

## Effect of the Fermentation Time of Purple Sweet Potato (*Ipomea batatas L*) Synbiotic Yoghurt with Probiotic Starter on Product Quality and Antioxidant Activity

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**Abstract:** Purple sweet potato (*Ipomea batatas L*) has the potential as a fermentation medium because it has a high carbohydrate content of around 20.1%. The combination of purple sweet potato extract, skimmed milk, and probiotic starter is expected to be a synbiotic yogurt that can help the digestive system. This study aims to determine the effect of long-term fermentation of purple sweet potato synbiotic yogurt with probiotic starter culture on product quality and antioxidant activity. The observed quality was chemical, microbiological, and organoleptic, with variations in fermentation time of 0, 4, 8, and 12 hours. The study's results showed a long-term effect of the fermentation of purple sweet potato synbiotic yogurt with probiotic starter culture on the quality of yogurt. The variation of 0-hour fermentation time did not meet the pH chemical quality standard with a value of 5.16. Still, it met the TAT chemical quality standard with a value of 0.513%, while the variation of fermentation time of 4-12 hours met the pH quality standard with a value of 3.88-4.33 and a TAT of 1.268-1.967%. In the quality of microbiology, there was a total increase in BAL, which was  $46.2 \times 10^7$  -  $90.61 \times 10^7$ . The best organoleptic quality of color, taste, and aroma was obtained in the 8-hour fermentation variation, while the best texture organoleptic quality was obtained in the 12-hour fermentation variation. The best antioxidant activity was received at 12 hours of fermentation with an IC50 value of 62.727 ppm. From this study, it can be concluded that the best yogurt results are obtained with a fermentation time of 12 hours.

**Keywords:** Antioxidant Activity; Probiotic Starter; Purple Sweet Potato; Quality; Synbiotic Yogurt.

### Introduction

Amid busy life and the pressures of modern times, people's awareness of the importance of health is declining. Many rely on practical, fast food but must consider its adverse health effects. In addition, extreme diet trends are also becoming more prevalent, where people focus more on weight loss than on the health and nutritional needs of the body. In declining awareness of health, functional food is becoming increasingly important. Functional foods offer solutions to meet dietary needs while still paying attention to the health of the body. Functional food can be interpreted as food that naturally has one or more compounds, where these compounds have specific physiological functions that are useful for the health of the human body [1]. Indonesia has a variety of natural food sources that can be used as functional food, but its use as functional food is still not optimal. One of the food ingredients that has the potential to be developed for use is purple sweet potatoes. Indonesia can produce 2,029,353 tons of sweet potatoes. The consumption rate of sweet potatoes in 2015 (3,389 kg/capita/year), 2016 (3,598 kg/capita/year), and 2017 (3,666 kg/capita/year) [2]. The use of purple sweet potatoes is still limited; most people use them by consuming them directly or processed into chips [3].

Purple sweet potato (*Ipomea batatas L*) is a food source of inulin known to have benefits as a prebiotic. One of the bioactive compounds that acts as a prebiotic is inulin, a dietary fiber [4]. Prebiotics can be defined as food ingredients that cannot be digested by the body but can be utilized because they can stimulate the growth/activity of

microbes in the intestines [5]. The inulin content in purple sweet potatoes is known to be 7.47% [6]

Inulin is a polymer containing a fructose group with a  $\beta$ -2-1 fructofuranside bond, where this form of fiber is soluble in water and cannot be digested by digestive enzymes [7]. Human digestive enzymes cannot digest inulin, but it can be metabolized by intestinal microorganisms into short-chain fatty acids (ALRP) [8]. ALRP is a compound that has many health benefits, so the food processing process that produces ALRP has the potential to be developed as a functional food. ALRP is known to maintain the body's homeostasis, modulate metabolic processes and the immune system, and provide direct protection against pathogens [9]. In addition, ALRP can preserve the health of epithelial cells in the colon, can also act as an anti-tumour and anti-inflammatory, control glucose homeo-statics, regulate appetite, and maintain the circulatory system [10]. Inulin itself is part of polysaccharides called fructans [11]. In the process of forming ALRP, inulin will be hydrolyzed into a simple form by the activity of the inulinase enzyme [12]. In addition to inulin, purple sweet potatoes are known to have a high anthocyanin content.

The anthocyanins in purple sweet potatoes can increase the antioxidant activity of the product. The antioxidant content in purple sweet potatoes comes from the purple color pigment that purple sweet potatoes possess [13]. The pigment is anthocyanin, which contains a high content of  $\beta$ -carotene, so it has high antioxidant activity. The antioxidant activity value of processed purple sweet potato products is usually in the range of 7.54% - 41.65%, and the antioxidant activity value is due to the presence of *an asylated caffeoyl*

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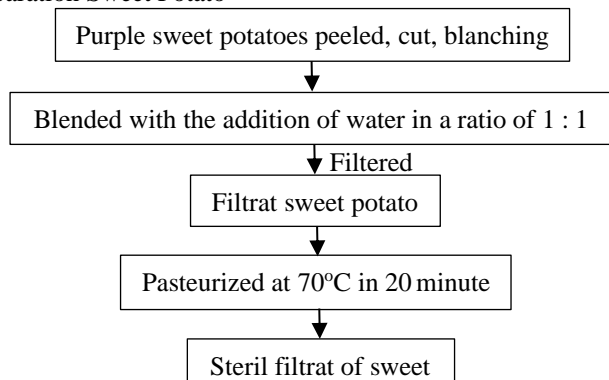
group in anthocyanins [14]. A longer fermentation process can allow time for probiotic bacteria such as *Lactobacillus* and *Streptococcus* to produce lactic acid and other bioactive compounds [15]. Lactic acid has antioxidant properties that can help protect the body's cells from oxidative damage. The duration of fermentation can also affect the activity of enzymes in purple sweet potatoes [16]. These enzymes can help break down complex polyphenols into simpler forms that are easily absorbed by the body. For example, anthocyanins, the pigment that gives sweet potatoes their purple color, can be broken down into more biologically active compounds during a long fermentation process. Time of fermentation can reduce antioxidant activity because some antioxidant compounds may be degraded due to improper fermentation conditions or excessive oxygen exposure [17].

Based on the background, it is known that purple sweet potatoes can be added to manufacture yogurt. Adding purple sweet potatoes with a specified percentage as an additional source of nutrients for starter bacteria will affect the growth of bacteria in yogurt. This research needs to be carried out to determine the best fermentation time by adding purple sweet potatoes to yogurt by analyzing the pH value, total Lactic Acid Bacteria (BAL), Total Titrated Acid (TAT), antioxidant and organoleptic activity. The results of this research are expected to be used as an alternative to functional food development.

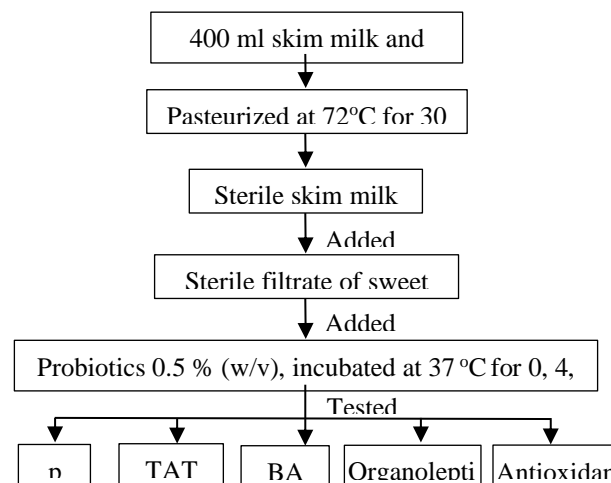
### Research Methods

This type of research is a true experimental research. True experimental research is research in which the subject is given a treatment and then measured as a result of the treatment on the subject. True experimental research aims to determine whether the treatment given to the studied topic is influenced. In other words, this research aims to determine whether the long fermentation of purple sweet potato synbiotic yogurt affects the quality of synbiotic yogurt. The pH value decreased until 12 hours of fermentation, followed by increased TAT. A decrease in IC50 value characterizes increased antioxidant activity. The resulting product has the highest preference for color, aroma, and taste at a fermentation time of 8 hours, while the highest level of choice for texture is 12 hours.

#### Preparation Sweet Potato



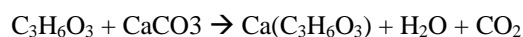
#### Making Purple Sweet Potato



### Results and Discussion

In this research, yogurt is made from purple sweet potato and skim milk with commercial propitic starter culture and fermented for a certain period. Purple sweet potatoes are washed, peeled, and cut into small pieces, then blanched for ± 15 minutes with a ratio of water and purple sweet potatoes 1:1. The purpose of the blanching process is to reduce the damage of functional compounds such as anthocyanins and phenolic compounds that play an important role in antioxidant activity [18].

The results showed a relationship between total BAL, pH, and TAT. The longer the fermentation time, the more BAL grows, the lower the pH, and the more total lactic acid is formed. This indicates that a yogurt probiotic starter consisting of 10 bacteria can utilize purple sweet potato synbiotic yogurt for its growth and development. The formation of a clear zone around the colony that grows on the growth medium results from the reaction between lactic acid produced by BAL and CaCO<sub>3</sub> during the incubation period so that Ca-lactate is formed. The reaction occurred as follows:



The results of TAT, pH, BAL, and antioxidant activity analysis can be seen in Table 1.

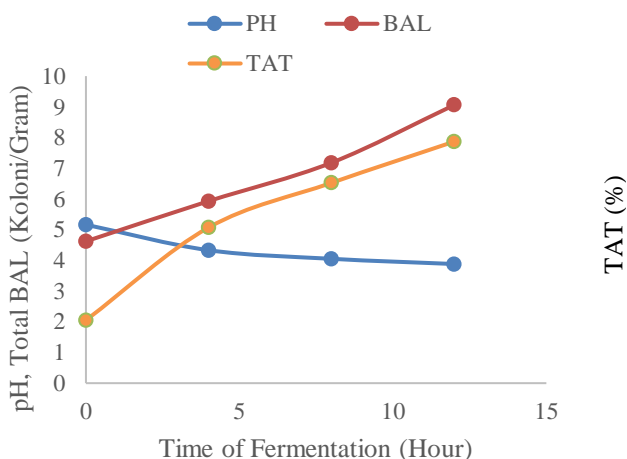
**Table 1.** Table results of TAT, PH, BAL, and antioxidant activity analysis.

Time Fermentation (hours)	TAT (%)	pH	BAL (colonies/gram)	Antioxidant (IC <sub>50</sub> )
0	0.513	5.16	46.2×10 <sup>7</sup>	114.713
4	1.268	4.33	59.3×10 <sup>7</sup>	104.279
8	1.631	4.05	71.8×10 <sup>7</sup>	86.636
12	1.967	3.88	90.6×10 <sup>7</sup>	62.727

In this research, the fermentation medium used is inulin, a prebiotic. The inulin level in fresh purple sweet potatoes was 6.073% [11]. Purple sweet potato inulin degraded into glucose and fructose and fermented will be

further metabolized to produce ALRP. In addition, adding 5% sugar (w/v) to purple sweet potato synbiotic yogurt not only gives flavor to the product but can also affect the growth of BAL. Bacteria can utilize sugars to become an additional source of nutrients, which will then be hydrolyzed into monosaccharides to produce BAL growth [8].

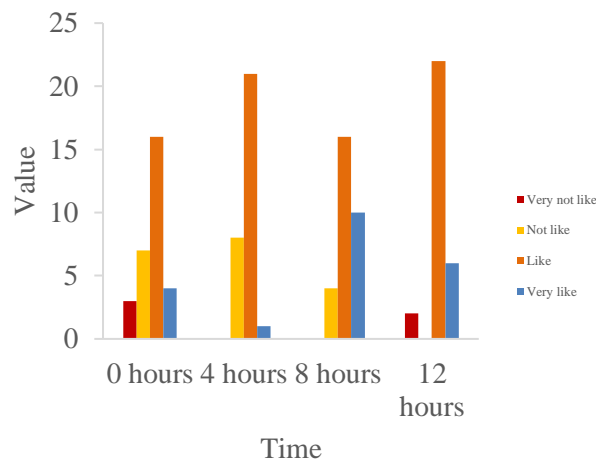
Increased BAL growth can lead to pH depletion and an increase in total titrated acid. Purple sweet potato synbiotic yogurt has an initial pH of 5.16 at 0 hours, then decreases to 3.88 at 12 fermentation. The TAT value in fermentation from 0 to 12 hours increased by 0.513% to 1.967%. The decrease in pH in yogurt samples is due to the activity of lactic acid bacteria that convert lactose in milk into lactate [19]. The fermentation process by BAL is characterized by an increase in the amount of BAL that grows in yogurt, which causes the accumulation of organic acids that cause a decrease in pH. The amount of lactic acid produced during fermentation will undergo the dissociation of lactic acid into H<sup>+</sup> ions and CH<sub>3</sub>CHOHCOO<sup>-</sup> ions. The increasing number of H<sup>+</sup> ions can affect the decrease in pH and increase the sample's acidity [20]. The reduction in pH causes the taste to become acidic due to the formation of lactic acid as the main product of lactic acid bacteria metabolism [21]. The more sources of nutrients obtained are metabolized, the more organic acids are produced, especially lactic acid, which causes a decrease in the pH value of fermented milk [22]. Figure 1 proves a relationship between BAL growth and pH decrease and TAT value increase.



**Figure 1.** Graph of BAL Growth, pH decrease, and TAT increase relative to fermentation duration.

The results of this study are also supported by previous research [24], which states that the fermentation time in purple sweet potatoes affects the growth of BAL and the chemical quality (pH and TAT) of fermented drinks. Fermented beverages for 12 hours have the best quality with a total amount of BAL 1.86×10<sup>9</sup> CFU/mL. Another study [25] showed that the fermentation time of purple sweet potato yogurt affected microbiological and chemical quality. The best microbiological quality occurred at a fermentation period of 12 hours, with a total value of lactic acid bacteria reaching 1.86 × 10<sup>9</sup> CFU/mL. The best chemical quality occurs at a fermentation period of 12-48 hours with a total titrated acid value of 0.240-0.630 and a pH of 3.9-3.4. Supporting data was obtained from this data, showing that the fermentation duration affected the total BAL, pH value, and TAT percent. In this study, purple sweet potatoes were processed into synbiotic yogurt, where inulin is classified as

a type of soluble fiber that dissolves in water, making it easier for BAL to use inulin for its growth. The BAL in the fermentation of gembili flour (5%) was smaller than that used in this study (8%).



**Figure 2.** Color organoleptic test results.

In this research, the organoleptic quality of purple sweet potato synthetic yogurt tested was color and taste. Color is the first sensory attribute in choosing a product, affecting consumer preferences. The colors should be attractive, fun, and uniform and can represent the added flavors. The organoleptic results showed that in the color aspect test, the most value was obtained by the variation of 8-hour fermentation, with a value of 16 likes and ten very likes. The level of color preference obtained varies depending on the color seen in the fermentation results of purple sweet potato synbiotic yogurt. The color of fresh purple sweet potato juice is dark purple. In contrast, purple sweet potato synbiotic yogurt is light purple, so there is a significant difference between the color of sweet potato juice and purple sweet potato synbiotic yogurt. The color produced results from anthocyanin compounds in purple sweet potatoes [23].

The discoloration of yogurt occurs because anthocyanins can form complex equilibrium in a water solution. The flavillium cation is a dominant species at low pH (<2) with a red color and is easily deprotonated to form a quinoidal base with a blue color or colorless carbinol. The pH of purple sweet potato synbiotic yogurt showed a value of 5.16 at the fermentation time of 0 hours. It is suspected that the dominant anthocyanin structure is quinonyroidal andiroba, so the color seen is purple, a mixture of red (cation flavylium) and blue (Quinodal). During the fermentation time of 4 – 12 hours, the pH of fermented beverages was in the range of 3.88-4.33, with the suspected dominant anthocyanin structure in the form of carbinols. The light purple color that appears in this synbiotic yogurt is a combination of colors from red (cation flavylium), blue (Quinodal), and colorless (Carbinol). The structural changes that occur can be observed in Figure 3.

Synbiotic yogurt is the most preferred flavor level and is fermented for 8 hours. Different levels of liking are influenced by the taste of the compounds contained in each yogurt. At 0-hour fermentation, the panelists' preference level was the lowest, and this was suspected because the taste of purple sweet potato synbiotic yogurt was too sweet. The sugar produces this sweetness in the purple sweet potato

juice and the addition of sucrose. Then, at the 12-hour fermentation time, the synbiotic yogurt flavor becomes sweet with a slightly sour mixture; the panelists prefer this variation. The panelists assessed the sweetness and deliciousness of the synbiotic yogurt product to be balanced at this variation in fermentation time. The resulting taste is the result of metabolites from probiotics derived from commercial probiotic starter cultures. The possible metabolites formed are lactic acid, acetic acid, propionate acid, butyric acid, glucose, and fructose. In the 12-hour fermentation variation, the level of liking decreased, with seven panelists choosing not to like it; this is suspected to be due to the sour taste that is too dominant.

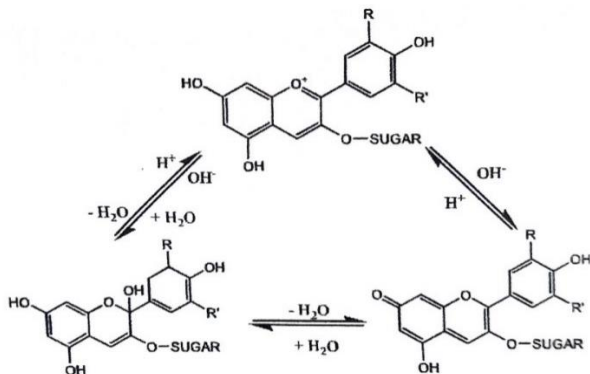


Figure 3. Flavylium cation complex equilibrium at pH 1-8.

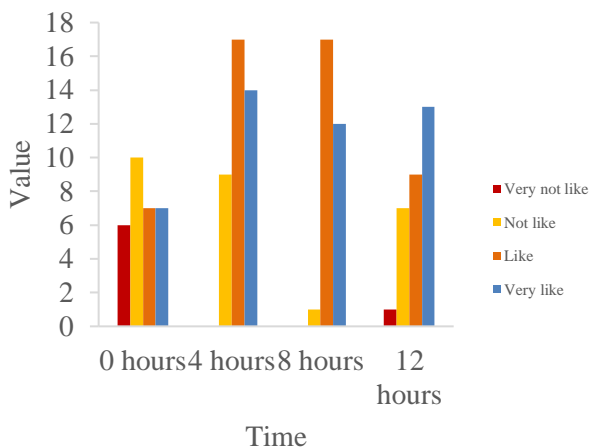


Figure 4. Taste organoleptic test results.

Antioxidant activity was measured using the DPPH method and measured using a UV-Vis spectrophotometer at a maximum wavelength ( $\lambda$ ) of 516.3 nm. DPPH gives a deep purple color to the maximum absorption of that wavelength. When antioxidants react with DPPH, the antioxidants will donate hydrogen atoms to DPPH, which decreases the amount of DPPH. A reduction in the amount of DPPH causes the adsorption value of DPPH to decrease, changing the color of dark purple to faded purple. This change in warning occurs due to a decrease in the conjugate double bond in DPPH, where DPPH radicals get hydrogen atoms from the sample. As a result of this reaction, DPPH radicals will experience a reduction to DPPH-H, as shown in Figure 5.

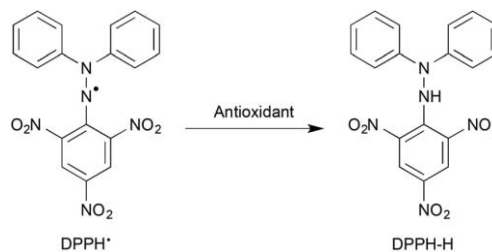


Figure 5. Reaction of antioxidant hydrogen atom donor to DPPH

The antioxidant potential of purple sweet potato synbiotic yogurt has reached a substantial  $IC_{50}$  value of 62.727 ppm at a fermentation period of 12 hours. The antioxidant potential of purple sweet potato synbiotic yogurt has reached a strong  $IC_{50}$  value of 62.727 ppm at a fermentation period of 12 hours. This result is supported by previous research [23], which reported an increase in antioxidant activity in purple sweet potato yogurt by two times higher than purple sweet potato yogurt without fermentation [26]. It also explained that yogurt with as much as 8% purple sweet potato flour had 6x higher antioxidant activity than without purple sweet potato flour.

### Conclusion

Based on research and discussion related to the effect of fermentation time of purple sweet potato synbiotic yogurt with probiotic starter culture on product quality and antioxidant activity that researchers have developed, it can be concluded that fermentation time affects increasing BAL, TAT, and Antioxidant Activity, as well as affecting a decrease in pH value. The total BAL continues to increase up to a fermentation time of 12 hours. The best yogurt is made with a fermentation time of 12 hours. Further research is needed regarding phenolic levels in yogurt.

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