



Comparison of Chemical Characteristics of Sinbiotik Yogurt Fermentation Using *Streptococcus thermophilus* Bacteria *Lactobacillus delbrueckii subsp. bulgaricus* and Chili Stalks

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ABSTRACT

Background: This study aimed to determine the comparison of nutritional content, Lactic Acid Bacteria content and acidity levels of milk-based yogurt using the stater *Streptococcus thermophilus* and *Lactobacillus delbrueckii subsp. bulgaricus* with chili stalks.

Research Methods: The method used in this study was an experimental method consisting of 3 treatments. The addition of prebiotics in the form of stone banana tuber flour aims to improve the quality of yogurt, namely increasing the growth of lactic acid bacteria.

Research Result: The results showed that the addition of prebiotics using chili stalks as a stater contained nutrients and the growth of lactic acid bacteria was better than the stater *Streptococcus thermophiles* and *Lactobacillus delbrueckii subsp. bulgaricus*.

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1. INTRODUCTION

A healthy digestive tract is related to a person's health status, because the digestive tract contains normal microbial flora. The process of metabolizing food in the body requires the presence of bacteria. Bacteria function to ferment nutrients so that they can be absorbed by the body. In addition, the secondary metabolites of bacteria provide many benefits to the body. Secondary metabolites such as vitamin K, B12, and folic acid are needed by the body for blood clotting, blood cell formation and brain cell development (Barkhidarian et al., 2021; Thorakkattu et al., 2022). Another important role of bacterial metabolites in the intestine is in the form of bacteriocins as antibacterial substances, butyrate and verbasosid which can act as anti-inflammatory (Jaye et al., 2022).

Microbial diversity in the gut is determined by several factors including geographical conditions, diet, living conditions, knowledge, gender, and age. This is due to the type of food available in an area, the habit of choosing to consume food, knowledge about healthy food, farming, hunting and the ability to buy food ingredients (Gupta et al., 2017; Leeming et al., 2019; Porras et al., 2021). Foods that enter our body, especially fermented foods, contribute greatly as a source of probiotic bacteria that are good for health (Kerry et al., 2018). Indonesia has a variety of fermented foods and is known to contain lactic acid bacteria that are good for gut health. Fermented foods in Indonesia include tempeh, kombucha, seredele, petis, wild horse milk, oncom, tuak, sequence and tempoyak (Nuraida, 2015; Tamang, 2016). Fermentation occurs because lactic acid bacteria obtain the nutrients they need to survive from the food materials being fermented. These nutrients include inulin, lactulose, soybean oligosaccharides, fructo-oligosaccharides (FOS), galacto-oligosaccharides (GOS), and resistant starch types 1, 2, 3, and 4. These substances are not only found in fermented foods but also naturally occur in whole grains, fruits, vegetables, milk, honey, and soybeans—especially those rich in dietary fiber. Fiber is a type of complex carbohydrate that cannot be easily digested by the human digestive system. Therefore, the role of microorganisms, particularly gut bacteria, is essential to break down and utilize these compounds through the fermentation process in the digestive tract. Microbes have specific enzymes that are able to ferment so that nutrients from fiber foods can be absorbed by the body. Foods that contain fiber, inulin, fructo-oligosaccharides (FOS), lactose and other complex carbohydrates are called prebiotics. Prebiotics are a food source for normal gut microflora to survive and thrive and provide benefits to gut health (Ayodeji et al., 2017; Kerry et al., 2018; Rivera-Piza & Lee, 2020).

In addition to normal gut microflora, prebiotics are required as a source of nutrients by probiotics. Probiotics are active bacteria that provide human health benefits through the gastrointestinal tract. The combination of prebiotics and probiotics is called synbiotics, both of which will contribute positively to human health. Therefore, this study will investigate the prebiotic ability of banana stump flour, sorghum flour and chicken egg membrane flour. The ability of all three will be tested on probiotic *Lactobacillus fermentum* and pathogenic bacteria *Escherichia coli*. *Lactobacillus fermentum* is known in the research of Zhang et al., (2020), to play an important role in supporting the recovery rate of autoimmune diseases, platelet formation in patients with idiopathic thrombocytopenia and maintaining the condition of the kidneys and heart of people with lupus (Guo et al., 2021). Bonggol pisang batu which is made into lawar is known to have a crude fiber content of 4.45% (Komalasari et al., 2016), white sorghum flour from the East Nusa Tenggara variety has 2.72% fiber (Avif & Oktaviana TD, 2021) and the results of research by Jia et al., (2017), found that egg membranes despite having properties as proteins that are not easily digested are known to be able to act like fiber

in the intestine by increasing the activity of intestinal microflora, to treat inflammatory bowel disease. If in this study stone banana pomace flour, sorghum flour and chicken egg membrane flour are able to increase the growth of *L. fermentum* and suppress the growth of *E. coli*, then the three flours can be used as prebiotic ingredients and the manufacture of synbiotic supplement products for autoimmune patients.

2. METHODS

2.1. Material and Tools

The materials used in this study were stone banana tuber (*Musa balbisiana*), Sorghum flour (*Sorghum bicolor* L.), *Lactobacillus fermentum* (Gadjah Mada University), Cimory (Commercial yoghurt), Nutrient Agar media (Oxoid), Nutrient Broth (Merck), MRSA Agar (Oxoid), MRSA Broth (Oxoid), Alcohol (Onemed), distilled water, 70% alcohol (Onemed), cleaning soap (Mama Lemon).

The tools used in this research are hot plate (Corning), Graduated cylinder (Pyrex), beaker glass (Iwaki), oven (Memmert), analytical balance (Acis), blender (Miyako), Erlenmeyer (Iwaki), thermometer (Pyrex), spatula, pan, magnetic stirrer, aluminium foil.

2.2. Research Procedure

2.2.1. Preparation of Stone Banana Tuber Flour

Stone banana stems are selected from banana trees that have already borne fruit, are not affected by plant diseases and are grown in a clean environment. The stem is cleaned from the outer skin, roots, fronds and soil. Washed thoroughly and cut into ± 0.5 cm pieces, washed again. The cut bamboo shoots were then baked at 60°C for 18 hours. Then mashed using a blender and sieved using an 80 mesh sieve. The finished tuber flour is sterilized using an autoclave at 70°C for the last 2 hours of flour stored in polypropylene plastic (Bernatal, S., 2013; Hardisari et al., 2016).

2.2.2. Processing of Synbiotic Yogurt with Chili Stem Starter

Milk powder of 25 g was added to 500 ml of water and heated at 80°C for 30 minutes. Sugar was added as much as 2% (w/b) and stirred well, then the temperature was reduced to 45°C, and 25 stalks of chili stem probiotic starter were added. While the control group used starter from cimory products, without the addition of stone banana stem flour.

2.2.3. Product Analysis

In this study, the application of the addition of banana stem flour as a source of prebiotic inulin in the manufacture of yogurt with chili stem starter and probiotics *Streptococcus thermophilus*, *Lactobacillus delbrueckii subsp. bulgaricus* derived from commercial yogurt. Analysis was carried out by measuring the degree of acidity (pH) and chemical characteristics and total lactic acid bacteria (Total Plate Count). In this preliminary research, data analysis was carried out using descriptive analysis (Hardisari et al., 2016; Sari & Puspaningtyas, 2019).

3. RESULTS AND DISCUSSION

The results of pH analysis, chemical characteristics and total growth of lactic acid bacteria (Table 3.) were obtained from synbiotic yogurt and positive control. The starter used chili stalks and probiotics *Streptococcus thermophilus*, *Lactobacillus delbrueckii subsp. bulgaricus* were from commercial yogurt.

Table 1. Chemical Characteristics and Total Lactic Acid Bacteria

No.	Sample	Water (%)	Ash (%)	Protein (%)	Fat (%)	Carbohydrate (%)	Fiber (%)	pH	TPC (cfu/g)
1.	Kontrol	86,721	0,644	4,014	2,830	5,791	0,407	5,61	1,1 x 10 ⁸
2.	Yt5	83,439	1,327	3,861	2,954	8,419	1,225	5,63	2,1 x 10 ⁸
3.	Yt10	80,592	1,970	7,915	3,123	6,400	2,340	6,41	2,6 x 10 ⁸
4.	Standar SNI	-	Maks 1,0	Min 2,7	Min. 3/0,6-	-	-	3,80-4,50	Min. 10 ⁷
	Yoghurt				2,9				

Source: Research Analysis

Synbiotic yogurt has a fat content in accordance with the quality standards of SNI 2981: 2009, for fatty yogurt (Yt10) was higher than the minimum value (3%) and for low-fat yogurt (Yt5) had value between 0.6-2.9%. The source of fat content in yogurt in this research, comes from the dairy milk used. However, in the present study there is no difference in fat content in plant-based yogurt and dairy yogurt. The only difference is that plant-based yogurt contains more fibers, lower sodium, sugar, protein, calcium and potassium (Marlapati, et al., 2024).

Ash content of Yt5 and Yt10 were higher compared to the SNI standard, more than 1% but the ash component in yogurt has a function to increase protein. Stone banana stem flour contains a high mineral content of 3-5%. Some of the mineral elements contained in banana stone tuber flour are magnesium, iron, calcium, ammonium nitrate, ammonia and potassium oxide. Therefore, the addition of banana stone tuber flour into yogurt will increase the mineral content. The longer the incubation time of yogurt, the mineral content also increases (Haryanto et al., 2023; Cahyadi et al., 2024).

The protein content of both formulations was higher compared with SNI, which the minimum value is 2.7%. Stone Banana tuber flour also contains with protein. In previous research conducted by Cahyadi et al., (2024) the protein content of kepok banana tuber flour that has been fermented using Bimo Cf stater contains quite high protein 10.24%. The results of this study showed that with the increase in the concentration of stone banana flour in the fermentation of Yt10, the protein content also increased. In the previous study found that content of protein from yogurt full-fat dairy was not significantly different compared to yoghurt made from plant-based such as almond (D'Andrea et al., 2023).

The pH acidity level does not meet the SNI standard. Samples Yt5 and Yt10 also the control has higher values of pH than the limit value on SNI which between 3.80-4.50. Higher pH value conditions can be caused by the addition of banana stone tuber flour which has high pH around 6, according to research by Rahmawati et al., (2023). The length of fermentation times and storage temperature of yogurt can affect acidity levels. In this study, the incubation duration was 18 hours, the incubation times between 18-42 hours should be due to increased yogurt acidity (Adiputra et al., 2022). But unlike what was found in this study, the value of yogurt acidity becomes alkaline, even still under 7. The use of chili stalks which are plants and concentration of banana stone tuber flour will increase the alkaline pH content of yogurt. However, the measured pH is not much different from the positive control yogurt. The growth of lactic acid bacteria is higher with the addition of stone banana stem flour and chili stalks starter. This shows that chili stalks can be used as a starter for growing lactic acid bacteria and can further develop with the addition of banana stone tuber flour. Banana stone tuber flour contains fiber and inulin which are used by lactic acid bacteria to develop (Hardisari, et al., 2016; Nuraida, 2016; Sabrina et al., 2021). This condition may cause the acidity level to slowly

decrease because lactic acid bacteria are still developed. As the number of live stater bacteria in yogurt, for example *Lactobacillus acidophilus*, decreases, the acidity of yogurt will increase according to the length of storage time (Tavakoli et al., 2019).

4. CONCLUSION

The addition of banana pomegranate flour as a source of prebiotic inulin and chili stems as a starter for the growth of lactic acid bacteria resulted in an increase in chemical characteristics and growth of lactic acid bacteria in accordance with the standard, only the disadvantage is that the pH level becomes more basic than the standard yogurt and requires a longer incubation time so that the pH becomes more acidic.

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