

# The Characteristics of Pineapple Probiotic Drinks Enriched by Inulin's Mangrove Apple

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## Abstract:

Probiotic drinks are functional drinks made from the fermentation process of lactic acid bacteria. (LAB) This drink usually uses milk-based ingredients, but diversification of vegetable-based products is carried out with pineapple because it contains sugar, and vitamins, and has an attractive color and aroma. The probiotic drink was given the addition of a prebiotic in the form of inulin. This research aims to determine the characteristics of pineapple probiotic drinks enriched with inulin's mangrove apple. The research procedures carried out were making pineapple juice, making inulin's mangrove apple, and making pineapple fruit probiotic drink which enriched by inulin's mangrove apple. The optimization of inulin's mangrove apple concentration was significant ( $P < 0.05$ ) on the characteristics of pineapple fruit probiotic drinks. The pH values of 3.70-4.10, total sugar 9.30-10.53°brix, and total LAB  $9.5 \times 10^7$ – $4.33 \times 10^8$  CFU/mL. There was a significant effect ( $P < 0.05$ ) on the aroma, taste, and overall variables in the hedonic test results and no significant effect on the color. The concentration of 3% inulin's mangrove apple is the best concentration.

**Keywords:** Inulin, *Mangrove* apple, pineapple, probiotics.

## Introduction

Over time, public awareness regarding the importance of health is increasing. People are starting to pay attention to functional food because it is felt that consuming functional food can positively improve body health. One of the functional food products is a probiotic drink. Probiotic drinks can positively affect body health when consumed regularly because they are made through a fermentation process with lactic acid bacteria [1]. The requirements for strains that can be used as probiotic agents are resistant to bile and acid so that they can reach the intestine and have the ability to stick to the intestinal mucosa [2]. Consumption of probiotic drinks regularly can maintain microflora balance by suppressing the growth of harmful bacteria and perfecting the human digestive process by protecting the digestive tract from pathogenic bacteria. [3].

Probiotic drinks are generally made from animal-based ingredients, namely milk. Some people cannot consume cow's milk, so product diversification is carried out with fruit juices that are rich in nutrients such as rich in dietary fibre, vitamins, low in fat, and also have an attractive aroma so that they are suitable as ingredients probiotic drinks [4]. One of the tropical fruits that has the potential to be used as the base for probiotic drinks is pineapple. Pineapple (*Ananas comosus* (L) Merr.) is a tropical fruit and has become a commercial fruit with high nutritional value and quite an excellent sensory [5]. Pineapple is a perishable food commodity and can only last for 4-6 days because of its high water and sugar content. The water and sugar content in 100 grams of pineapple is 86 g of water and 13.12 g of carbohydrates [6]. Pineapple fruit that is quite widely produced in Indonesia is the Queen type honey pineapple and the Cayenne type [7]. Queen honey pineapple has a yellower color and sweeter taste than Cayenne pineapple because it contains more sugar than the water, namely the amount of sugar reaches 13-18°brix [8]. The sugar content in pineapple is used as a carbon compound for the metabolism of lactic acid bacteria. The more sugar concentration, the total lactic acid bacteria will increase [9]. LAB in large quantities can provide a beneficial effect for those who consume it. Lactic acid bacteria, as probiotic agents, can inhibit pathogenic bacteria's

growth and function as immunomodulators [10]. Immunomodulators improve the immune system by stimulating (immunostimulants) or suppressing abnormal immune reactions [11]. The amount of water-soluble dietary fiber in mangrove apple extract shows that mangrove apples contain inulin which is included in the fructan type polysaccharide in water-soluble dietary fiber. The application of inulin's mangrove apple to food products is still lacking, so it is necessary to research the addition of inulin's mangrove apple to pineapple fruit probiotic drinks.

To add functional value, probiotic drinks will be better if given the addition of prebiotics that can increase body immunity. The prebiotics most often added to low pH drinks, such as probiotic drinks, are inulin, FOS, GOS, and resistant starch [12]. Inulin can be obtained from local food plants, which is apple mangrove extract. Apple mangrove (*Sonneratia caseolaris*) is a mangrove plant that generally grows in the tropics and belongs to the *Sonneratiaceae* species [13]. Mangrove apples, both in fruit and extracts, also contain 5.08% inulin, 9.67% soluble dietary fibre from inulin and inulin fructans, and 0.66% FOS [14]. This research aims to determine the characteristics of pineapple probiotic drinks enriched by inulin's mangrove apple

## Materials

The materials used in this research were Queen honey pineapple fruit obtained from the Banyumanik market, Central Java with a harvest age of 4 months. Mangrove apples obtained from Purworejo Beach with a harvest age of 3 months, sucrose, tomatoes, glucose, starter Pure *Lactobacillus plantarum* from Gadjah Mada University Laboratory, yogurt starter powder, water, 70% ethanol, buffer solution pH 4.7, and 10, NaCl solution, aquadest, MRSA, 1% PP indicator, 0.1 N NaOH and tissue. The equipment used during the research were analytical balance, 60 mesh sieve, baking sheet, cabinet dryer, thermometer, freezer, micropipette, tip, oven, autoclave, incubator, pH meter, laminar airflow, vortex, refractometer, burette and stative, and test sheets for hedonic. Making probiotic drink modified, namely honey queen pineapple juice inoculated with 3% (v/v) pure *Lactobacillus plantarum* starter and 3% (v/v) yogurt powder starter and which previously added 0-4% (v/v) inulin's mangrove apple. The probiotic drink of honey pineapple added with starter and inulin is then fermented at 42°C for 4 hours.

## Methods

### Making Pineapple Probiotic Drinks

Making probiotic drink modified, namely honey queen pineapple juice inoculated with 3% (v/v) pure *Lactobacillus plantarum* starter and 3% (v/v) yogurt powder starter and which previously added 0-4% (v/v) inulin's mangrove apple. The probiotic drink of honey pineapple added with starter and inulin is then fermented at 42°C for 4 hours.

The research was conducted experimentally using a Completely Randomized Design (CRD) with 5 treatments and 4 replications. The treatments were giving different concentrations of inulin to pineapple juice probiotic drinks including T0 (0%), T1 (1%), T2 (2%), T3 (3%) and T4 (4%) and if it was obtained the best concentration. The parameters tested in this research were total lactic acid bacteria, total acid, pH value, total soluble solids and hedonic test.

## Results and Discussion

### Total LAB

The results of the total lactic acid bacteria in pineapple probiotic drinks with the addition of inulin's mangrove apples can be seen in Table 1.

**Table 1.** Total LAB in pineapple fruit probiotic drink with inulin's mangrove apple

Treatment	Total Lactic Acid Bacteria (CFU/ml)
P0	7,65 ± 0,24 <sup>a</sup>
P1	8,21 ± 0,56 <sup>b</sup>
P2	8,12 ± 0,35 <sup>b</sup>
P3	8,58 ± 0,99 <sup>c</sup>
P4	8,60 ± 0,20 <sup>c</sup>

\* Values (Mean ± standard deviation, four replicates). Different letters in the same row mean significantly differences among cultivars at P<0,05.

Based on Table 1, the difference in concentration had a significant effect ( $P < 0.05$ ) on the total lactic acid bacteria of pineapple probiotic drink with the addition of inulin's mangrove apples in treatments P0, P1, P2, P3 and P4. Total LAB test results in treatment P0 are 7.65 log CFU/ml or  $9.5 \times 10^7$  CFU/ml, P1 is 8.21 log CFU/ml or  $1.63 \times 10^8$  CFU/ml, P2 is 8.12 logs CFU/ml or  $1.58 \times 10^8$  CFU/ml, P3 is 8.58 log CFU/ml or  $3.83 \times 10^8$  CFU/ml and P4 is 8.60 log CFU/ml or  $4.33 \times 10^8$  CFU/ml. The total LAB test results showed that the addition of inulin from mangrove apples could increase the number of bacterial colonies more than without inulin. The number of lactic acid bacteria is significant in processing probiotic drinks because it will affect the quality of the product. According to SNI 2981:2009, the number of starter bacteria contained in yogurt must reach a minimum of  $10^7$  colony/gram. This research used a starter with a total number of  $2.9 \times 10^7$  CFU/ml and the total range of lactic acid bacteria produced in the product ranged from 7.65 to 8.60 to  $10^8$  CFU/ml. The number of lactic acid bacteria that grows on the product still accepted SNI requirements, so it can be concluded that the total lactic acid bacteria in pineapple probiotic drinks with the addition of inulin's mangrove apple still accept the requirements to be declared as probiotic drinks. The more lactic acid bacteria contained in probiotic drinks with their proteolytic ability, the more beneficial it is for the digestive organs [16]. The higher the concentration of inulin added to the pineapple probiotic drink, the more the number of lactic acid bacteria that grow. Lactic acid bacteria are thought to utilize inulin for fermentation and produce lactic acid. LAB will ferment inulin to produce lactic acid and short-chain fatty acids (acetic, butyric, propionate). In pineapple probiotic drinks, there is no lactose, so besides inulin, lactic acid bacteria use the sugar in pineapple for the fermentation process [17]. Lactic acid bacteria utilize monosaccharides contained in fruit during fermentation so that lactic acid is formed which causes the atmosphere to become acidic [18]. Lactic acid bacteria can utilize the high sucrose content in pineapple as a carbon source

### pH Value

The results of testing the pH value of pineapple probiotic drinks with the addition of inulin's mangrove apple can be seen in Table 2.

**Table 2.** pH value in pineapple fruit probiotic drink with inulin's mangrove apple

Treatment	pH Value
P0	$4,10 \pm 0,82^a$
P1	$4,05 \pm 0,57^a$
P2	$4,00 \pm 0,82^{ab}$
P3	$3,90 \pm 0,81^b$
P4	$3,70 \pm 0,63^c$

\*Values (Mean  $\pm$  standard deviation, four replicates). Different letters in the same row mean significantly differences among cultivars at  $P < 0,05$ .

Based on Table 2. it can be seen that the difference in the concentration of mangrove apples had a significant effect ( $P < 0.05$ ) on the pH value of the pineapple probiotic drink with the addition of inulin's mangrove apples. The resulting pH value is in accordance with the standard of probiotic drinks. A good yogurt probiotic drink has a pH value between 3.5-4.5 [19]. The decrease in pH is related to the activity of lactic acid bacteria in probiotic drinks. Which states that the increasing number of lactic acid bacteria that use lactose and the more sources of sugar that are metabolized, the more organic acids are produced so that automatically the pH will also be lower. In pineapple probiotic drinks do not use milk so that no lactose is used as a nutrient for bacterial growth but the function of lactose can be replaced by inulin. The higher the concentration of inulin's mangrove apple added, the lower the pH of the product. The decrease in pH was caused by the activity of lactic acid bacteria which utilize inulin for growth, that the greater the amount of inulin as a carbon source used for LAB metabolism, the more lactic acid is produced [20].

### Total Soluble Solids

The results of testing the Total Soluble Solids of pineapple probiotic drinks with the addition of inulin's mangrove apple can be seen in Table 3.

**Table 3.** Total soluble solids in pineapple fruit probiotic drink with inulin's mangrove apple

Treatment	Total Soluble Solids
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P0	10 ± 0,45 <sup>ab</sup>
P1	10,53 ± 0,15 <sup>b</sup>
P2	9,53 ± 0,30 <sup>bc</sup>
P3	9,38 ± 0,42 <sup>c</sup>
P4	9,30 ± 0,35 <sup>c</sup>

\*Values (Mean ± standard deviation, four replicates). Different letters in the same row mean significantly differences among cultivars at P<0,05.

Based on the data in Table 3. shows that the difference in inulin concentration of mangrove apples has a significant effect (P<0.05) on the total sugar of pineapple probiotic drinks. The sugar range of the resulting product is 9.30 – 10.53°brix. The highest total sugar was obtained in pineapple probiotic drink products with the addition of 1% inulin's mangrove apple, which was 10.53 ± 0.45, while the lowest total sugar value was obtained in pineapple probiotic drinks with the addition of 4% inulin's mangrove apple, which was 9.30 ± 0.35. The total amount of sugar in the pineapple probiotic drink with the addition of inulin's mangrove apple decreased over time. The decrease in the amount of sugar is due to the activity of lactic acid bacteria which utilize sugar as a nutrient for metabolism, optimally growing microorganisms require more sugar to be broken down to reproduce so that there is less sugar left in the solution [21].

### Hedonic Test

After analyzing the hedonic test of pineapple probiotic drinks with the addition of inulin's mangrove apple, the data obtained as shown in Table 4.

Table 4. Hedonic results in pineapple fruit probiotic drink with inulin's mangrove apple

Treatment	Color	Flavor	Taste	Overall
P0	4,00 ± 0,7	3,68 ± 0,63 <sup>a</sup>	3,88 ± 0,83 <sup>ac</sup>	3,60 ± 0,76 <sup>ab</sup>
P1	4,12 ± 0,78	3,64 ± 0,95 <sup>ab</sup>	3,64 ± 0,64 <sup>ab</sup>	3,80 ± 0,58 <sup>ab</sup>
P2	4,16 ± 0,62	3,44 ± 0,82 <sup>ab</sup>	3,72 ± 0,79 <sup>ab</sup>	3,80 ± 0,7 <sup>ab</sup>
P3	4,32 ± 0,56	3,28 ± 0,98 <sup>c</sup>	4,32 ± 0,63 <sup>c</sup>	4,20 ± 0,64 <sup>c</sup>
P4	4,20 ± 0,7	3,16 ± 0,85 <sup>c</sup>	2,56 ± 0,96 <sup>d</sup>	2,96 ± 0,94 <sup>d</sup>

\*Values (Mean ± standard deviation, four replicates). Different letters in the same row mean significantly differences among cultivars at P<0,05

The results of the test on panelists' preferences including color, aroma, taste and overall are presented in table 6. Analysis of panelists' preferences for pineapple probiotic drinks showed that the addition of inulin's mangrove apple had no significant effect (P> 0.05) on panelists' preference for product color, but had a significant effect (P<0.05) to increase the preference value on aroma, taste and overall product. The highest value was obtained in pineapple probiotic drink with the addition of 3% inulin's mangrove apple.

### Color

The addition of inulin's mangrove apple had no significant effect (P>0.05) on the panelists' preference for color. The color appearance of all pineapple probiotic drinks with the addition of inulin's mangrove apple has almost the same value because the resulting color is the same, namely yellow. There was no significant difference in the color of the product due to the water-soluble nature of inulin. This is because inulin is soluble in water but cannot be digested in the digestive system but in the large intestine, inulin is fermented by intestinal bacteria [22].

### Flavor

In table 4 it can be seen that the addition of inulin's mangrove apple had a significant effect (P<0.05) on the panelists' preference for product aroma. Based on table 4, variations in inulin concentration significantly affect the aroma of pineapple probiotic drinks. The more the addition of inulin, the less favorable the aroma of the product, this is because the pedada fruit or mangrove apple has a distinctive aroma that panelists do not like, the more pedada fruit used in the manufacture of pedada products, the stronger the distinctive aroma of pedada. Pineapple probiotic drink with the addition of inulin has a rather distinctive aroma, namely sour aroma [23].

## Taste

The addition of inulin's mangrove apple had a significant effect ( $P < 0.05$ ) on the panelists' preference for taste. This is because the panelists do not like drinks with a taste that is too sour. Yogurt with a sweet taste is preferred by consumers and has the highest value on consumers' assessment of yogurt taste. P4 probiotic drink has a fairly low preference for taste. This could be due to the low pH of P4 so the taste is quite sour. The presence of lactic acid causes the sour taste in yogurt drinks as a metabolite product due to the activity of Lactic Acid Bacteria [24].

## Conclusion

Based on the results of the hedonic test, the pineapple probiotic drink product with the addition of inulin's mangrove apple can be accepted by the community. The added concentration of inulin's mangrove apple increases the total lactic acid bacteria, but decreases the pH value and total sugar. The best treatment in this research was the addition of 3% inulin because it was closest to the desired criteria, while in the research of storage time it could be concluded that different storage times affected product characteristics.

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## Author Profile



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ingredients for diversified food products that have functional capabilities in the human body that consumes them. As well as the use of microbes for fermentation-based food products for functional food products.