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CHARACTERISTICS OF JUWET FRUIT YOGHURT WITH THE ADDITION OF SKIM MILK AND LACTIC ACID BACTERIA STARTER

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ABSTRACT

In this study, the manufacture of juwet fruit yoghurt was studied with different treatments of adding skim milk and lactic acid bacteria starter. Yoghurt is a processed milk product made through bacterial fermentation. Yoghurt is known as a good source of protein, calcium, and probiotics, which can support digestive health and the immune system. Juwet fruit has a high antioxidant and vitamin C content so that juwet fruit has interesting potential to be made into yoghurt. Skim milk is used to utilize high lactose and protein so that the growth of lactic acid bacteria can grow optimally. Lactic acid bacteria are used for preservation, providing texture, and adding flavor to yoghurt drinks. The purpose of this study was to determine the addition of skim milk and lactic acid bacteria starter on the physicochemical, microbiological, and organoleptic characteristics of juwet yoghurt. This study used a Completely Randomized Design (CRD) factorial pattern consisting of two factors. Factor 1 addition of skim milk (5%, 7%, 9%). Factor 2 addition of starter (5%, 7%, 9%). Each treatment was repeated three times. The data obtained were analyzed using analysis of variance (ANOVA). If there is a significant difference between treatments, it is continued with the Duncan test (DMRT) and organoleptic test using the Scoring test with 25 trained panellists. The treatment of adding 9% skim milk and 9% starter was the best treatment that produced juwet yoghurt with a protein content of 2.1% ± 0.036, total titratable acid 2.1% ± 0.1, total soluble solids 22.7ºBrix ± 1.53, total lactic acid bacteria 10.5 log CFU/ml ± 0.023, pH 3.7 ± 0.058, antioxidants 34.0% ± 0.117, viscosity 6.8% ± 0.058, Vitamin C 5.8 ± 0.3, and organoleptic tests of color 2.88 (rather like), taste 3.24 (rather like), aroma 2.68 (rather like), texture 3.76 (like).

1. INTRODUCTION

1.1. Research Background

Yoghurt is a drink produced from the lactic acid fermentation process by lactic acid bacteria (LAB). The quality requirements for fermented milk drinks are a minimum fat content of 3%, a minimum protein content of 2.7%, an acidity of 0.5-2.0%, and a minimum starter culture of 107 cfu/ml [1]. Lactic acid bacteria are a group of bacteria that can ferment sugar into lactic acid. *Streptococcus thermophillus* and *lactobacillus bulgaricus bacteria* support each other in producing lactic acid and aroma-producing compounds. *Streptococcus thermophillus* produces pyruvic acid, formic acids, CO^2, and folic acid which stimulate the growth of *lactobacillus bulgaricus* [2]. In general, lactic acid bacteria originating from the human digestive tract such as *lactobacillus* and *bifidobacterium* can act as good probiotics, while *streptococcus* and *lactococcus* which are fermented cultures of dairy products cannot reach the human intestines alive [3]. Juwet fruit or jamblang fruit is a fruit plant originating from tropical Asia and Australia. Some uses of the juwet plant in Indonesia are that the fruit is eaten as fresh fruit and the fruit, leaves, and especially the seeds have long been used as a medicine for diabetes. This juwet fruit contains chemical compounds including vitamin C, gallic acid, tannin, anthocyanin group, cyanidin, petunidin and malvidin. The vitamin C content in juwet fruit can cause a sour taste when consumed. Vitamin C in this fruit can also function as an antioxidant. The antioxidant content in juwet fruit plays an important role in preventing various diseases. The antioxidant content in juwet fruit is influenced by the level of ripeness of the fruit. The highest antioxidant content is found in juwet fruit that has turned purple-black [4]. This study will see the best effect between the addition of skim milk and lactic acid bacteria starter on the juwet fruit yoghurt that will be made. The addition of skim milk is used to provide carbohydrates and lactose for the growth of LAB and can ferment sugar into lactic acid..

1.2. Literature Review

Yoghurt drinks are drinks that include fresh products or processed products that are not only refreshing but also have a good impact on health and reduce the risk of disease for consumers. Yoghurt drinks contain beneficial components or compounds such as dietary fiber, oligosaccharides, sugar alcohols, amino acids, peptides, proteins, alcohols, glycosaccharides, vitamins, lactic acid bacteria, polyunsaturated fatty acids and antioxidants [5]. Beverage products fermented with lactic acid bacteria can provide health benefits, such as preventing aging, reducing allergies, and maintaining the walls of the digestive tract [6]. Fermentation is one of the microbial activities to use organic compounds or carbon sources to obtain energy for its metabolism with by-products in the form of gas as a carbon source in fermentation is lipid. Microbes that play a role in fermentation can be classified into bacteria, molds, and yeasts. The results of fermentation mainly depend on several factors, namely the type of food (substrate), the type of microbe and the surrounding conditions that affect the growth and metabolism of the microbe. Fermentative microbes can convert carbohydrates and their derivatives, especially into alcohol, acid and CO^2. Proteolytic microbes can break down proteins and nitrogen components so that they produce an undesirable odor, while lipolytic microbes will break down or hydrolyze fats, phospholipids and their derivatives by producing a rancid odor The principle of fermentation is to activate the growth and metabolism of alcohol and acid-forming microbes and suppress the growth of proteolytic and lipolytic microbes. Factors that affect fermentation are the number of microbes, fermentation time, pH (acidity), substrate (medium), temperature, oxygen, alcohol, salt, and water. Fermentation is a process that produces various products both aerobically and anaerobically by involving the activity of microbes or their extracts in a controlled manner. Fermentation can increase food diversity and produce products with distinctive tastes, aromas, and textures, in addition to extending the shelf life of products [7].

1.3. Research Objective

The purpose of this study was to determine the effect of adding skim milk and lactic acid bacteria starter on the physicochemical, microbiological, and organoleptic characteristics of juwet fruit yoghurt, and to determine the best combination of treatments between the addition of skim milk and lactic acid bacteria starter that produces yoghurt drinks with the best characteristics and preferred by panelists.

2. MATERIALS AND METHODS

The making of juwet yoghurt in this study used a formulation by combining the concentration of skim milk (A) and lactic acid bacteria starter (B), consisting of 9 treatment formulations:

A1B1 (Addition of 5% Skim Milk and Addition of 5% Starter),
A2B1 (Addition of 7% Skim Milk and Addition of 5% Starter),
A3B1 (Addition of 9% Skim Milk and Addition of 5% Starter),
A1B2 (Addition of 5% Skim Milk and Addition of 7% Starter),
A2B2 (Addition of 7% Skim Milk and Addition of 7% Starter),
A3B2 (Addition of 9% Skim Milk and Addition of 7% Starter),
A1B3 (Addition of 5% Skim Milk and Addition of 9% Starter),
A2B3 (Addition of 7% Skim Milk and Addition of 9% Starter),
A3B3 (Addition of 9% Skim Milk and Addition of 9% Starter).

This study used a Completely Randomized Design (CRD) factorial pattern consisting of two factors. Factor 1 addition of skim milk (5%, 7%, 9%). Factor 2 addition of starter (5%, 7%, 9%). Each treatment was repeated three times. The data obtained were analyzed using analysis of variance (ANOVA). If there is a significant difference between treatments, it is continued with the Duncan test (DMRT).

2.1. Tools and Materials

The raw materials used were juwet obtained from the Mangga Dua market in Surabaya, lactic acid bacteria (*Lactobacillus bulgaricus* : *Streptococcus thermophilus* : *Lactobacillus Acidophillus* 1: 1 : 1) obtained from Gadjah Mada University, sucrose (sugar) with the brand Gulaku and skim milk with the brand Ammerland 25/ls, DPPH solution obtained from the food analysis laboratory of UPN Veteran JATIM, and Iodine, starch, methanol, aquades and phenolphthalein indicator obtained from the Fajar Kimia store. The tools used were an autoclave, laminar air flow, incubator, micropipette, analytical balance, viscometer, pH meter, micro kjehdahl unit and glassware for analysis.

2.2. Implementation of Research

2.2.1. Preparation of Juwet Fruit Juice

The process of making juwet fruit juice involves cutting the juwet fruit, then crushing it using a blender (ratio of juwet fruit: water 1:3), then filtering the juwet fruit juice with juwet fruit dregs.

2.2.2. Preparation of Juwet Yoghurt

The process of making yoghurt is done by mixing juwet fruit juice with sugar (10%) and skim milk (according to the treatment of 5%, 7%, and 9%), then pasteurization is carried out at a temperature of 70ºC for 15 minutes, then cooling is carried out to a temperature of 40ºC, then culture inoculation is carried out (according to the treatment of 5%, 7%, and 9%), then incubation is carried out at a temperature of 37ºC for 18 hours.

3. RESULT AND DISCUSSION

3.1. Raw Material Analysis

Table 1. Juwet Fruit Raw Material Analysis

Parameter	Analysis Result	
	Analysis	Literature
pH	3.47	3.77
Antioxidan (%)	65.52	65.46
Vitamin C (mg/100g)	17.19	17.41
Anthocyanin (mg/100g)	18.73	18.89

The results of the analysis in Table 1 show that the raw material of juwet fruit in 100 grams has a pH of 3.47, has antioxidants of 65.52%, has vitamin C of 17.19mg, and has anthocyanin of 18.73mg. While in the literature [8] shows that juwet fruit in 100 grams has a pH of 3.77, has antioxidants of 65.46%, has vitamin C of 17.41mg, and has anthocyanin of 18.89mg.

3.2. Analysis of Juwet Yoghurt

Table 2. Analysis of Juwet Yoghurt

Treatment		Protein Content (%)	Total Titrated Acid (%)	Total Dissolved Solid (°Brix)	Total LAB (Log CFU/ml)	pH	Antioxidant (%)	Viscosity (%)	Vitamin C (%)
Skim Milk	Starter								
5%	5%	1.1	1.5	15.3	8.7	4.4	7.1	3.9	4.5
5%	7%	1.2	1.6	15.7	9.2	4.2	10.5	4.1	5.3
5%	9%	1.5	1.9	16.0	9.8	4.2	14.6	4.4	5.5
7%	5%	1.6	1.6	16.7	9.0	4.1	16.5	4.9	5.2
7%	7%	1.8	1.6	17.0	9.5	4.1	17.3	4.9	5.4
7%	9%	1.8	1.7	18.0	10.1	3.8	17.9	5.1	5.6
9%	5%	1.8	2.0	18.3	9.9	3.7	18.6	5.8	5.6
9%	7%	2.0	2.1	18.3	10.2	3.7	21.4	6.5	5.7
9%	9%	2.1	2.1	22.3	10.5	3.7	34.0	6.8	5.8

Note: Mean values accompanied by different letters indicate significantly different results at $p \leq 0.05$.

3.2.1. Protein Content

Table 2 shows that the higher the addition of skim milk and the addition of starter, the higher the protein content of juwet yoghurt. According to [9] skim milk contains protein (3.3 grams) per 100 grams while according to [10] BAL starter also contains protein (12.5 grams). The higher the addition of skim milk and BAL starter will increase the soluble protein of the yoghurt produced. The addition of skim milk will add nutrients (lactose) for the growth of BAL, and the more lactose is available, the faster the BAL will grow. This is supported by the statement [11] which states that the growth of BAL which increases along with the increase in proteolytic enzymes results in more hydrolyzed protein and soluble protein also increases. Increasing the concentration of starter also increases the number of lactic acid bacteria which will carry out fermentation activities on the content in the product and break down protein bonds, resulting in more soluble protein.

3.2.2. Total Titrated Acid

Table 2 shows that the higher the addition of skim milk and starter, the higher the total titrated acid of juwet yoghurt. The total titrated acid ranges from 1.53% -2.10%. This is because the more skim milk and starter, the more the process of breaking down lactose into lactic acid. The higher fermentation of lactose by LAB produces lactic acid as the main product. Lactic acid lowers the pH of the environment, which inhibits the growth of pathogenic bacteria and spoilage bacteria, increasing the durability of yoghurt products. In addition, lactic acid also plays a role in providing a distinctive sour taste to yoghurt products. This is supported by literature [12] which states that the increase in lactic acid levels is due to the activity of LAB which breaks down lactose and other sugars into lactic acid. LAB activity will affect the acidity level of yoghurt because of the metabolite product in the form of lactic acid.

3.2.3. Total Dissolved Solids

Table 2 shows that the higher the addition of skim milk and starter, the higher the total dissolved solids of juwet yoghurt. This is because the higher the addition of skim milk and starter, the more lactose can be converted into lactic acid for the growth of LAB so the more lactose, the higher the total dissolved solids. According to Ref. [13] total solids are the solid part consisting of mixed materials and the nutrients contained therein, namely fat, protein, carbohydrates, minerals, vitamins and soluble fiber. The remaining total sugar, lactic acid and organic acids formed are counted as total dissolved solids.

The fermentation process causes bacterial activity in breaking down the yoghurt media substrate to increase so that products such as carbohydrates and proteins will be broken down into simpler compounds, causing the total dissolved solids to increase. The fermentation process of lactic acid bacteria will degrade more substrates so that they will produce higher solubility than before so that the final total solids will increase. The total dissolved solids value will affect the texture of the yoghurt produced. According to [14] total dissolved solids also come from the decomposition of proteins into simple molecules and are soluble in water such as amino acids and peptones. According to [1] the total solids of yoghurt are at least 8.2% while the total solids of juwet yoghurt obtained the lowest value of 15.33% and the highest value of 22.67%..

3.2.4. Total Lactic Acid Bacteria

Table 2 shows that the higher the addition of skim milk and the addition of starter, the higher the total LAB of juwet yoghurt. This is because lactic acid bacteria utilize lactose from skim milk as a source of energy for their growth, so the total LAB is higher. According to Ref. [14] an increase in the concentration of skim milk can increase the growth of LAB. Lactose in skim milk acts as a source of energy and carbon. LAB will break down lactose into glucose and galactose to produce lactic acid. Testing the total LAB in juwet yoghurt drinks using a dilution factor of 10^{-7}. The total LAB in juwet yoghurt increases due to the higher addition of skim milk and the higher LAB starter. According to [15] the higher the concentration of skim milk and the concentration of the starter added, the higher the number of bacteria that will grow so that the total number of lactic acid bacteria also increases. Lactic acid bacteria are thought to utilize the lactose contained in yoghurt as a source of energy for their growth which will ultimately produce lactic acid. According to Ref. [1] the minimum dilution factor for BAL testing is 10^{-7}.

3.2.5. pH

Table 2 shows that the higher the addition of skim milk and the addition of starter, the lower the pH value of juwet yoghurt. This is due to lactic acid bacteria breaking down lactose into lactic acid, thereby lowering the pH of yoghurt. The fermentation process in milk will produce organic acids which will cause the pH of the milk to drop until it reaches the isoelectric point of milk protein (around 4-4.5). This is supported by literature [16] which states that the pH of newly formed yoghurt products can reach 3.65 - 4.40. If the incubation process is continued, the pH of yoghurt can drop to 3.50 with an increase in lactic acid of up to 2 percent. The decrease in pH in yoghurt products during incubation occurs due to the accumulation of lactic acid produced by the activity of the culture.

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3.2.6. Antioxidant

Table 2 shows that the higher the addition of skim milk and the addition of starter, the higher the antioxidants of juwet yoghurt. This is because Vitamin C in skim milk can function as an antioxidant that can inhibit cell damage due to oxidation. The levels of vitamin C and anthocyanins found in juwet yoghurt play an important role in increasing antioxidant activity. Vitamin C not only acts as a direct antioxidant but also helps strengthen and maintain antioxidant activity. The combination of these two components in juwet yoghurt can significantly increase protection against oxidative damage and provide greater health benefits. This means that lactic acid can slow down the degradation or decomposition of antioxidants, thereby extending the shelf life and effectiveness in fighting free radicals. According to [17] in yogurt fermentation, the amount of acid excreted by lactic acid bacteria will increase due to the process of acid accumulation in this substrate is marked by a decrease in the pH of the substrate, so the increasing amount of lactic acid can increase antioxidant activity.

The greater the difference in addition, the faster the diffusion process will be. During yoghurt fermentation, the amount of acid excreted by lactic acid bacteria will increase due to the process of acid accumulation in the substrate, the increase in acidity in the substrate is marked by a decrease in the pH of the substrate, so the increase in the amount of lactic acid can increase antioxidant activity. In juwet fruit which has a blackish-purple color also has high anthocyanin compounds. This is supported by the literature [18] which states that the blackish purple color of juwet fruit is caused by high anthocyanin content.

3.2.7. Viscosity

Table 2 shows that the higher the addition of skim milk and starter, the higher the viscosity of juwet yoghurt. This is because the more skim milk and starter, the more lactose is converted into lactic acid which causes the protein contained in yoghurt to coagulate, increasing the viscosity of yoghurt. According to Ref. [19] states that yoghurt viscosity describes the properties of a liquid that has resistance to a flow that can withstand relative movement. The viscosity of yoghurt is influenced by pH, protein content, type of strain culture, incubation time and total milk solids.

3.2.8. Vitamin C

Table 2 shows that the higher the addition of skim milk and starter, the higher the vitamin C of juwet yoghurt. This is because the more skim milk and starter added can maintain the vitamin C content. According to [9] skim milk has 1µg of vitamin C in 100 grams. The content of juwet in 100 grams has 14.3mg of vitamin C. Lactic acid bacteria convert lactose into lactic acid, which lowers the pH of milk and helps form gel in yoghurt. This fermentation process does not directly affect the vitamin C content, but changes in pH can affect the stability of vitamin C. While skim milk can affect the consistency and quality of yoghurt. Vitamin C can decrease during the heating and storage process. According to [20] vitamin C in a food ingredient will decrease if the temperature is increased.

3.3. Organoleptic Test Results of Juwet Yogurt

Table 3 shows the Organoleptic Test Results of Juwet Yogurt based on the treatment.

Table 3. Organoleptic Test Values of Juwet Yogurt with the Addition of Skim Milk and the Addition of Starter

Treatment		Colour	Taste	Flavor	Texture
Skim Milk	Starter				
5%	5%	2.48	2.6	2.88	2.92
5%	7%	2.52	2.64	3.12	3
5%	9%	2.56	2.68	3.04	3.12
7%	5%	2.64	2.76	2.76	3.28
7%	7%	2.68	2.88	3.04	3.36
7%	9%	2.72	2.96	3.08	3.52
9%	5%	2.76	3.04	2.52	3.56
9%	7%	2.84	3.16	2.72	3.64
9%	9%	2.88	3.24	2.68	3.76

Note: The higher the value, the more the panellists prefer it

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3.3.1. Color

Table 3 shows that the panellists' preference level for the organoleptic color of juwet yoghurt has a value between 2.48-2.88 (dislike-quite like). The highest panellists' preference level was for the addition of 9% skim milk and the addition of 9% starter with an average value of 2.88, while the lowest panellists' preference level was for the addition of 5% skim milk and the addition of 5% starter with an average value of 2.48. This is following the literature [21] which states that color can be used as a quality parameter that is first considered by consumers before assessing other organoleptic qualities. Color can also provide clues about chemical changes in food such as browning and caramelization. The color of yoghurt products is generally almost the same as the raw materials, namely yellowish white. The effect of color on consumer acceptance is an important complement to quality so that it can indicate a quality product.

3.3.2. Flavor

Table 3 shows that the panelists' preference level for the organoleptic taste of juwet yoghurt has a value between 2.6-3.24 (dislike-quite like). The highest preference level of the panelists was in the addition of 9% skim milk and the addition of 9% starter with an average value of 3.24, while the lowest preference level of the panelists was in the addition of 5% skim milk and the addition of 5% starter with an average value of 2.6. According to [21] sour taste is caused by proton donors whose intensity depends on the H+ ions produced by acid hydrolysis. The sour taste of yoghurt is mostly caused by the presence of lactic acid produced by the starter culture during the fermentation process. This sour taste is what forms the uniqueness of yoghurt products.

3.3.3. Aroma

Table 3 shows that the panelists' level of preference for the organoleptic aroma of juwet yoghurt has a value between 2.52-3.12 (dislike-quite like). The highest level of panelists' preference was for the addition of 5% skim milk and the addition of 7% starter with an average value of 3.12, while the lowest level of panelists' preference was for the addition of 9% skim milk and the addition of 5% starter with an average value of 2.52. Indicating that many panellists did not like the aroma of the juwet yoghurt sample because of the very sour smell. This is supported by the literature [21] which states that a sour smell is a smell that is easily accepted by the human nose and brain, along with three other smells, namely fragrant, rancid and good.

3.3.4. Texture

Table 3 shows that the panellists' preference level for the organoleptic texture of juwet yoghurt has a value between 2.92-3.76 (dislike-quite like). The highest preference level of panellists was in the addition of 9% skim milk and the addition of 9% starter with an average value of 3.76, while the lowest preference level of panellists was in the addition of 5% skim milk and the addition of 5% starter with an average value of 2.92. This is because skim milk can increase the thick texture of juwet yoghurt. This is supported by the literature [22] which states that the addition of skim milk can increase the amount of coagulated protein in the fermentation process so that the resulting yoghurt texture is thicker.

4. CONCLUSION

The treatment of adding skim milk and adding starter significantly interacted with the levels of protein, total titratable acid, total soluble solids, total lactic acid bacteria, pH, antioxidants, viscosity, vitamin C of yoghurt, and the combination of these treatments significantly affected the scores of preference for color, taste, aroma, and texture of yoghurt. The results showed that the treatment of adding 9% skim milk and adding 9% starter was the best treatment that produced yoghurt with a protein content of 2.1%, total titratable acid 2.1%, total soluble solids 22.7ºBrix, total lactic acid bacteria 10.5 log CFU/ml, pH 3.7, antioxidants 34.0%, viscosity 6.8%, vitamin C 5.8% and hedonic organoleptic tests including color 2.88 (quite like), taste 3.24 (quite like), aroma 2.68 (quite like), and texture 3.76 (like).

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