The Effect of Differences in Sugar Concentration on The Amount Of Sauerkaut Lactic Acid Bacteria from Cabbage (*Brassisca oleracea L.*)

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Abstract. Lactic acid bacteria are probiotic bacteria that have an important role in preventing and curing various diseases. Affects the content of lactic acid bacteria produced, this is because sugar is a carbon source that is used as an energy source for lactic acid bacteria. This study was conducted from April to August 2021 at the Padang State University Research Laboratory of the Biology Faculty of Mathematics and Science. 5 treatments, 3 replications, and a completely randomized design (CRD) were employed in this investigation. The treatments in this study were K (cabbage 100 g + salt 2.5%), A (cabbage 100 g + salt 2.5% + sugar 5%), B (cabbage 100 g + salt 2.5% + sugar 10%), C (cabbage 100 g + salt 2.5% + sugar 15%), D (cabbage 100 g + salt 2.5% + sugar 20%). With a significance level of 0.05, data on the impact of additional sugar on the number of lactic acid bacteria in cabbage were analyzed using ANOVA (Analysis of Variance) and a significant difference test using Duncan's Multiple Range Test (DMRT). According to the study's findings, sugar can have an impact on how many lactic acid bacteria are formed by sauerkraut made from cabbage. From the results of data analysis, F count < F table so that the DMRT test was conducted. The higher the concentration of sugar used, the greater the number of bacteria produced. In treatment A the number of lactic acid bacteria was 29.21667x 105 cfu/g while in treatment D the number of lactic acid bacteria produced was 224,83333 105 cfu/g.

Keywords: Isolation · Lactic Acid Bacteria · Probiotics · Fermentation · Sugar

1 Introduction

A type of bacteria called lactic acid bacteria (LAB) is responsible for the fermentation process' major byproduct, lactic acid [1]. Lactic acid bacteria are classified as gram-positive bacteria, do not form spores, form coccus or bacilli, and are generally catalase negative. LAB is a facultative anaerobic bacterium capable of living in a wide range of habitats in nature, including the digestive tract of animals and humans, canned food, dairy products, fermented products, fruits, and vegetables.

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LAB has a role to prevent and cure various diseases so this bacterium is classified as one of the probiotics [2]. Any type of microbial cell preparation or microbial cell component that has a positive impact on the host's health is referred to as a probiotic. Probiotics are often a group of non-pathogenic bacteria that, when routinely taken in appropriate amounts, have a favorable impact on the physiology and health of the host's digestive tract. If there are enough probiotic bacteria present, the digestive tract's health will therefore improve. These probiotic bacteria help control pathogenic microorganisms in the digestive tract.

Probiotic bacteria have a positive effect on health such as lowering blood cholesterol, increasing intestinal motility and detoxification, inducing the immune system, producing various metabolites, and metabolism of vitamins, minerals, and hormones. So eating foods with sufficient LAB content can have a positive effect on body health.

LAB can be isolated from various processed fermented foods, one of which is sauerkraut. Sauerkraut (salted cabbage) is a biotechnology processed product that is the result of lactic acid fermentation from chopped cabbage (*Brassisca oleracea* L.) which is finely sliced and added with 2.5% salt [3]. by lactic acid bacteria for successful fermentation. Gradually the fermented microflora was dominated by *L. mesentereides, L. plantarum, L. curvatus, L. brevis* and Pediococcus naturally without the addition of microorganism cultures.

In making sauerkraut sugar can be added to improve the quality of the sauerkraut. The fermentation process is impacted by the addition of sugar; the faster the fermentation, the higher the sugar concentration given. This is because high sugar concentrations can increase microbial growth. Sugar is a nutrient used for microbial growth so the higher the sugar concentration allows the growth of more bacteria [4].

The results of Putri's research, et al. 2020, show that the concentration of sugar is directly proportional to the time of fermentation, namely the higher the concentration of sugar, the faster the fermentation due to increased microbial growth [5]. Sugar plays a role as a nutrient that is used for microbial growth so that microbes can grow more [6]. This study's objective was to ascertain how varying sugar concentrations affected the quantity of lactic acid bacteria present in sauerkraut made from cabbage (*Brassica* oleracea L.).

2 Materials and Methods

2.1 Sauerkraut Sampling

Choose whole cabbage that is fresh and not rotten. Next, remove the outside, rotten part, and liver from the cabbage. Then the cabbage is sliced thinly about 0.5 cm and then washed with water until clean. After that, each cabbage was weighed as much as 100 g, added 2.5% salt, and given sugar according to the treatment, namely 5%. 10%, 15%, 20%. The results of mixing each concentration are put into the fermentation jar while pressing it slowly until the water in the cabbage comes out covering the surface of the cabbage slices and close the jar tightly. Fermentation will last for 4 days at room temperature [7].

2.2 Isolation of Lactic Acid Bacteria

Using the pour plate method with a selective medium for lactic acid bacteria, such as MRSA media, lactic acid bacteria were isolated. To create a sample suspension with a dilution of 10^{-1}, a total of 1 ml of the liquid sample from the sauerkraut was obtained and placed into a test tube containing a dilution of 9 ml of sterile, 0.9 percent physiological NaCl. Additionally, 1 cc was taken from the 10–1 dilution and transferred to the second test tube to create the 10^{-2} dilution, and so on until the 10–8 dilution was created. Furthermore, as much as 1 ml of suspension from a dilution of 10^{-3} to 10^{-5} is pipetted into a sterile petri dish that has been provided, and pour about 15 ml of MRSA medium then the bacteria are spread in the media by rotating it to form a figure of eight. After the agar solidifies, close the petri dish with wrapping and incubate in an inverted position for 1 x 24 h.

2.3 Calculation of Total BAL

The method used to calculate the number of LAB is the cup count method (Total Plate Count). The number of colonies used to calculate the total LAB is with a colony scale of 25–250 expressed in cfu/ml using the formula: Number of bacteria = number of colonies x 1/dilution factor [8].

2.4 Data Analysis

The ANOVA test (Analysis of Variance) was used to analyze the data from the calculation of the number of lactic acid bacteria. If there was a significant difference between treatments, the Duncan's Multiple Range Test (DMRT) method was used to further analyze the difference at a significant level of 0.05.

3 Results and Observation

To determine the effect of sugar concentration on the number of lactic acid bacteria in sauerkraut, lactic acid bacteria were isolated using MRSA media. Lactic acid bacteria can develop more readily in MRSA. With its overall selective nature, colonies of lactic acid bacteria from several strains can grow on this medium.

Based on the results of the study, it was found that the highest number of lactic acid bacteria was found at 20% sugar concentration, which was 225 x 105 cfu/mL and the least was in the control treatment without added sugar, which was 29.55 cfu/mL. Table 1 data shows that the difference in sugar concentration affects the number of lactic acid bacteria produced.

A greater quantity of sugar leads to a greater production of lactic acid bacteria. The results of the ANOVA test showed that the calculated F F table at the 5% level of significance. So that H_0 is rejected and H_1 accepted, so it can be concluded that the treatment of different glucose concentrations affect the amount of lactic acid bacteria produced sauerkraut. Sugar is one of the factors that affect the growth of lactic acid bacteria. This is because sugar is a nutrient and carbon source used by microbes for

Table 1. ANOVA Test Results Effect of Sugar Concentration on the Number of Lactic Acid Bacteria in Sauerkraut

Source Diversity	DB	Amount Square	Square middle	F Count	F Table
Treatment	4	76,974,996	19,243,749	338,019	3.48
Error	10	569.31	56,931		
Total	14	77,544,306			

Table 2. DMRT test results The Effect of Sugar Concentration on the Number of Lactic Acid Bacteria in Sauerkraut

Treatment	Average
Control	29.55 [a]
A (5%)	42,617 [ab]
B (10%)	50.21 [b]
C (15%)	88,627 [c]
D (20%)	225 [d]

metabolism and cell growth. With the availability of optimal nutrition, the number of bacteria produced is also increasing [9].

Probiotic microorganisms with a beneficial impact on health include lactic acid bacteria. The capacity of lactic acid bacteria to convert carbohydrates into lactic acid is its most crucial characteristic. The growth of unwelcome microbes is inhibited by lactic acid [10].

From Table 2, it can be seen that treatment K was not significantly different from treatment A, but there was a significant difference between treatments K and B, C and D. The results of the calculation of the number of lactic acid bacteria with higher sugar concentrations produced more lactic acid bacteria, which can be seen in Fig. 1.

During fermentation, lactic acid bacteria use sugar for development and create metabolites in the form of lactic acid. Microbes will remodel carbon compounds (sugars) into energy for growth and lactic acid as metabolites. This affects the increase in the number of bacterial cells, the more the amount of sugar present, the more the number of bacteria.

In the process of fermenting cabbage sauerkraut with the addition of sugar, in addition to adding flavor variations, it also affects the number of bacteria produced. The addition of sugar can increase the number of lactic acid bacteria. It can be seen that the administration of sugar with a concentration of 20% is able to produce more lactic acid bacteria.

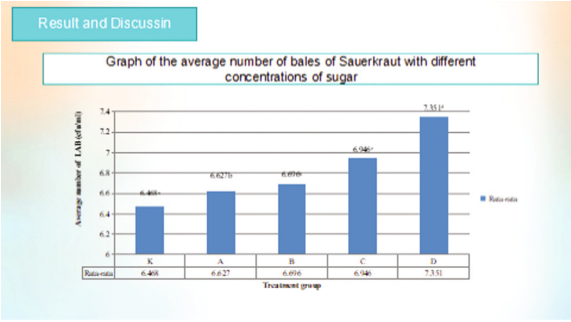

Fig. 1. Average Total Lactic Acid Bacteria

4 Conclusion

The quantity of lactic acid bacteria produced by sauerkraut is influenced by the number of lactic acid bacteria present in cabbage when various sugar concentrations are added. In the control treatment the amount of lactic acid bacteria produced 29.55 x 10^5 cfu/ml, 5% sugar concentration 42.617 cfu/ml, 10% sugar concentration 50.21 cfu/ml, 15% sugar concentration 88.627cfu/ml, sugar concentration 20% 225 cfu/ml. This proves that the higher the concentration of added sugar, the more the number of lactic acid bacteria produced.

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