste and texture were still not homogeneous, while the color and aroma were homogeneous. Because there were still non-homogenous data mentioned above, the Kruskal-Wallis test was applied as seen in Table 4.

The color, aroma, taste, and texture of *dadih* pudding have average values of 4.20, 4.91, 4.56, and 4.54 in the normal category. F3 has the highest average value in the treatment formula, namely 5.03, 4.57, 4.07, and 4.70, with slightly like categories. Based on the result of the statistical test on formula treatment, F1, F2, and F3 that founded were significantly different, namely $p \le 0.05$. It means that the three formulas in terms of color, aroma, taste, and texture were somewhat preferred.

The hedonic quality of the *dadih* pudding are selectable from very sour, sour, slightly sour, medium, slightly sweet, sweet, and very sweet, and the hedonic quality of the *dadih* pudding texture are very hard, hard, slightly hard, medium, slightly soft, soft, very soft. The results of the hedonic quality tests' data analyzed of *dadih* pudding in terms of aroma are still not homogeneous. So it is followed by the Kruskal-Wallis test, the results of the Kruskal-Wallis test can be seen in Table 4.

Good quality *dadih* has creamy-white color, smooth and glossy surface, good consistency, milk-like aroma, and acidic taste (Surono, 2015). Based on the result of the hedonic quality statistical test on formula treatment, the panelists prefer the slightly bright in terms of color, moderate in terms of aroma, slightly sour in terms of taste, and somewhat soft in terms of texture.

Selected formulas

Based on Table 5, it can be seen that F3 has the highest score (18.37) while F2 has the lowest score (14.93). Based on the result of the calculation, the F3 is determined as the selected formula.

CONCLUSION

The nutritional value of 100 g of *dadih* pudding is 11.84% of carbohydrates, 3.05% of protein, and 3.13% of fat. Therefore, pregnant women in the first trimester need 200 g of *dadih* pudding, while in the second and third trimesters 400 g of *dadih* pudding. The amount of LAB in the *dadih* pudding of the three formulas is described as follows: F1 1.7 × 10 ° CFU/g, F2 2.4 × 10° CFU/g, and F3 2.4 × 10° CFU/g. Based on the organoleptic test of *dadih* pudding, the best and preferred formula is the F3 with the addition of 100 g of *dadih*. Therefore, the provision of *dadih* pudding can be applied consistently and regularly as a food supplement for pregnant women during pregnancy.

ACKNOWLEDGEMENTS

The authors would like to acknowledge the support of the Faculty of Public Health at Andalas University

Table 4. Kruskal-Wallis test of hedonic and hedonic quality test of dadih pudding

	Hedonic Test			Hedonic Quality Test		
	Mean	<i>p</i> -value	Information	Mean	p-value	Information
Colour	4.20	0.000	Ordinary	4,82	0,001	Rather bright
Aroma	4.91	0.003	Somewhat like	4,30	0,000	Moderate
Taste	4.56	0.001	Somewhat like	3,50	0,000	Slightly sour
Texture	4.54	0.000	Somewhat like	4.98	0.000	A little soft

Note: The hedonic test rating scale for the color of *dadih* pudding is 1 = really dislike, 2 = dislike, 3 = somewhat disliked, 4 = ordinary, 5 = somewhat like it, 6 = like it and 7 = really like it

Table 5. Scores of the hedonic test dadih pudding formula

Parameter	Average Value				
	F0	F1	F2	F3	
Color	5.60	4.60	4.40	5.03	
Aroma	5.47	4.17	4.03	4.57	
Taste	5.47	3.97	3.30	4.07	
Texture	5.97	4.30	3.20	4.70	
Total Score	22.51	17.04	14.93	18.37	

Information: F0 = standard formula without adding dadih, F1 = 80 g of dadih addition, F2 = 90 g of dadih addition and F3 = 100 g of dadih addition.

for providing the facilities and financial support for this research.

ETHICAL STATEMENT

This study was approved by the ethical committee of Medical Faculty Andalas University, number 372/UN.16.2/KEP-FK/2021

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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