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## **Glucose Levels and Organoleptic Quality Probiotic Tepache of Pineapple Peel on Variation of Sugar and Fermentation Duration**

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### **ABSTRACT**

**KEYWORDS:**

*Fermentation  
Pineapple peel  
Probiotic  
Sugar  
Tepache*

The probiotic beverage tepache is a variety of natural or processed functional products that contain bioactive compounds with specific physiological functions for health. Pineapple peel is one of the plant base ingredients that can be used as a base for probiotic beverages. The levels of antibacterial compounds, carbohydrates, and sugars in pineapple peel are sufficient to be an essential medium for the growth of Lactic Acid Bacteria (LAB) in probiotic beverages. This research was aimed to determine the glucose levels and organoleptic quality of probiotic beverages tepache of pineapple peel on variation of sugar and fermentation duration. This research used experimental method Completely Randomized Design (CRD) with two-factors, Factor 1 was variation of sugar (G) palm sugar and granulated sugar. Factor 2 was the variation of fermentation duration (F) 3 and 5 days. The results of the research showed that the tepache of pineapple peel had the highest glucose levels in the treatment of G1F1 (palm sugar 100 g with 3-days fermentation duration) at 12,2 %. The best organoleptic quality of tepache pineapple peel was in G2F1 treatment (granulated sugar 100 g with 3-days fermentation duration) with light yellow color, tepache-specific aroma, and sour flavor.

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## **1. INTRODUCTION**

Probiotic beverages are a type of functional food with physiological benefits for the body's health. In addition, it can reduce the risk of hypercholesterolemia, hypertension, diabetes, and obesity, and suppress inflammation in the intestines (Refita, 2021). Furthermore, certain functions of probiotic beverages may reduce lactose intolerance reactions and inhibit the proliferation of diarrhea-causing pathogens (Sultan, 2022). Dairy fermentation products such as yogurt and kefir are the most well-known probiotic beverages. However, fermented milk beverages are relatively more expensive and can trigger lactose intolerance in some individuals. To accomplish this, other plant-based alternatives must be used as raw materials in producing probiotic beverages so that they can be made at a price that is more affordable and relatively safe for public health. Oranges, apples, guava, and pineapples can be used as the base components for probiotic beverages, then further developed through extensive research. Tepache is one of the probiotic beverages produced by the pineapple peel.

Tepache is a traditional Mexican beverage made possible by developing functional products and nutritious ingredients. The pineapple rind accounts for 25 % of the fruit's weight. 81,72 % of pineapple peel is water, 20,87 % of crude fiber, 17,53 % of carbohydrates, 4,41 % of protein, and 13,65 % of reduced sugar (Setyawati & Rahman, 2017). Due to its high glucose content, pineapple peel is an essential medium for developing Lactic Acid Bacteria (LAB). Pineapple peel contains flavonoids, tannins, saponins and bromelain. Bromelain is effective against both aerobic and

anaerobic bacterial isolates (Putri et al., 2016). The fermentation of pineapple peel with lactic acid microorganisms produces an alternative functional beverage. Tepache is produced by placing fruit flesh and pineapple peel in a container with brown sugar to encourage spontaneous fermentation (Ojeda-Linares et al., 2021).

Palm and granulated sugar are the forms of sugar used by the community. The top of the palm tree stalk is tapped to obtain palm sugar. The sap water from the lead is heated to a shallow moisture content (6 %) so that the sap will harden throughout the water-cooling process. Palm sugar contains vitamin B complex, glucose, and mineral salts (Lalisang, 2018). Palm sugar has a lower glycemic index value than granulated sugar, measuring 35 glycemic index with a total of 368 calories per 100 g of ingredients, compared to granulated sugar's glycemic index value of 58 with a total of 400 calories per 100 g of ingredients. This glycemic index regulates triglyceride levels in the blood (Aprilia & Suryana, 2022).

Granulated sugar is refined, dehydrated sucrose crystal sugar derived from sugar cane. Several steps are involved in producing granulated sugar: preparation, grinding, sap purification, crystallization, centrifuging, and drying. Due to the minimal production process and low temperatures, the phytonutrient content of palm sugar is more stable than granulated sugar. The addition of pineapple peel and sugar in a ratio of 3:1 results in a balanced flavor between the acidic pineapple peel and the sweet sugar; the aroma and color of golden brown enhance the appealing qualities of tepache beverages (Sukriadi et al., 2022). High concentrations of sucrose in palm sugar and granulated sugar nourish lactic acid bacteria, affecting their activity during fermentation.

The fermentation duration depends on the incubation period of lactic acid bacteria and determines the properties of the fermented beverage. Several variables, including microbial population, pH, substrate, temperature, alcohol concentration, oxygen, salinity, water, and fermentation duration, affect the fermentation process (Widyantara et al., 2020). The longer the duration of the fermentation process, the lower the pH in a fermented beverage, the concentrations of lactic acid, acetic acid, butyric acid, and propionic acid increase as pH decreases. The production of acid results from the conversion of glucose to alcohol. (Simanjuntak et al., 2017). This increase in sourness reduces the panelists' evaluations of flavor. The bacterial curve during the lengthy fermentation period has three phases. During the logarithmic phase, the number of bacteria increases; during the stationary phase, the number of living and deceased bacteria equals; and during the death phase, the number of lactic acid bacteria decreases (Masengi et al., 2020).

In Indonesia, probiotic tepache beverages have not been the subject of thorough studies. In the research conducted by Hujjatusnaini et al., 2022, pineapple peel tepache probiotic beverage was combined with a LAB starter in the form of *Lactobacillus casei* and sugar concentration solutions of varying concentrations. Probiotic beverage tepache pineapple peel with added sugar can undergo spontaneous fermentation due to the presence of microorganisms such as *Lb. Plantarum*, *Leuconostoc mesenteroides*, and yeast *Saccharomyces cerevisiae* var. *boulardii* in the pineapple peel. (Romero-luna et al., 2017) The pineapple peel tepache probiotic beverage is usually made with piloncillo sugar, a form of brown sugar. This research aims to determine the glucose level and organoleptic quality of probiotic tepache pineapple peel on variations of sugar types (palm and granulated sugar) and fermentation durations (three days and five days fermentation).

## 2. MATERIALS AND METHODS

This research was conducted in February 2023 at the UMS Biology Laboratory on Ahmad Yani Tromol Street, Post 1, Pabelan Kartasura. In the meantime, testing for glucose levels was conducted in March 2023 at the Chem-Mix Pratama Laboratory in Bantul, Yogyakarta. Manufacturing pineapple peel tepache fermented beverages requires 1000 ml glass jars, 500 ml plastic measuring cups, test containers, digital scales, pH meters, water baths, vortices, centrifuges, and spectrophotometers. Probiotic beverages tepache pineapple peel was produced with honey pineapple peel, honey pineapple flesh, palm sugar, granulated sugar, and distilled water.

This research employs an experimental method. The experimental design was a Complete Randomized Design (CRD) with two factors: sugar type (G1 = 100 g of palm sugar, G2 = 100 g of granulated sugar) and the duration of fermentation (F1 = 3 days, F2 = 5 days). The research was conducted in the following stages: 1. sterilization of equipment, 2. preparation of materials and tepache solution, 3. production of tepache, 4. pH measurement, 5. glucose level testing, 6. organoleptic test. A pH meter is utilized for the pH test. The parameter test: glucose levels and organoleptic quality. The glucose level test is carried out by the spectrophotometric, Nelson-Somogy method at 540 nm absorption wave. The data was collected using experimental techniques, surveys, literature, and documentation, while organoleptic quality and acceptability tests were analyzed qualitatively and descriptively using questionnaire sheets conducted on 15 panelists with an age range of 20 – 25 years. Quantitative descriptive methodologies applied to glucose levels testing analysis. The organoleptic test consists of color, aroma, flavor, and acceptability with hedonic scale. Quantitative data analysis utilizing two-track variance, the Kruskal-Wallis test, and qualitative data utilizing Microsoft Excel.

## 3. RESULTS AND DISCUSSION

The results of a research conducted on the pH and glucose levels of probiotic beverages tepache pineapple peel on variation of sugar and fermentation duration show the following values:

### 3.1. pH and Glucose Analysis of Probiotic Beverage Tepache Pineapple Peel on Variations of Sugar Types and Fermentation Duration.

**Table 1.** pH Values and Glucose Levels of Probiotic Beverage Tepache Pineapple Peel on Variations of Sugar Types and Fermentation Duration.

No	Treatment	pH	Glucose (%)	Annotation
1	G1F1	3.4**	12.22**	Palm sugar 100 g with a fermentation duration of 3 days
2	G1F2	3.3	8.46	Palm sugar 100 g with a fermentation duration of 5 days
3	G2F1	3.2	8.43	Granulated sugar 100 g with a fermentation duration of 3 days
4	G2F2	3.14*	5.91*	Granulated sugar 100 g with a fermentation duration of 5 days

Notes: \*) pH value and lowest glucose level

*\*\*\*) pH value and highest glucose level*

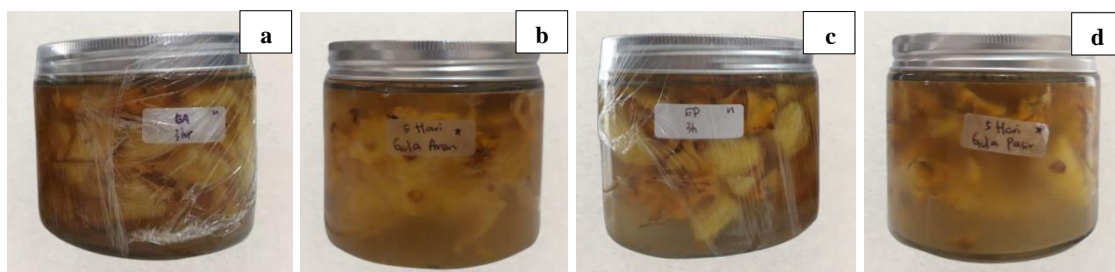
Table 1. shows that the glucose levels of pineapple peel tepache on sugar and fermentation duration variations have different values. The highest glucose concentration in pineapple peel tepache treated with G1F1 was 12.22 % per 100 g of palm sugar fermented for three days. In contrast, the lowest glucose level of pineapple peel tepache in the G2F2 treatment is 5.91 % per 100 g of granulated sugar after five days of fermentation. Within Table 1. the concentration of glucose decreases. The longer pineapple peel tepache is fermented, the lower its glucose concentration. The glucose decrease caused by Lactic Acid Bacteria (LAB) has not effectively decomposed the sucrose levels. Lactic Acid Bacteria is in its beginning growth phase (lag phase), an adaptation phase on the third day. Lactic Acid Bacteria begin experiencing rapid growth (exponential phase) on the fourth day. During the exponential phase, additional glucose substrate must be converted into lactic acid to sustain the growth and division of bacterial cells, resulting in a decrease in glucose levels. This phase involves the division or proliferation of bacteria because optimal conditions exist (Astuti et al., 2020). On day five, treatment with G1F2 and G2F2 decreased glucose levels. The glucose decrease indicates that bacteria are in an exponential growth phase, in which sucrose is broken down into glucose and glucose is converted into lactic acid and other organic acids used for bacterial growth. The decrease in glucose explains that bacteria and microorganisms require sugar as a carbon source for nutrition, which is then converted into organic acids and alcohol (Puspitasari et al., 2017).

The type of sugar in probiotic tepache pineapple peel affects the glucose levels in the beverage. Due to the hydrolysis of sucrose into simpler sugar components such as glucose and fructose under acidic conditions, reduced sugar levels decrease (Diniyah et al., 2013). This explains why G1F1 and G1F2 treatments have higher glucose concentrations than G2F1 and G2F2 treatments. Palm sugar has a more significant concentration of sucrose than granulated sugar. One hundred grams of palm sugar contain 84.31 percent sucrose, whereas 100 g of refined sugar contain 71.89 percent (Hendry et al., 2021). The cooking process in palm sugar can inhibit the activity of the invertase enzyme, resulting in less harm to the produced sucrose and a lesser decrease in pH value (Wilberta et al., 2021). During the fermentation process, the pH value serves as an indicator because it affects the tepache's aroma, flavor, and acceptability. The optimal pH for LAB activity in acidic conditions is 4 (Hujjatusnaini et al., 2022). Therefore, the optimal pH range for pineapple peel tepache is between 2.8 and 3.8. All formulations of pineapple peel tepache beverage achieve the minimum pH value of tepache, with G1F1 having the highest pH at 3.44 and G2F2 having the lowest pH at 3.14. The LAB fermentation process converts carbohydrates into lactic acid, increasing lactic acid and decreasing pH (Rizal & Nurdin, 2020). Therefore, the more fermentation duration, the lower pH of pineapple peel tepache. Additionally, the variety of sugar determines the pH of pineapple peel tepache. Sucrose concentration is a nutrient for LAB that regulates the increase in lactic acid and decrease in pH (Dipu et al., 2016).

According to the results of statistical studies, the influence of glucose levels with variations in sugar types and fermentation duration has a probability value of 0.012 at a level of Sig. based on the significance level of  $0.012 < 0.05$ , this indicates that glucose levels significantly affect sugar type and fermentation duration variations.

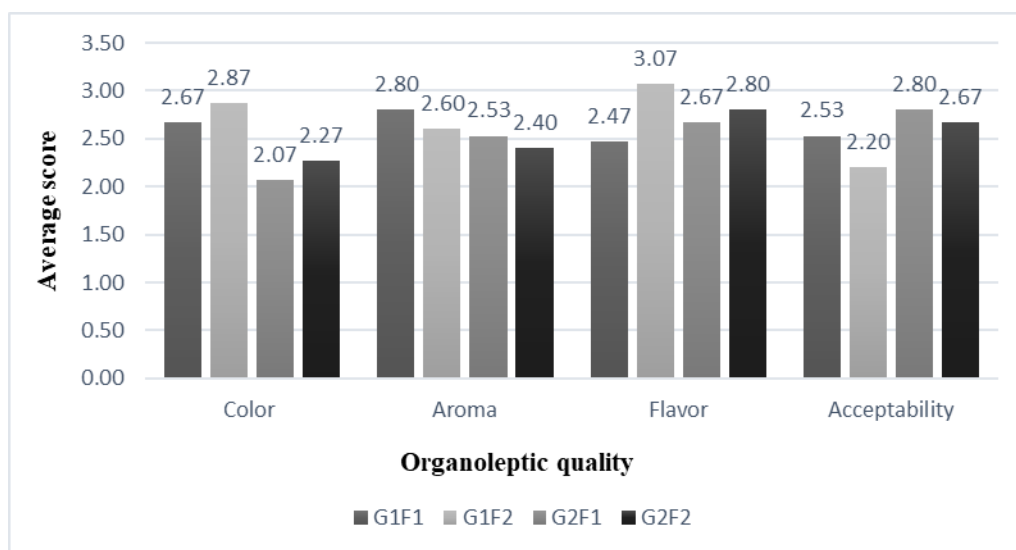
### 3.2. Organoleptic Quality of Probiotic Beverages Tepache Pineapple Peel on Variations of Sugar Types and Fermentation Duration

The results of organoleptic quality of probiotic beverages tepache pineapple peel on variations of sugar types and fermentation duration there are color differences as shown in figure 1. as follows:



**Figure 1.** The organoleptic color quality of tepache beverages pineapple peel on variations sugar and fermentation duration: (a) G1F1; (b) G1F2; (c) G2F1; (d) G2F2.

The organoleptic quality of probiotic beverages tepache pineapple peel on variations of sugar and fermentation duration with aspects of aroma, flavor and acceptability can be seen in Figure 2. as follows:



**Figure 2.** Results of Probiotic Beverages Organoleptic Quality Tepache Pineapple Peel on Variations of Sugar Types and Fermentation Duration.

#### 3.2.1. Color

The color quality of G1F1 and G1F2 treatments for pineapple peel tepache beverages is brownish yellow, whereas G2F1 and G2F2 treatments are light yellow. In light yellow color, the G2F1 treatment (100 g of granulated sugar with a 3-days fermentation duration) was favored by panelists for its superior color quality. The color of pineapple peel tepache is determined by the variety of pineapple peel and sugar used as ingredients. The treatment of G1F1 and G1F2 has a brownish-yellow color due to the effect of the brown color of palm sugar. On the other hand, due

to the effect of clear cane sugar sap, the G2F1 and G2F2 treatment has a light yellow color, resulting in the white color of granulated sugar (Milaniyah et al., 2022).

### 3.2.2. Aroma

The aroma quality of pineapple peel tepache beverage treated with G1F1, G1F2, and G2F1 is characterized by the aroma of tepache. However, the G2F2 treatment (100 g of granulated sugar with a 5-days fermentation duration) has a tepache-specific aroma. The panelists found the aroma of pineapple peel tepache to be fresh and aromatic, as is typical of pineapple peel. Due to LAB metabolism, lactic acid gives probiotic products a typical aroma (Utami, 2018). This indicates the more fermentation duration, the sharper aroma and the lower panelists' preference for the aroma.

### 3.2.3. Flavor

Figure 2. shows the flavor characteristics of the pineapple peel tepache beverages. According to the results of organoleptic tests, the average preference of panelists for the flavor of tepache pineapple peel probiotic beverages ranges between 2.47 and 3.07 (quite acidic - acidic). The G2F1 treatment (100 g of granulated sugar with 3-days a fermentation duration) resulted in the highest flavor value of 2.67 (acidic). The flavor of the probiotic beverage tepache pineapple peel is affected by the ingredients used as its base (Kusuma et al., 2019). Each treatment, including palm and granulated sugar balances the sour flavor of pineapple peel with a sweet flavor. However, the duration of fermentation reduced the panelists' organoleptic quality. This is because the fermentation process generates lactic acid and reduces the minimal pH value, enhancing the sour flavor (Rahayu et al., 2021).

### 3.2.4. Acceptability

Acceptability is the ability to determine whether the community acknowledges a product. The G2F1 treatment (100 g of granulated sugar with a 3-days fermentation duration) produced the pineapple peel tepache probiotic beverage the panelists preferred the most. It had an average value of 2.8 and a light-yellow color, tepache aroma, and acidic flavor. The public favors G1F1 and G2F2 treatment with acceptability scores of 2.53 and 2.67. The treatment that they did not like much by the panelists was G1F2 (100 g of palm sugar with a 5-days fermentation duration). which had an average score of 2.20 with aspect a brownish-yellow color, an original tepache aroma, and a sour flavor. This is because palm sugar is the less preferred form of sugar in the G1F2 treatment. Palm sugar lends tepache a brownish color, a sour flavor, and an aroma that panelists don't like much (Iskandar et al., 2015). The panel of panelists do not like much the 5-days fermentation duration much because it gave the tepache sourer flavor and sharper aroma.

## 4. CONCLUSION

Based on the results and discussion of the research, it can be concluded that the G1F1 treatment (100 g of palm with a 3-days fermentation duration) produced the highest glucose level of 12.22 % and the G2F1 treatment (100 g of granulated with a 3-days fermentation duration) produced the highest organoleptic quality with a light-yellow color, tepache-specific aroma, and sour flavor.

## 5. ACKNOWLEDGMENTS

The authors thank the Biology Education Program at Muhammadiyah Surakarta University for providing financial support INCOBEST article, as well as Mrs. Titik Suryani for guiding the research.

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