The microbiological characteristics of modified tapioca by liquid fermentation using ginger and curcuma extract

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Abstract. This research was conducted to determine the microbiological characteristics of modified tapioca by liquid fermentation using ginger and curcuma extract. The study was using randomized block design with two factors i.e.: type of extract (P): (water, curcuma extract, giant ginger extract, red ginger extract and small ginger extract) and ratio of starch to extract volume (U): (1 kg : 1 L, 1 kg : 2 L and 1 kg : 3 L. The interaction of type of extract with ratio of starch to extract volume had highly significant effect on lactic acid bacteria (LAB) on fermentation liquid. The type of extract and ratio of starch to extract volume had a highly significant effect on total plate count (TPC) on fermentation liquid. Based on the total of lactic acid bacteria, P1U3 (using water with a volume of 3 L of extract) was the treatment with the highest number of LAB, namely $1.79 \times 10^7$ CFU/mL. The treatment of extract types gave the result that P1 (water) had the highest TPC value, and the treatment of the ratio of starch to the volume of extract gave the result that U3 (1 kg : 3 L) had the highest TPC value.

1. Introduction

Indonesia is a country that has a high potential for cassava production. According to the Central Statistic Agency (BPS) of Indonesian cassava production in 2018 reached 19,341,233 tons [1]. Besides, the price of cassava is also cheap. This causes cassava to have good prospects in processing it into starch. In general, natural starch still has some weaknesses that can hinder the application process. One of the objectives of the modification process is to improve the characteristics of the starch so that it can facilitate the application [2].

One of the starch modifications processes that is often carried out in Indonesia is through the fermentation process. The fermentation process that is usually carried out by the community is fermentation using ordinary water. In this study, the fermentation process was carried out by soaking tapioca using various types of extract (giant ginger extract, red ginger extract, small ginger extract and curcuma extract).

Currently, the use of cassava is more to a food source, so it is necessary to pay attention to other parts of cassava whose use has not been optimally utilized [3]. From the production of so much acidic tapioca, a fermented liquid is produced from tapioca which has other components as well as microorganisms that may grow such as lactic acid bacteria.

Lactic acid bacteria is one of the bacteria that play a role in the fermentation process and is useful for improving the quality and safety of foodstuff through natural inhibition of pathogenic microbes. Lactic acid bacteria produce several antimicrobial components, namely organic acids, carbon dioxide,
hydrogen peroxide, diacetyl, reuterin, and bacterioisin [4]. This study aimed to determine the microbiological characteristics contained in the modified tapioca fermented liquid using types of extract (giant ginger extract, red ginger extract, small ginger extract, and curcuma extract).

2. Materials and methods

The materials that used in this research were cassava obtained from Kreasi Lutvi cassava chips, Medan Tuntungan and giant ginger, red ginger and curcuma obtained from Sipintu Angin Village, Dolok Pardamean District, Simalungun Regency and small ginger obtained from Sidikalang, Medan. The reagents used in this study were distilled water, MRS Agar, PCA, and 0.9% NaCl.

2.1. Research methods

The schematic of research design can be seen in Figure 1.

2.1.1. The process of making fermentation liquid. Cassava is peeled, then washed under running water to clean the remaining dirt. After that, as many as 15 kg of washed cassava are cut into pieces, then the pieces of cassava are crushed using a shredder to produce tuber pulp [5]. The resulting tuber slurry is then added with water in a ratio of 1:2. Then filtered then pulp of the tubers using a filter cloth so that...
the pulp and liquid are separated. Then let the liquid stand for 1 hour so that the starch sediment is separated from the water. Next, the resulting wet starch was weighed 1 kg each and put into each plastic container that had been prepared. Furthermore, 1 L, 2 L, and 3 L of juice were added to each treatment as a fermentation solution. The plastic container is covered with a filter cloth and tied so that dirt such as dust cannot enter so that the fermentation process is not contaminated. Furthermore, the starch is fermented for up to 16 days. After 16 days the starch precipitate was separated from the fermentation solution. The fermented liquid was analysed for the BAL and TPC.

2.1.2. Lactic acid bacteria [6]. Weighed the sample as much as 1 mL then added with 9 mL of 0.9% NaCl and diluted $10^{-1} - 10^{-6}$. Two series of $10^{-5} - 10^{-6}$ dilutions were peptized as much as 1 mL and the growth was carried out using the pour method on deMann Rogosa Sharp Agar (MRSA) medium, then incubated at 37 °C for 48 hours. BAL colonies were calculated using the Bacteriological Analytical Manual (BAM) standard at the amount of 25-250 CFU/mL.

2.1.3. Total plate count [7]. A sample of 1 mL is diluted with 9 mL of 0.9% NaCl which is called a 10-1 dilution. The mixture is homogenized and 1 mL of the solution is taken using a micropipette then diluted with 9 mL of 0.9% NaCl which is then referred to as the $10^{-2}$ dilution and so on until dilution $10^{-6}$ is piped aseptically 1 mL of sample suspension by using a micropipette, then put into a sterile petri dish done in duplo. Next, add 15-20 ml of PCA (plate count agar) medium. Shake like number eight so that the sample solution and medium are mixed homogeneously. Waited for the medium to freeze and then incubated it upside down in an incubator at 36 °C for 48 hours. All procedures were performed aseptically and the equipment and medium were sterilized.

2.1.4. Data analysis. This research was using a factorial randomized block design (RBD) consisting of two factors, namely factor 1 is the type of extract (P) and factor 2 is the ratio of starch to extract volume (U) which is made as much 3 replicates. The levels for each factor 1 and 2 can be seen in Figure 1.

3. Results and discussion

3.1. Lactic acid bacteria on liquid fermentation

![Image of LAB analysis](image-url)

**Note:** $P_1$ (Water), $P_2$ (Curcuma extract), $P_3$ (Giant ginger extract), $P_4$ (Red ginger extract), $P_5$ (Small ginger extract) and $U_1 = 1$ kg : 1 L, $U_2 = 1$ kg : 2 L, $U_3 = 1$ kg : 3 L

**Figure 2.** Interaction type of extract and the ratio of starch and extract volume with LAB of fermented liquid.
The interaction between the type of extract and the ratio of starch to the volume of the extract had a very significant effect (P<0.01) on the total lactic acid bacteria fermentation liquid. The interaction relationship between the type of extract and the ratio of starch and extract volume with LAB of fermented liquid can be seen in Figure 2.

In Figure 2, it can be seen that there is a decrease in the total lactic acid bacteria in the fermentation liquid along with the increasing volume of extract used. This is because the growth of lactic acid bacteria is inhibited by the increasing volume of the extract used. After all, the more volume of the extract used, the antimicrobial components will also increase so that the growth of lactic acid bacteria is disrupted. However, the P₁ fermentation solution increased the number of lactic acid bacteria as the volume of extract used increased. This is because in the P₁ treatment the fermentation liquid used is in the form of plain water which does not contain antimicrobial compounds in it so that the more water volume is used, the number of lactic acid bacteria also increases.

3.2. Total plate count on fermentation liquid

3.2.1. The effect of the type of extract on the total plate count fermentation liquid. The type of extract had a very significant effect (P<0.01) on the total plate count (TPC) contained in the resulting fermentation liquid and can be seen in Figure 3.

In Figure 3, it can be seen that the highest total plate count was found in treatment P₁, namely 1.60 x 10⁷ CFU/mL, and the lowest total plate count in treatment P₅ namely 1.38 x 10⁷ CFU/mL. The high total plate count in treatment P₁ compared to other treatments was due to the fermentation liquid used in the form of water, while other treatments used rhizomes such as ginger and curcuma. Curcuma and ginger have antimicrobial compounds that can inhibit the growth of microorganisms during fermentation [8].

3.2.2. Ratio of starch to extract volume on the total plate count fermentation liquid. The ratio of starch and extract volume has a very significant effect (P<0.01) on the total plate count contained in the resulting fermentation liquid which can be seen in Figure 4.

Figure 4 shows that there is an increase in the value of the total plate count along with the increasing volume of extract used. This is because during fermentation there is growth of microorganisms. Microorganisms that growth during the fermentation process can be bacteria, yeast and so on. So that with the increasing volume of extract used, the growth media for microorganisms will also increase the total number of microbes in the fermentation liquid.
4. Conclusions
The interaction between types of extract with the ratio of starch and volume of extract had a very significant effect on the total lactic acid bacteria and resulted in P₁U₃ (soaking using water with a volume of 3 L of extract) was the treatment with the highest number of lactic acid bacteria. The type of extract gives the result that P₁ (soaking using water) has the highest total plate count value, namely $1.6 \times 10^7$ CFU/mL. The ratio of starch to the volume of extract gives the result that U₃ (1 kg : 3 L) has the highest total plate count value, namely $1.63 \times 10^7$ CFU/mL.

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