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Lactic Acid Levels Yogurt Red Beans with Addition of Honey Trigona sp

Ruri Widyati*, Leka Lutpiatina, Neni Oktiyani, Haitami

Medical Laboratory Technology Poltekkes Kemenkes Banjarmasin
Jl Mistar Cokrokusumo Street 4a Banjarbaru, Indonesia.
e-mail: ruriwidy@gmail.com

Abstract. Red beans (Phaseolus vulgaris L.) are grains that are rich in nutrients and useful as a substitute for vegetable milk which can be processed into yogurt using Streptococcus thermophillus and Lactobacillus bulgaricus. Carbohydrates in red beans consist of a group of oligosaccharides that have little ability as an energy source for bacteria to produce lactic acid. Therefore, the making of red bean yogurt needs another source of sugar by adding Trigona sp. Honey. The purpose of this study was to determine the effect of adding Trigona sp honey by 0%, 1.9%, 3.8%, 5.7%, 7.6%, 9.5% to the total level of lactic acid in red bean yogurt incubated for 24 hours at 37oC. This type of research is an experiment with the Postest Only With Control Group Design research design using the alkalimetry titration method. The sampling technique used was purposive sampling technique, namely Trigona sp honey bee taken at Trigona sp bee farm in Tambangan, Tanah Laut. Indonesia. The results showed that there was an effect of adding Trigona sp honey to total lactic acid levels in red bean yogurt with the addition of Trigona sp honey 1.9% - 9.5% fulfilling the requirements of SNI 01.2981-2009. The highest average value of total lactic acid in red bean yogurt with the addition of Trigona sp honey 9.5% which is equal to 0.6672%. It is recommended for future researchers to use other types of carbohydrate sources and additives in the form of proteins. For the community, it is recommended to use alternative sources of other sugar and optimize the addition of Trigona sp. Honey.
Keywords: red beans; yogurt; honey; lactic acid

INTRODUCTION

Yogurt is a product obtained from pasteurized cow's milk and then fermented with bacteria that produce acid, with a distinctive taste and thick texture. The principle of making yogurt is fermented milk by adding lactic acid bacteria such as Streptococcus thermophilus and Lactobacillus bulgaricus. The addition of lactic acid bacteria will convert lactose to milk into lactic acid so that the pH will drop. Decreasing pH or increasing acid will cause milk protein to coagulate to form a yogurt texture thick with distinctive aroma and taste[1].

Yogurt contains live bacteria as probiotics, which are microbes from foods that are beneficial for microflora in the digestive tract. By far the most common types of probiotics are lactic acid bacteria from the Lactobacillus bulgaricus, Streptococcus thermophilus, and Lactobacillus casei groups. Yogurt usually contains millions to billions of cells of these

bacteria every milliliter. The presence of many bacteria in yogurt is indeed related to the manufacturing process[2]. Streptococcus thermophilus grows at a temperature of 43-45°C and produces titrated acid 0.9-1.0%; whereas for Lactobacillus bulgaricus it grows at a temperature of 43-46°C and produces 2.0-4.0% titrated acid[3].

The benefits of consuming yogurt alone include the source of probiotic bacteria for the intestine and as an antioxidant to counteract free radicals. Currently yogurt has developed in the manufacturing process to produce yogurt with many variations[4].

Bean seeds are a source of protein for most of the world's population, especially for people in developing countries like Indonesia. Even the consumption patterns of people have shifted from animal foods to plant-based foods. Animal milk can increase cholesterol levels so it is not recommended to consume excessively, especially for someone who suffers from certain diseases and allergies.

The material used in this study is red bean milk. Red beans are common in Indonesia at a relatively affordable price. Red bean is a local food that has the potential as a raw material for fermented milk because its nutritional content is quite complete. Red beans have almost the same protein as meat protein and are a high source of folic acid. Red bean (Phaseolus vulgaris L.) is a good source of protein plants with a protein content of around 23.1%, complex carbohydrates, fiber, vitamin B1, calcium, phosphorus, iron, and folasin[5].

The type of carbohydrates in red bean milk is very different from the carbohydrates found in cow's milk. Carbohydrate in red bean milk itself is a group of oligosaccharides, this causes the sugar content used by microorganisms in making yogurt is very limited, therefore it is necessary to add another sugar as a carbon source [5,6]. Honey is chosen as an additional source of sugar because it has sucrose 6.175%[7]. Honey also has antioxidant content[8] and has the ability to improve body fitness[9].

Red beans have the potential as a substitute for animal milk sources to be used as fermented milk by adding honey as a source of sugar in order to get good quality yogurt based on measurements of total lactic acid levels. Therefore researchers are interested in conducting research on the Effect of Addition of Honey Trigona sp to Total Lactic Acid Levels in Red Bean Yogurt. This study aimed to determine the effect of adding Trigona sp honey to total lactic acid levels in red bean yogurt.

MATERIALS AND METHODS

This research is using experimental method. This type of research is the most ideal for studying the causal mechanism, because almost all sources of invalidity can be well controlled by design.

The research design used in this study is Postest Only With Control Group Design, which measures the effect of treatment in the experimental group by comparing these groups with the control group. In this study an examination of the effect of adding Trigona sp honey on total lactic acid levels in red bean yogurt with a concentration of 0%, 1.9%, 3.8%, 5.7%, 7.6%, 9.5%. The number of samples in this study were 25 samples, which consisted of 5 treatments and 5 repetitions for each treatment.

The materials in this study were red beans (Phaseolus vulgaris L.) and Trigona sp honeybee taken at beekeeping in Tambangan, Tanah Laut. Indonesia. By means of sampling using purposive sampling technique. The study used analytical balance (Acis AD 600i), Buret (Iwaki) starter Lactobacillus bulgaricus and Streptoccocus thermophillus,

the independent variable was the addition of Trigona sp honey with concentrations of 0%, 1.9%, 3.8%, 5.7%, 7, 6%, 9.5%. The dependent variable is the total level of lactic acid in red bean yogurt.

RESULTS AND DISCUSSION

Making red bean milk from 750 gr which is soaked for 12 hours, mashed in 3 liters of distilled water with a ratio of red beans and distilled water 1: 4. Red bean milk on the hotplate to a temperature of ± 70oC for 15 minutes. Making the concentration of Trigona sp honey 0%, 1.9%, 3.8%, 5.7%, 7.6%, 9.5% can be seen in the table below:

Table 1. Concentration of honey bee Trigona sp

Concentration (%)	ml honey bee	ml of red bean milk	ml of bacterial starter
0	0	100	5
1,9	2	98	5
3,8	4	96	5
5,7	6	94	5
7,6	8	92	5
9,5	10	90	5

Inoculation of yogurt bacteria starter is carried out at 30oC, then stirred evenly and tightly covers with cotton, then coated with aluminum foil and plastic wrap. Incubated at 37oC with 24 hours fermentation time. Then an analysis of the red bean yogurt is carried out.

Standardize 0.1 N NaOH solution using H2C2O4.2H2O 0.1 N. Examination of total lactic acid levels from 10.0 grams of yogurt samples added to 100 ml aquadest and 3 drops of 1% PP indicator. Titrate the sample with 0.1 N NaOH until the reddish color and color do not disappear for 30 seconds (constant).

Calculation:

$$\% \text{ lactic acid} = \frac{(V \times N)\text{NaOH} \times \text{BE lactic acid}}{\text{Material weight (gram)} \times 1000} \text{ X } 100\%$$

Table 2. Results of alkalimetric titration

No	Concentration of Trigona sp. Honey (%)	Volume of end point of titration (ml)				
		1	2	3	4	5
1	0	5,3	5,2	5,3	5,2	5,2
2	1,9	6,9l	6,7	7,2	7,0	6,7
3	3,8	7,0	7,5	7,3	6,9	7,1
4	5,7	7,2	7,4	7,3	7,2	7,0
5	7,6	7,3	7,1	7,0	7,6	7,3
6	9,5	7,4	7,1	7,5	7,4	7,7

Table 3. Total lactic acid in red bean yogurt with various concentrations of Trigona sp. Honey.

NO	Concentration of Trigona sp. Honey (%)	Total lactic acid in red bean yogurt (%)					Average (%)
		1	2	3	4	5	
1	0	0,473	0,472	0,477	0,470	0,473	0,4730
2	1,9	0,619	0,601	0,645	0,603	0,629	0,6194
3	3,8	0,629	0,674	0,656	0,620	0,638	0,6434
4	5,7	0,647	0,656	0,665	0,630	0,647	0,6490
5	7,6	0,657	0,639	0,630	0,684	0,656	0,6532
6	9,5	0,665	0,638	0,674	0,666	0,693	0,6672

Table 3. shows the total levels of lactic acid in red bean yogurt with the addition of Trigona sp. Honey. in various concentrations. In this study the group without the addition of Trigona sp honey had the lowest total lactic acid level, which was an average of 0.4730%, while the increase in total acid content was seen in the addition of Trigona sp honey with concentration, 1.9%, 3.8%, 5.7%, 7.6%, 9.5% with the average total lactic acid levels respectively 0.6194%, 0.6434%, 0.6490%, 0.6532% and 0.6672%. And the highest total lactic acid level was seen in the concentration of Trigona sp honey 9.5%. Thus the total level of lactic acid increases with increasing concentration of Trigona sp honey added to red bean yogurt.

Based on Table 3. can be graphed the total levels of lactic acid in red bean yogurt with the addition of Trigona sp honey as

This study was conducted to find the effect of the independent variables in the form of the concentration of Trigona sp honey with the dependent variable, the total level of lactic acid. Analysis of the data is done quantitatively using linear regression test.

A good regression model is a regression model that has normal distribution data. For the normality test, the Shapiro-Wilk Test is used. The research data obtained a significance value for each variable of more than 0.05, so it can be interpreted that the data on each variable is normally distributed, thus the data meets the requirements to proceed to the regression test.

Regression test was conducted to determine how much influence Trigona sp honey had on total lactic acid levels in red bean yogurt. while the linear regression equation that will be formed is:

$$Y = a + bX$$

explanation: Y = Total lactic acid levels
a = Constants
b = Regression coefficient
X = Concentration of Trigona sp. Honey

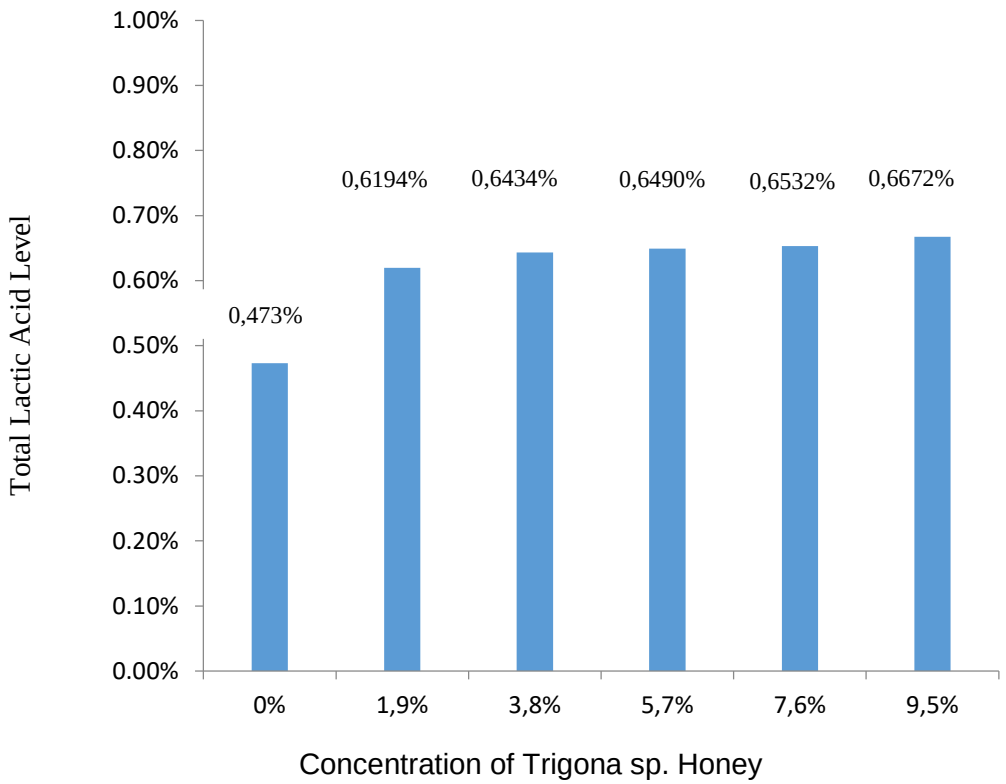

Figure 1. Increase in the average total level of lactic acid in red bean yogurt with the addition of Trigona sp. Honey

Based on the results of the linear regression test obtained a significance value of 0.0001 (α <0.05) stated that the treatment of the addition of different concentrations of honey in yogurt red beans affect the total level of lactic acid, with a linear regression equation

Y = 0.510 + 0.031X.

Values a and b in the above equation can be interpreted as follows:

The value of a = 0.510 is the variable value Y (total lactic acid level) estimated when the variable X (concentration of Trigona sp honey) is equal to zero. This means that if the concentration of Trigona sp honey is zero, then the total lactic acid value is 0.510. The value of b = 0.031 is the regression coefficient which indicates that the effect of variable X (concentration of Trigona sp honey) on the variable Y (total lactic acid level) is unidirectional (positive). This means that if the concentration of Trigona sp honey increases by one unit, then the total level of lactic acid will increase by 0.031 units.

To see how much influence the addition of Trigona sp honey on red bean yogurt to the total level of lactic acid was analyzed the coefficient of determination or can be symbolized by R2. R2 test results obtained R-Square value of 0.599. This value indicates that the effect of adding Trigona sp honey to the total level of lactic acid in red bean yogurt is 59.9%.

The influence of the addition of Trigona sp honey on the total levels of lactic acid is because according to Kuan (2017) Trigona sp honey has a monosaccharide carbohydrate content in the form of glucose at 16.4%. Glucose found in Trigona sp honey adds glucose already in red beans[10].

According to Purwoko (2007) lactic acid bacteria are able to convert glucose into lactic acid. There are two groups of lactic acid fermentation, namely homofermentative and heterofermentative. Homofermentatives use glycolysis through the Embden Meyerhof Pathnas (EMP) and heterofermentative pathways using the Hexosa Monophosphat Pathway (HMP) pathway[11]. So that with the increase in the content of Trigona sp honey in yogurt, it will be directly proportional to the increase in the total level of lactic acid.

Although there was an increase in the total levels of lactic acid but the value of the increase produced was very little in each treatment, this was due to the lack of carbon sources needed by lactic acid bacteria as energy to produce lactic acid. Andualem (2014) suggested that glucose levels in Trigona sp honey were lower than bee honey which reached 31.2%[12]. This can be seen in Figure 5.1, at the concentration of Trigona sp honey 9.5% the total level of lactic acid only reached 0.6672%. In contrast to the research conducted by Rahmayuni (2013), using the addition of other bee honey concentrations of 5%, 10% and 15% in red bean yogurt obtained total lactic acid levels of 0.29% -0.75%. Likewise the research conducted by Nofrianti (2013), added another honey bee with a concentration of 0%, 1.9%, 3.8%, 5.7%, 7.6%, 9.5% in corn yogurt, the level of the total lactic acid produced is in the range of 0.45% -0.85%[13].

The red bean yogurt produced in this study has different consistency and homogeneity than yogurt in general, where the yogurt produced is not too thick and there are white lumps at the bottom and on the surface of the yogurt.

According to Novia (2012), lumps at the base are the result of clumping of protein[14]. Clumping that occurs is caused by a decrease in substrate pH which results in protein denaturation, whereas clumps that occur on the surface are components in red beans which are smaller in density compared to water, so the yogurt produced is different from the usual yogurt.

Yusmarini et al. (2004) stated that during fermentation organic acids were formed which gave rise to distinctive tastes[15]. The sugar content found in red bean milk is used by culture for its metabolic processes so that organic acids, especially lactic acid, are produced. These acids will clump the protein in milk.

With the addition of Trigona sp honey, the lactic acid produced is still not maximal but is in accordance with the quality requirements recommended by SNI 01.2981-2009, namely 0.5% - 2.0%.

CONCLUSION

Red bean yogurt which has been added Trigona sp honey with a concentration of 0%, 1.9%, 3.8%, 5.7%, 7.6%, 9.5%, the average total lactic acid levels are 0.4730 %, 0.6194%, 0.6434%, 0.6490%, 0.6532% and 0.6672%.

The highest average value of total lactic acid is found in red bean yogurt with the addition of Trigona sp honey 9.5%, which is equal to 0.6672%

There is the effect of adding Trigona sp honey to the total level of lactic acid in red bean yogurt with a significance value of 0.0001 ($\alpha < 0.05$).

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