

The Effects of the Addition of Banana Puree to the Total Number of Total Probiotic Bacteria, pH Value and Organoleptic Characteristics of the Synbiotic Yogurt Made from Goat Milk and Banana Puree

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Abstract

Synbiotic yogurt is a yogurt with the addition of prebiotics and lactic acid bacteria (LAB) that serves as probiotic which helps to improve digestion, able to survive in gastric acid condition and able to improve immune system. The milk that was used in this study is Saneen goat milk which has higher digestibility than cow milk. The banana used in this study was processed into puree form. The purpose of study was to determine the concentration of banana puree on synbiotic yogurt made from goat milk and banana puree with the desired amount of microbiological, pH value, viscosity and organoleptic characteristics based on SNI. The research method used was randomized block design with 4 treatments and 3 replications. Synbiotic yogurt was made with difference concentrations of banana puree of 25%, 50%, 75% and 100%. The 50% addition of banana puree produced the best synbiotic yogurt with 4.15 pH, , 9,43 Log CFU/mL acid resistance probiotic bacteria and 9,16 Log CFU/mL bile salt resistance probiotic bacteria, and the organoleptic characteristics including color, flavor, taste, texture and overall appearance rather preferred by panelists.

Keywords: *Banana puree, goat milk, L. acidophilus, synbiotic yogurt*

1. INTRODUCTION

The increasing public awareness of the importance of healthy living has created a shift in consumer's demand for foods. Consumers are not only demand for foods that are nutritious and delicious, but they also want foods that have physiological functions for the body, which is called functional food.

According to [1], functional foods are foods (not capsules, pills nor flour) that are derived from natural ingredients. Functional foods are foods whose active component content can provide health benefits, beyond the benefits provided by the nutrients contained therein [2]. Foods are said to be functional when they contain

nutrients or non-nutrients (active components) that can affect the physiological function of the body in a positive direction such as to prevent certain diseases, strengthen the body's defense mechanisms, maintain physical and mental conditions, and slow the aging process.

The currently popular functional food is the food products that contain probiotics and prebiotics or so-called synbiotic foods. Probiotics and prebiotics are components that have been proven to provide health benefits for humans. Probiotics are microorganisms that provide health benefits to its host through its effects in the intestinal tract [3]. Prebiotics are components of food that cannot be

hydrolyzed by digestive enzymes in the human digestive tract but these components can benefit the body by stimulating the growth or activity of a number of bacteria in the colon that ultimately can improve the health [4].

One of the synbiotic foods that start to catch the public's interest is synbiotic yogurt. Synbiotic yogurt is a yogurt with the addition of probiotic lactic acid bacteria (LAB) that can live and metabolize in the intestine such as *Lactobacillus acidophilus* and *Lactobacillus casei*, *L. acidophilus*. These LABs are probiotic bacterium which can improve human digestive function, survive in stomach acid pH conditions, and boost the immune system [5].

Most yogurt products available on the market are made from cow's milk. Making yogurt by using milks other than cow milk has not been done much. [6] in [7] mentioned that there is a high demand for new dairy products due to the presence of cow's milk allergy and indigestion. Cow's milk allergy is an undesirable immunologically mediated reaction to cow's milk protein. The presence of β -immunoglobulin (β -Ig) in cow's milk is considered to be one of the major protein antigens that cause allergic reactions in infants [8] in [9]. Based on this, synbiotic goat yogurt will be made.

According to [6], goat milk is reported to have higher digestibility and lower allergic properties compared to cow's milk. The number of fat globules which are homogenous and small in diameter are more commonly found in goat milk which make goat milk more easily digested by human digestive organs and does not cause diarrhea for those who consume it. In addition, goat milk has a high content of short chain fatty acids, and has a high content of zinc, iron and magnesium and has anti-bacterial characteristics [10]. In addition, high florin content in goat milk can be antiseptic.

The use of goat milk is also done in order to improve the utilization of goat milk in Indonesia. Goat milk is less preferred because it has a distinctive aroma of goat commonly called *prengus*. Processing goat

milk into yogurt can reduce the smell of *prengus* because the process of fermentation in milk will depress gases or volatile compounds such as acetaldehyde, diacetyl, and CO₂ that can reduce the smell of *prengus*. This is supported by the statement of [11] that the original aroma of milk will be reduced if the acetaldehyde content is high.

The goat milk that is going to be used in this study comes from a type of goat called *Saanen*. According to [12] *Saanen* goat is a superior dairy goat in the world that can produce milk about 3-4 l/day. The peak of *Saanen* goat production can produce milk about 5-6 l/day [13].

In order to increase the functional food variation of synbiotic yogurt, local raw materials will be used as a source of prebiotics namely banana (*pisang kepok*). Based on [14], bananas contain inulin and fructose-oligosaccharides (FOS) used as a prebiotic substrate. [15] states that Kepok bananas has an inulin content of 3%. Bananas also has a high resistant starch content compared to other bananas that amounted to 22.01% [16].

In an effort to increase the economic value of bananas, it is necessary to diversify the utilization of bananas. The banana would be processed into a form of banana puree added to yogurt products as a source of prebiotics. The addition of banana puree to synbiotic yogurt is done to obtain the distinctive aroma of a banana.

Synbiotic yogurt has the same characteristics as yogurt in general, but in synbiotic yogurts, they must contain probiotic bacteria at least 7 log CFU/ ml. Yogurts have a texture ranging from slightly thicker to thick or semi-solid with a homogeneous viscosity resulting from protein clumping due to organic acid produced by starter cultures [17].

Based on the above description, the authors are encouraged to conduct a research on the effect of the addition of banana puree to goat milk in order to obtain a synbiotic yogurt with good characteristics that is favored by people.

2. MATERIALS AND METHODS

The materials used in this research are fresh goat milk from *Saanen* goat type obtained from goat breeder in Ciparanje, bananas with maturity index VII or fully ripen, “Yogurmet” freeze dried starter (*L. bulgaricus*, *S. thermophilus*, and *L. acidophilus*), caster sugar, and aquades. The media used in the analysis were “Merck” deMann Rogosa Sharpe agar (MRSA), “Merck” deMann Rogosa Sharpe Broth (MRSB), aquades, 1N NaOH, HCl, Bile Salt, 1% phenolphthalein, 70% alcohol, spiritus, physiologic NaCl 0.85 %, buffer solution pH 4 and pH 7. The tools used in the making of yogurt in this study were refrigerator, incubator, autoclave, pot, gas stove, gas cylinder, jar, grinder, erlenmeyer 500 ml, plastic, mask, rubber glove, bunsen, funnel, aluminum foil, tissue, basin, Storage plastic box, and spoon. The tools used for analysis are microscope, viscometer, pH meter, analytical balance, micropipette, 100 ml measuring cup, beaker, ose, buret, test tube, tube rack, thermometer, petri dish, filter cloth, bulb pipette, dropper drops, stirrer rods, spatulas, calculators, and stationery.

The research method employed in this research is the experimental method using the experimental design of Randomized Block Design with the treatment variation of the addition of Kepok banana puree to the fresh goat milk by 25%, 50%, 75% and 100%. Banana puree was made by steam blansing kepok banana in 63°C for 7 minutes and grinding the kepok banana with blender. Each treatment was done with 3 repetitions after 2 days of storage. The data of this research were then analyzed statistically with Duncan’s test using SPSS 17.0.

2.2 Probiotics’ Acid and Bile Salts

Tolerance test (Modification of [18])

The probiotics’ acid tolerance test was carried out by pouring 10 ml of MRSB into the test tube. HCl was added until pH 4 is achieved. 10 ml of sample was incorporated into MRSB plus HCl and incubated for 2 hours at 37 ° C. Meanwhile, for probiotic testing against bile salts, bile salt of 0.3% was added into the 10 ml MRSB. A sample of

10 ml was incorporated into MRSB which had been added with bile salt and incubated for 4 hours at 37°C. A 1 ml suspension was taken from both test tubes for dilution 10⁻⁶, 10⁻⁷ and 10⁻⁸. Samples of 1 ml were taken each from the last 3 dilutions and inserted into a petri dish. MRS agar (Merck) was poured as much as 12-15 ml.

2.1 Production Process of Banana Puree Synbiotic Yogurt

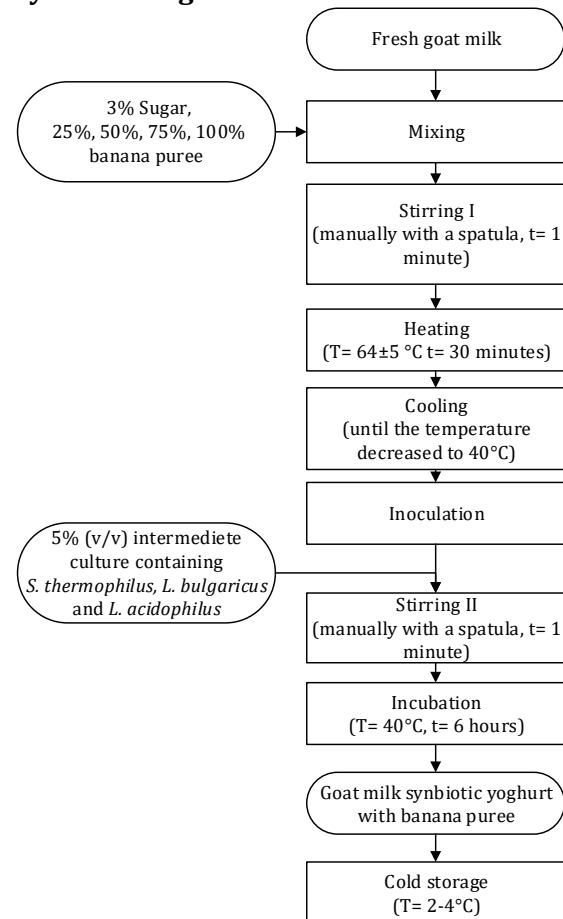


Figure 1. Diagram of the Manufacturing Process of Synbiotic Goat Milk Yogurt with Banana Puree

Petri dishes were incubated for 48 hours at 37 ° C. The amount of colonies grown on the agar were calculated using the BAM calculation method.

$$N = \frac{\sum C}{[(1 \times n_1) + (0,1 \times n_2)] \times (d)}$$

With:

N = the number of product colonies, expressed in colonies per ml or colony per g.

ΣC = number of colonies in all calculated petri dish

n_1 = the amount of petri dish in the calculated first dilution

n_2 = the amount of petri dish in the calculated second dilution

d = the calculated first dillution.

pH Test [19]

The pH measurement was performed using pH meters that had been calibrated with buffer solutions of pH 4 and 7. The sample measurements were made by taking 20 ml of the sample. Previously, the electrodes were rinsed with aquades and then dried with tissues. After that, the electrode is immersed in the sample for a while until it reads a stable pH value.

Hedonic Test [20]

The hedonic testing was conducted by 20 trained panelists. The trained panelists were randomly selected in order to be asked to assess the degree of preference for the color, flavor, aroma, texture and overall appearance of the sample. A sample of 20 mL was prepared in a small cup. The test table was provided in the form of questionnaire form that contains the assessment in the forms of numbers with a scale of one to five, where 1 is not liked, 2 that is less liked, 3 is rather favored, 4 is favored and 5 is very favored.

3. RESULTS AND DISCUSSION

3.1 The Total of the Probiotics

Based on the statistical analysis, synbiotic *Saanen* goat milk yogurt with the addition of 25% (A), 50% (B), 75% (C) and 100% (D) banana puree gives different effects significantly to the total of acid and bile salt resistance bacteria. The effect of the addition of banana puree to the total probiotic of synbiotic goat milk yogurt can be seen in Table 1.

According to [21] the minimum standard of probiotic bacteria living in

dairy-based products is 10^7 cfu / ml so that the *Saanen* goat milk synbiotic yogurt in all treatments had fulfilled the standard. Based on Table 1, there was an increase of the total probiotic due to the 25% addition of banana puree (A) as well as the 50% addition of banana puree (B), then there was a decrease of total probiotic in 75%(C) and 100%(D) addition of banana puree

Table 1. The Effect of Banana Puree Addition to the Total Probiotics of the Goat Milk Synbiotic Yogurt with Banana Puree

Treatment	Mean (Log cfu/ml)	
	Acid Tolerance Test	Bile Salts Tolerance Test
A (25%)	7,92 b	8,65 b
B (50%)	9,43 a	9,16 a
C (75%)	7,93 b	7,85 b
D (100%)	7,90 b	7,80 b

The average treatments which were marked with the same lower case of same column were not significantly different at the 5% level according to Duncan's Test.

The decrease of probiotic in treatment B and C is suspected due to the addition of excessive banana puree. The nutrition contained in banana puree such as glucose and protein is needed for bacterial growth, but glucose that serves as a substrate can also be one of the growth inhibiting factor when its existence is excess or greater than its critical value [22] in [23] while the excess nitrogen in general can reduce substrate conversion into polysaccharides [24].

The condition of the banana used was fully ripen where the banana has a high glucose content. Excessive glucose can be a bacterial growth inhibitor. According to [25], bacterial inhibition mechanisms occur because of the high concentrations of glucose that can form an environment with low water activity (suddenly decreasing amount of water) causing an increase in osmotic pressure. The high osmotic pressure causes hypertonic environmental

conditions so that the cells become dehydrated and cell death occurs.

3.2 pH

Based on statistic analysis, synbiotic *Saanen* goat milk yogurt with the addition of 25% (A), 50% (B), 75% (C) and 100% (D) banana puree gave significant effect on the pH. The effect of adding banana puree to pH of synbiotic goat milk yogurt can be seen in Table 2.

Table 2. The effect of the addition of banana puree to the pH of the goat milk banana puree synbiotic yogurt

Treatment	Mean
A (25%)	4,38 ab
B (50%)	4,15 b
C (75%)	4,67 ab
D (100%)	4,85 a

The average treatments which were marked with the same lower case were not significantly different at the 5% level according to Duncan's Test.

The obtained *Saanen* goat milk synbiotic yogurt had a pH range 4.15-4.85. Based on SNI no. 2981-2009, a good yogurt has a pH range of 4.0-4.5. This shows that *Saanen* goat milk synbiotic yogurt with the treatment C and D has not met the standard because it has pH above 4.5.

The addition of banana puree give effects to the of pH *Saanen* goat milk synbiotic yogurt. Banana puree provides additional nutrients such as carbohydrates. This carbohydrate is utilized by *L. acidophilus*. This is in accordance with [26] that the availability of carbohydrates as a source of energy leads to an increase in activity metabolism by *L. acidophilus* bacteria that contributes to a decrease in pH. The content of lactose in goat's milk also affects the pH. This is consistent with [27] who claimed that lactic acid bacteria utilize lactose and produce lactic acid which can decrease pH as metabolites because lactose is the main source of carbon and energy source for BAL especially by *S. thermophilus* bacteria species.

3.3 Organoleptic assessment of color, flavor, taste, texture and overall appearance

Color

Based on statistical analysis, the *Saanen* goat milk synbiotic yogurt with the 25% (A), 50% (B), 75% (C) and 100% (D) addition of banana puree has no significant effect on the rate of favorable color of the *Saanen* goat milk synbiotic yogurt. The effect of the addition of banana puree to the rate of favorable color of the *Saanen* milk goat synbiotic yogurt color can be seen in Table 3.

Table 3. The effect of the addition of banana puree to the rate of favorable color of the goat milk banana puree synbiotic yogurt

Treatment	Mean
A (25%)	2,90 a
B (50%)	3,55 a
C (75%)	3,30 a
D (100%)	2,90 a

The average treatments which were marked with the same lower case were not significantly different at the 5% level according to Duncan's Test.

The color of the *Saanen* goat milk synbiotic yogurt is yellowish white goat milk and the yellow color is intensified along with the increasing concentration of banana puree. The panelists favor the goat milk synbiotic yogurt with treatment B that has a slightly yellowish white color and slightly dislike the goat milk synbiotic yogurt whose color is white and too yellow like those owned by yogurts with treatment A and D.

The yellow color of in the *Saanen* goat milk synbiotic yogurt comes from the added banana puree and is not caused by the metabolism activity of lactic acid bacteria in yogurt. This is based on [28] assertion that the results of lactic acid bacteria metabolism activity in yogurt, lactic acid, acetaldehyde, diacetyl, acetic acid, and other volatile agents have no effect on yogurt color. The bananas used for the banana puree is a banana with a level of maturity VI / VII which is characterized by the evenly

distributed yellow color on the banana skin. The yellow color of the banana comes from the carotene that is formed due to the loss of chlorophyll during the maturation process [29].

Aroma

Based on statistical analysis, synbiotic milk goat yogurt with the 25% (A), 50% (B), 75% (C) and 100% (D) addition of banana puree gave no significant effect to the rate of favorable aroma of the synbiotic saanen goat milk yogurt. The effect of adding banana puree to the rate of favorable aroma of the synbiotic saanen goat milk yogurt. can be seen in Table 4.

Table 4. The effect of the addition of banana puree to the rate of favorable aroma of the goat milk banana puree synbiotic yogurt

Treatment	Mean
A (25%)	3,50 a
B (50%)	3,55 a
C (75%)	2,90 a
D (100%)	3,05 a

The average treatments which were marked with the same lower case were not significantly different at the 5% level according to Duncan's Test.

Saanen goat milk synbiotic yogurt has a mixture of a typical sour yogurt aroma and typical banana scent. This goat milk synbiotic yogurt has fulfilled the requirements of a yogurt aroma according to [30] which is having a normal/distinctive aroma of yogurt. According to [31] the flavor component of yogurt is divided into four main categories: non-volatile acids (pyruvic acid), volatile acids (acetic acid, propionic acid or butyrate and formic acid) carbonyl components (acetone, acetaldehyde, acetoin and diacetyl) and other components such as amino acids. Volatile acid compounds act as a balancing aroma component in yogurt products, formed by metabolic activity of both types of yogurt bacteria.

The bananas used are bananas with full maturity level. According to [32], in fully mature banana, there is a volatile compound which is amyl acetate, the main component

of a typical banana odor. In addition to amyl acetate, volatile components derived from bananas which also provide a role in forming the final flavor of yogurt namely acetate, amyl acetate, and hexyl alcohol [33].

The making of *Saanen* goat milk synbiotic yogurt using goat milk is generally less favored by people because it has a distinctive aroma which is called prengus. The prengus aroma of goat milk comes from the short-chain and medium-chain fatty acids such as caproic acid, caprylic acid and capric acid [34]. The reduced odor of prengus in synbiotic *Saanen* goat milk yogurt is caused by the fermentation process in milk that condenses gases or volatile compounds such as acetaldehyde, diacetyl and CO₂ which can reduce the scent of prengus. This is supported by the statement of [11] that the original aroma of milk will be reduced if the acetaldehyde content is high after the fermentation process.

Taste

Based on the statistical analysis, synbiotic goat milk yogurt with the 25% (A), 50% (B), 75% (C) and 100% (D) addition of banana puree gave no significant effect to the rate of favorable taste of the synbiotic *Saanen* goat milk yogurt. The effect of adding banana puree to the rate of favorable taste of synbiotic goat milk yogurt can be seen in Table 5.

Table 5. The effect of the addition of banana puree to the rate of favorable taste of the goat milk banana puree synbiotic yogurt

Treatment	Mean
A (25%)	2,70 a
B (50%)	3,10 a
C (75%)	2,95 a
D (100%)	3,00 a

The average treatments which were marked with the same lower case were not significantly different at the 5% level according to Duncan's Test.

Synbiotic goat milk yogurt has a sour taste with the mixture of sweet taste from the banana. It meets the requirement of

yogurt flavor based on [30] which is a typical sour taste of yogurt. According to Table 5, the panelists favor the samples from treatment B, of which yogurt has a balanced sour and sweet taste. The panelists slightly dislike the samples from treatment A which have a very dominating sour taste.

The sour taste in synbiotic *Saanen* goat milk yogurt is derived from milk carbohydrates (lactose) which are transformed during the fermentation process [35]. This is supported by the statement from [36] which states that fermented lactic acid provides a sour taste and donates a distinctive flavor of yogurt.

The sweet taste of the yogurt comes from the added banana puree. The banana used is a banana with full mature ripeness level where in this maturity level a high sugar level is generated. [37] stated that the chemical changes that occur during banana maturation are the increased levels of sugar, the production of volatile substances and decreased levels of starch. The increase of sugar tends to cause sweetness in the fruit [38].

Texture

Based on the statistical analysis, synbiotic *Saanen* goat milk yogurt with the 25% (A), 50% (B), 75% (C) and 100% (D) addition of banana puree gave a significant effect on the rate of favorable texture of the synbiotic *Saanen* goat milk yogurt. The effect of the addition of banana puree to the rate of favorable texture of the synbiotic *Saanen* goat milk yogurt can be seen in Table 6.

Table 6. The effect of the addition of banana puree to the rate of favorable texture of the goat milk banana puree synbiotic yogurt

Treatment	Mean
A (25%)	3,60 a
B (50%)	3,80 a
C (75%)	2,70 b
D (100%)	2,15 b

The average treatments which were marked with the same lower case were not

significantly different at the 5% level according to Duncan's Test.

This synbiotic *Saanen* goat milk yogurt has a thick and homogeneous texture. This is in accordance with yogurt texture requirements based on [30] which is a liquid to viscous appearance and has a homogeneous concentration. The panelists favor B treatment samples that have a slightly thicker and denser texture.

The addition of banana puree affects the texture of the synbiotic *Saanen* goat milk yogurt. One of the things that affect the texture of the yogurt is the total concentration of dissolved solids. According to [39], the higher the content of dissolved solids in yogurt, then it will produce yogurts with a high viscosity. The higher the banana puree concentration is added, the more viscous and dense the texture is produced. The fermentation process also plays a role in texture formation in yogurt, this is explained by [40] which states that during fermentation there is a decrease in pH and causes the casein protein to clot and form gel. The formation of this gel causes the yogurt viscosity to increase and the texture of yogurt becomes thicker.

Overall

Based on the statistical analysis, synbiotic *Saanen* goat milk yogurt with the 25% (A), 50% (B), 75% (C) and 100% (D) addition of banana puree gave no significant effect on the overall acceptance rate of the synbiotic *Saanen* goat milk yogurt. The effect of adding banana puree to the overall acceptance rate of the synbiotic *Saanen* goat milk yogurt can be seen in Table 7.

Table 7. The effect of the addition of banana puree to the overall acceptance rate of the goat milk banana puree synbiotic yogurt

Treatment	Mean
A (25%)	3,4 a
B (50%)	3,45 a
C (75%)	3,45 a
D (100%)	3,3 a

The average treatments which were marked with the same lower case were not significantly different at the 5% level according to Duncan's Test.

The overall assessment is the panelist assessment of the samples of four types of the addition concentration of banana puree, which includes sensory attributes, ie color, flavor, aroma and texture. Based on the statistical tests, the addition of banana puree to the yogurt only significantly influences the texture favorability while other sensory attributes such as color, taste and flavor are not significantly affected. Overall, the addition of 50% banana puree (B) and 75% (C) is the most preferred sample by panelists with a score of 3.45. Meanwhile, the addition of 100% banana puree (D) is the least preferred sample with a score of 3.3.

4. CONCLUSION

The addition of various concentration of banana puree on the making of synbiotic *Saanen* goat milk yogurt gave significant different effects to the total probiotic, pH, panelists' favorability to the organoleptic characteristics of texture. However that did not give significantly different effect to panelists' level of preference to organoleptic characteristics of color, aroma, taste, viscosity, and overall appearance.

The addition of 50% banana puree (B) on the making of synbiotic *Saanen* goat milk yogurt yields the best characteristics by having the total probiotic 9,43 Log cfu / ml with the bile salt resistance test and 9,16 Log cfu / ml by the testing of acid resistance, pH value of 4.15, organoleptic color characteristics with an average preferred value of 3.55 (rather favored); aroma 3.55 (rather favored); taste 3.10 (rather favored); texture 3.80 (rather favored); overall appearance of 3.45 (rather favored).

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