

# Quality of Kefir Combination Between Sweet Corn Milk and Skim Milk on Variations of Sugar Types and Fermentation Duration

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**Abstract:** This study aims to determine the quality (total acid content and organoleptic quality) of kefir combination between sweet corn milk and skim milk on variations of sugar types and fermentation duration. The research method was experimental, with a completely randomized design (CRD) factorial pattern of 2 factors, namely the sugar types (ant sugar and granulated sugar) and the fermentation durations (36 hours and 48 hours), each treatment of 3 replications. Data analysis was the quantitative and qualitative descriptive methods. The results showed that the quality of kefir combination between sweett corn milk and skim milk was the K2T1 treatment (30 grams of granulated sugar + 36 hours fermentation) that was total acid content of 0.92%, pH of 3.83, and the organoleptic quality of slightly sour taste, white color, tasty aroma, smooth and thick texture, and favored acceptability.

**Keywords:** kefir; sweet corn; skim milk; sugar type; fermentation duration; total acid content; organoleptic.

# 1. INTRODUCTION

Probiotic bevarages are one of the fermented drinks with microbial agents that play a role in maintaining the health of the digestive tract (intestines) (Setiarto and Karo, 2021). Probiotic drinks have the potential to be developed as functional drinks. This is studied from the content in fermented foods or drinks that are beneficial for health (Aprilia, 2021). Kefir is one of the probiotic beverage products resulting from fermented milk with grain kefir starter, which consists of a group (biomass) of lactic acid bacteria, yeast, casein and polysaccharides (Hanum et al., 2021). Kefir contains protein with essential amino acids, vitamins (vitamins A, B1, B2, B5, B6, B7, B9, B12, C and vitamin K), and minerals (potassium, calcium, phosphorus, magnesium, iron, zinc, copper, and manganese) (Arvanta, 2021). According to the Indonesian National Standard (SNI), (2009) the total acid content of kefir is about 0.5% - 0.9%, pH 3.8-4.6, and protein 3.2%.

In general, cow's milk is the raw material in making kefir. However, not a few people are allergic to lactose contained in animal milk, so other alternative raw materials are needed in making kefir with vegetable milk (Muhsinin et al., 2020). One of the plant-based food ingredients as a kefir base is sweet corn. Sweet corn contains high starch (72-73%) consisting of amylose and amylopectin starch in the ratio of 25-30%: 70-75 %. Sweet corn contains 8-11% protein, unsaturated fatty acids (omega 9/oleic acid and omega 6/linoleic fatty acid), vitamins (A, K, Na, P, Ca and Fe), and minerals (Lestari et al., 2021). Sweet corn is a basic ingredient for making kefir because it contains carbohydrates and reduces sugars which are quite high. Sweet corn is utilized into vegetable milk and processed through a process so that it becomes a kefir probiotic beverage (Putri, 2017). The protein content in sweet corn milk tends to be lower when compared to cow's milk, so if it is used as a base material in





making kefir, it will be inhomogeneous and quickly settle.

The addition of skim milk in making kefir is necessary to maintain the stability and consistency of sweet corn milk (Hakiki et al., 2022). Sweet corn milk also does not contain lactose because the sugar in sweet corn is not the same as sugar in animal milk. Therefore, in making kefir, skim milk needs to be added, because skim milk contains lactose or milk sugar. Skim milk sugar will be utilized by LAB as an energy source, the more available the energy source, the faster the growth of LAB. Skim milk is also rich in carbohydrates and proteins which are good growth media for LAB. Skim milk contains 90.42% water, 3.68% protein, 0.10% fat, 5.00% lactose, and 0.80% ash (Rihastuti and Soeparno, 2018).

Sugar is a source of nutrients for lactic acid bacteria during the fermentation process. In 100 g of sugar or white sugar contains 394 kcal, 0g protein, 0g fat, and 94g carbohydrates (Basagili, 2018). Ant sugar contains Thiamin (Vitamin B1) which functions as a coenzyme in energy metabolism and contains Riboflavin (Vitamin B2) which helps form red blood cells. The nutritional content of ant sugar is 3.75% moisture content, 8.21% ash content, 98.68% total sugar, 95.79% sucrose, and 9.04% reducing sugar (Assah and Makalalag, 2021). Various microorganisms in kefir grains break down palm sugar (complex) into simple sugars. The simple sugars formed may not have been utilized by LAB entirely during the fermentation process (Setvoningsih et al., 2020).

Fermentation duration is one of the factors that can affect the results of fermentation products. The duration of fermentation will affect the decrease in pH due to the activity of LAB in breaking down carbohydrates into lactic acid (Kinteki *et al.*, 2019). The longer the fermentation duration, the more it increases the total acid and increases the total lactic acid bacteria (Rohman *et al.*, 2019). This can occur due to the availability of sufficient food for the growth and proliferation of LAB obtained from the added sugar.

This study aims to determine the quality (total acid content and organoleptic quality) of kefir combination between sweet corn milk and skim milk on variations of sugar types and fermentation duration.

### 2. **RESEARCH METHOD** *Time and Place of Reaserch*

The research was conducted during December 2023-March 2024 at the Industrial Microbiology Laboratory, Faculty of Teacher Education, Training and Universitas Muhammadiyah Surakarta. This research includes the process of making kefir with a combination between sweet corn milk and skim milk, testing pH parameters and organoleptic quality at the Industrial Microbiology Laboratory, Faculty of Teacher Training and Education, Universitas Muhammadiyah Surakarta. Testing the total acid content of kefir was carried out at Chem-Mix Pratama Laboratory, Yogyakarta.

# Tools and Materials

- a. Tools used in this study: pot, knife, scissors, refrigerator, stailness spoon, blender, sieve, measuring cup, pH stick, pH meter, measuring cup, beaker glass, glass bottle, sieve, spatula, spoon, analytical balance, thermometer, laminar air flow, incubator, sprayer, drop pipette, flakon bottle, glass jar.
- b. Materials used in this research: sweet corn milk, skim milk, distilled water, 70% alcohol, ant (palm) sugar, granulated sugar, kefir grain, NaOH, 70% ethanol, 1% PP indicator, aluminum foil, spiritus, label paper, tissue, plastic wrap.

# Research Design

This research method is experimental. The experimental design used Completely Randomized Design (CRD) factorial pattern of 2 factors and 3 replications each. The treatment factor consists of two factors, namely factor 1 sugar types: ant (palm) sugar and granulated sugar and factor 2 duration: fermentation (36 hours and 48 hours).

### Sweet Corn Milk Making

500 grams of sweet corn (best quality) was added with water in the ratio of 1:4. Then blended for  $\pm$  10 minutes or until smooth, filtered corn paste and corn dregs are set aside until the filtrate is obtained (which is sweet corn milk).





## Kefir Making

Sweet corn milk and skim milk (250 ml each) were pasteurized at 70 °C for 15 minutes, then put into glass bottles in a ratio of 1:1 (125 ml sweet corn milk: 125 ml skim milk), so the total volume was 250 ml. Adding 30 g of ant (palm) sugar to the treatment for 36 hours and 48 hours of fermentation (two treatment combinations). Furthermore, adding 30 g of granulated sugar to the treatments for fermentation duration of 36 hours and 48 hours (two treatment combinations). Add 8 g of starter (grain kefir) to each treatment combination, then cover with aluminum foil and wrap with plastic wrap. Next, incubate at room temperature and a sterile place for 36 hours and 48 hours according to the treatment.

# pH Test

The pH of the sample was measured using an AZ pH-meter. The electrode was rinsed with distilled water and dried with tissue paper, then the electrode was dipped in the sample and the pH-meter was set at pH measurement, the electrode was left for a while until the pH-meter needle stabilized and showed the pH of the sample (Yurliasni *et al.*, 2019).

# Total Acid Test Titration Method

The percentage of lactic acid was measured by titration method. A 10 mL sample was added with 2 drops of 1% phenolphtalein (PP) indicator and then titrated with 0.1 N NaOH solution until a pink color appeared. The amount of acid with the following formula:

Lactic acid(%) =  $\frac{mL NaOH \times 0.1 NaOH \times 90.08}{mL sample \times 1000} \times 100\%$ (Afiyah, et al., 2021)

# **Organoleptic Test**

The testing parameters using the hedonic test method consist of color, aroma, taste, texture and acceptability. Organoleptic quality testing was carried out by asking the panelists to give a value on the assessment sheet (questionnaire) that had been provided. The panelists used amounted to 15 panelists consisting of moderately trained and untrained panelists (Yudiastuti *et al.*, 2023).

# Data Analysis

Research data is presented in tabular form. Analysis of total acid testing with quantitative descriptive methods using the two-way analysis of variance (Two Way ANOVA) test. Testing organoleptic quality with qualitative descriptive methods.

# 3. **RESULTS AND DISCUSSION** *Results of Total Acid Content and pH*

The results of total acid content and pH of kefir combination between sweet corn milk and skim milk on variations of sugar types and fermentation duration can be seen in Table 1.

Table 1.	Results	of Total	Acid	Content	and pH
		of Ke	fir		

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Treatment	Total Acid Content (%)	рН					
K1T1	0.99*	3,8					
K2T1	0.92	3,83**					
K1T2	1.21	3,6*					
K2T2	1.18**	3,7					

Description: \*) Lowest score

\*\*) Highest score

K1T1: 30 g ant (palm) sugar + 36 hours fermentation duration

K2T1: 30 g granulated sugar + 36 hours fermentation duration

K1T2: 30 g ant (palm) sugar + 48 hours fermentation duration

K2T2: 30 g granulated sugar + 48 hours fermentation duration

### **Organoleptic Quality Result**

The results of organoleptic quality of kefir combination between sweet corn milk and skim milk on variations of sugar types and fermentation duration including color, texture, taste, aroma and acceptability can be seen in Table 2.





Table 2. Results of Organoleptic Quality of Kefir									
No	Treatment -	Aspect of Organoleptic Quality							
		Taste	Color	Aroma	Tekstur	Acceptability			
1	K1T1	Sour	Brown	Tasty	Smooth and thick	Dislikes			
2	K2T1	Slightly sour	White	Less tasty	Smooth and thick	Dislikes			
3	K1T2	Sour	Brown	Tasty	Smooth and thick	Dislikes			
4	K2T2	Sour	White	Tasty	Smooth and thick	Like			

Description:

K1T1: 30 g ant (palm) sugar + 36 hours fermentation duration

K2T1: 30 g granulated sugar + 36 hours fermentation duration

K1T2: 30 g ant (palm) sugar + 48 hours fermentation duration

K2T2: 30 g granulated sugar + 48 hours fermentation duration

#### DISCUSSION

#### Total Acid Content and pH

The results (Table 1) showed that the total acid content and pH of kefir were influenced by variations in sugar type and fermentation duration. The average value of total acid content is 0.99 -1.21% and the pH value of kefir is 3.6-3.83. The total acid content will increase along with the increase in fermentation duration, the higher the total acid content, the pH will decrease. The results of Stella's research (2019) showed that the more duration the fermentation, the activity of lactic acid bacteria increased. With increasing activity, the LAB produced also increases, causing the pH value to decrease. The activity of lactic acid bacteria is influenced by the availability of energy sources. The more energy sources available, the higher the total acid produced will be and will affect the decrease of pH in kefir. The energy source of lactic acid bacteria is obtained from sucrose in the added sugar. Lactic acid bacteria will use sucrose in sugar as an energy source and carbon source. Sucrose will be broken down by lactic acid bacteria through the permease system to form glucose and fructose through the glycolysis pathway to produce lactic acid (Salminen and Wright, 1998 in Hanzen, 2016).

### **Organoleptic Quality**

The results of organoleptic quality (Table 2) of kefir with a combination of sweet corn milk and skim milk, have varying assessments of each treatment. The taste of kefir combined with sweet corn milk and skim milk in the K2T1 treatment had a slightly sour taste, while in the K1T1, K1T2, and K2T2 treatments had a sour taste. The sour

taste in kefir is produced from the activity of lactic acid bacteria that break down lactose into lactic acid. According to Rumeen et al., (2018), the high acidity level in kefir is caused by the high content of lactic acid. Lactic acid bacteria produce the enzyme  $\beta$ -galactosidase which will convert lactose into lactic acid. The sourness of kefir will affect the aroma produced in each treatment. Kefir with a combination of sweet corn milk and skim milk K2T1 treatment has a less tasty aroma, while the K1T1, K1T2, and K2T2 treatments have a tasty aroma. This aroma can be formed due to the presence of lactic acid produced by LAB during the fermentation process. The results of Lestari's research (2018) stated that the sour aroma of kefir is formed due to the presence of lactic acid and acetic acid produced during the fermentation process. The number of volatile compounds such as lactic acid, acetic acid, and ethanol formed during fermentation affects the scent of the kefir aroma formed, so it will affect the hedonic results. This is supported by the opinion of Musdholifah and Zubaidah (2016) which states that the sour aroma of kefir is caused by the presence of volatile compounds formed in kefir, giving rise to a distinctive sour aroma in kefir.

Variations of sugar types will affect the total acid content and pH of kefir. The total acid content of kefir with added ant (palm) is higher (0.99% - 1.21%) compared to kefir with added granulated sugar (0.92% - 1.18%). This is because the sucrose content of ant (palm) sugar tends to be higher than that of granulated sugar, so that the energy source obtained by bacteria in kefir with added ant (palm) is higher than that of kefir with added granulated sugar. Ant sugar contains 81.78% sucrose (Handayani *et al.*, 2022). The



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results of <u>Widia's research (2018)</u> showed that palm sugar contains higher sucrose, which is 84% compared to cane sugar, which is 20%. This is supported by the opinion of Lempang (2012) in <u>Astuti (2020)</u> which states that palm sugar contains approximately 84% sucrose, while beet sugar and cane sugar only contain 20% and 17% sucrose, respectively.

The color of each treatment combination showed different results. Kefir combination between sweet corn milk and skim milk in the K1T1 and K1T2 treatments was brown, while in the K2T1 and K2T2 treatments it was white. This shows that the variations of sugar type affect the color produced in kefir. The results of research by Uswatiningty as (2023) show that the addition of different sugar types will affect the color produced in kefir results. Ant (palm) sugar has a brown color so that kefir with the addition of ant (palm) sugar (treatment K1T1 and K1T2) will produce a brown color. Granulated sugar is white so that the color of kefir in the K2T1 and K2T2 treatments is white. Color changes in fermented milk products are also influenced by the fat content and different milk solids (Mandang et al., 2016).

The texture of kefir combined with corn milk and skim milk showed the same results in all treatments, namely smooth and thick. The addition of skim milk affects the level of viscosity produced in kefir. This is in line with Lestari's research (2021) that the addition of skim milk in making corn milk kefir will affect the total solids of corn milk kefir. The higher the total solids of corn milk kefir, the thicker the texture of the resulting milk kefir product. The texture formed in sweet corn milk kefir bevarage is caused by protein coagulation due to the accumulation of acid due to the formation of lactic acid by lactic acid bacteria during the fermentation process (Ingga et al., 2019). The acceptability of kefir combination between sweet corn milk and skim milk showed different results. Panelists tended to like kefir in the K2T2 treatment because it had a sour taste, tasty aroma and smooth and thick texture.

The results of the 4 samples tested in the K2T2 treatment (30 g granulated sugar + 48 hours fermentation duration) showed the best results with a slightly sour taste, white color, tasty aroma, smooth and thick texture, and favorable

acceptance. The results of organoleptic quality of kefir in the K2T2 treatment are in accordance with <u>SNI (2009)</u> which explains that the standard criteria for physical characteristics in kefir must have the appearance of a thick-solid liquid, a normal/typical aroma, and a sour/typical taste.

### 4. CONCLUSION

The best quality of kefir combination between sweet corn milk and skim milk was treatment K2T1 with total acid content of 0.92% and pH 3.83. The best organoleptic quality of kefir combination between sweet corn milk and skim milk was the K2T2 treatment with a slightly sour taste, white color, tasty aroma, smooth and thick texture, and favorable acceptance.

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