Improving probiotic quality of cabbage waste with vitamin and minerals addition seen from the contents of potential hydrogen (ph), physical quality of organoleptic and lactic acid bacteria count

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Improving probiotic quality of cabbage waste with vitamin and minerals addition seen from the contents of potential hydrogen (ph), physical quality of organoleptic and lactic acid bacteria count

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Abstract. The aim of the study was to evaluate the quality of probiotics derived from fermented cabbage waste which was added by vitamins and minerals in terms of pH, organoleptic value and lactic acid bacteria count. The study was arranged based on a completely randomized design with five treatments and three replicates. The cabbage was chopped, added with 8% molasses and 6,7% salt. The mixture was fermented for 6 days in anaerobic conditions. After six days of fermentation, the substrate was added with vitamins and minerals for about 0%, 2,5%, 5%, 7,5% and 10%, and then was fermented for 2 days in anaerobic conditions. The results showed that the addition of vitamins and minerals to substrate had a effect (p<0,05) on pH, lactic acid bacteria count, and color with the best treatment was T4 (10%). The conclusion of this study was that adding vitamins and minerals to fermented cabbage waste improved the quality of probiotics from fermented cabbage waste juice.

1. Introduction

Feeds is the most important thing to make successful livestock business because it's needed to reach high animal productivity. Good animal production affected by addition of feed additives. The common feed additive is Antibiotic Growth Promotor (AGP). The Principle of antibiotics as feed additive is decrease the population of bacteria in digestive tract. Now days the use of antibiotics as additional needs already restricted because of the negative effect in meat product like antibiotics residue. The replacements of antibiotics are probiotics, prebiotics, and synbiotics (composite of probiotics and prebiotics). The effort to find replacement of AGP currently focused on natural ingerdients, such as microba and the metabolite products like organic acids. The applications of organic acids are expected to decrease and eliminate negative effect without decreasing the quantity and quality of livestock products. Natural ingredients can be used to produce synbiotic is fermented cabbage waste [1].

Fermented cabbage waste contains good high amounts of lactid acid bacteria (LAB) for animal's disgestive tract. The Fermentation of Lactic acid was created by *Streptococcus, Leuconostoc, Lactobacilllus* or *pediococcus* type. Lactid acid bacteria that reacts in vegetable fermentation are *Lactobacillus plantarum, Lactobacillus brevis, Rhizopus oryze* and *Saccharomyces cerevise* [1, 2, 3]. The production of lactic acid by lactic acid bacteria (LAB) run fastly so the growth of the other unexpected microorganism stunned. The advantage of fermented vegetable waste such as organic acid that can be useful as biological preservation and also as

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starter for feed fermentation. Use as a starter for fermentation of feed ingredients can increase mineral content by 20% and durable feed ingredients are stored for 1 year at room temperature without changing the proximate composition [4].

Probiotics are beneficial microorganisms. It can be used to influence landlady with microbial repair in the digestive tract [5]. The common microbes used are lactic acid bacteria (LAB), and the dosage use of probiotics that have significant effect is 10^6 - 10^9 CFU / g [6]. Probiotics from fermented cabbage waste can be improved by the addition of vitamins and minerals to help microorganisms work more optimally. The minerals are substances that stimulates the growth of natural antibiotics and antioxidants. Minerals in the fermentation process can increase the carbon and nitrogen content. It can be used by lactic acid bacteria as energy for the fermentation [7]. Vitamins as one part of micronutrients also have an important role that the body uses to grow and develop normally.

This research was conducted to to evaluate the quality of probiotics derived from fermented cabbage waste which was added by vitamins and minerals. The parameters measured are physical, organoleptic and biological parameters. Physical parameters include the pH of the product, organoleptic parameters including texture, color and odor. Biological parameters include the total analytical test of lactic acid bacteria (LAB) that found in fermented cabbage waste product.

2. Materials and Methods

2.1. Materials

The Materials used include materials cabbage waste, a mixture of vitamins and minerals which include POX (CoSO4, KAI (SO4) 2, Na2 SeO3), zagromix (H3BO3, Na2MaO4, Vitamin C, Vitamin B complex, Vitamin E), MgSO4, Urea, NaCl, MnSO4, ZnSO4, CaCl2, FeSo4, CuSO4, and MSG. The tools used in this study are chopper, autoclave, fermented plastic, knives, buckets, measuring cups, stirrers, rubber, digital scales, pH meters, filters, trays, analytical scales, a set of total LAB analysis tools.

2.2. Methods

2.2.1. Fermented Cabbage Waste. Cabbage waste fermentation is done by cutting the cabbage with small size to facilitate the grinding process of cabbage waste into cabbage waste juice, adding 6.7% salt and 8% molasses from the fresh weight of cabbage, after that it is mixed and homogenized and then put into a tightly fermented plastic (facultative anaerobes) for 6 days. After 6 days the fermentation process was carried out by adding vitamins and minerals in accordance with the treatment. The treatment consisted of adding of mineral vitamins as much as 0%, 2.5%, 5%, 7.5% and 10% and was fermented again for 2 days.

2.2. Organoleptic Test. Organoleptic test was conducted with a questionnaire test on panellists of 20 people. Organoleptic test is a test conducted based on the assessment of panellists on the product including color, odor and texture variables [8]. Color is categorized as the fastest and easiest response indicator in the impression of the product to be selected. Texture is an important factor to determine the quality of a product and aroma is also often used as a parameter to determine the good or bad quality of the product [9]. Organoleptic test scale includes:
Odor :
Score 1 : Foul
Score 2 : Odorless
Score 3 : Acid
Score 4 : Very Acid (Fermented)
Color :
Score 1 : Blackish Brown
Score 2 : Yellowish Brown

Score 3 : Light Brown
Score 4 : Dark Brown
Texture :
Score 1 : Wet
Score 2 : Mushy
Score 3 : Clot
Score 4 : Crumble

2.3. Potential Hydrogen Test (pH). The pH test conducted with pH meter.

2.4. LAB Total Test. Lactic acid bacteria is defined as a group of bacteria that form lactic acid, as the only product or main product in carbohydrate metabolism [2]. LAB count is determined by the Cup Count method (Total Plate Count) using the grow media of Man Rogosa and Sharpe (MRS). Lactic acid bacteria count is one of the factors that needs to be evaluated to assess the product has good quality [10].

2.5. Data Analyze. This study uses a complete randomized design (CRD) with 5 treatments and 3 replicates. The treatment consists of the addition of mineral vitamins as much as 0%, 2.5%, 5%, 7.5% and 10%. The data obtained were analyzed with ANOVA using the Statistical Product and Service Solutions 14 program if there is a difference between treatments conducted Duncan's test. The description of the treatment is as follows:

T0 = Fermented cabbage waste without the addition of vitamins and minerals
T1 = Fermented cabbage waste with the addition of vitamins and minerals 2.5%
T2 = Fermented cabbage waste with the addition of vitamins and minerals 5%
T3 = Fermented cabbage waste with the addition of vitamins and minerals 7.5%
T4 = Fermented cabbage waste with the addition of vitamins and minerals 10%

3. Results and Discussion

3.1. Physical Quality of Organoleptic

Organoleptic quality analysis is based on responses from respondents. The results of organoleptic quality test are presented in Table 1.

Table 1. Average of Organoleptic test result fermented cabbage waste with the addition of vitamins and minerals

Treatment	Addition (%)	Variables		
		Odor	Color	Texture
T0	0	3.00^{ab}	2.33^{c}	3.23^{ab}
T1	2.5	3.17^{a}	2.90^{b}	3.23^{ab}
T2	5	3.17^{a}	3.08^{ab}	3.50^{a}
T3	7.5	3.35^{a}	3.32^{a}	3.50^{a}
T4	10	3.50^{a}	2.12^{c}	3.00^{b}

Numbers followed by the same letter are non significant from the real level 5%.

3.1.1. Odor. Odor is another important variable in assessing feed product. Odor is also used as a parameter to determine the quality of the product produced. The organoleptic test scale with odor variable of fermented cabbage waste on each treatment ranged from foul odor (reek) to acid odor [9]. The average value of numerical scale aroma of fermented cabbage waste is highest in the fermentation of cabbage waste with the addition of 10% vitamins and minerals (T4) which is 3.50 and the lowest average value in the treatment of fermented cabbage waste by control treatment (T0) which is 3.00 (Table 1). The results of statistical calculations show that fermented cabbage waste with the addition of vitamins and minerals treatment had no significant effect ($P > 0.05$) on the aroma of fermented cabbage waste. The addition of vitamins and minerals does not significant effect aroma changes. The aroma of fermented cabbage waste is acidic, cabbage-smelling, and slightly smelling sweet because of the addition of molasses. Fermentation aroma will usually be more acidic if the amount of lactic acid bacteria also increased and enzymatic rate of salt can increase the aroma (odor). The occurrence of increased aroma can be caused by the enzymatic rate of salt that can inhibit the growth and development of strong odors (aromas) [11].

3.1.2. Color. Color is one of the considerations in choosing a product. Color is categorized as the fastest and easy response indicator in giving the impression of the product to be selected [9]. Organoleptic test scale on the color of fermented cabbage waste on each treatment ranged from black brown to dark brown. The average value of numerical scale color of fermented cabbage waste is highest in the treatment of fermented cabbage waste with the addition of 7.5% vitamins and minerals (T3) which is 3.32 and the lowest average value in the treatment of fermented cabbage waste with the addition of 10% vitamins and minerals (T4) which is 2,12 (Table 1). The results of the variance analysis showed that the treatment of the addition of vitamins and minerals to fermented cabbage waste products had significant affect ($P < 0.05$) on the color of fermented cabbage waste. This is due to visual observations that there are color differences between cabbage waste which gets the treatment with the addition of vitamins and minerals 2.5% (T1), 5% (T2), 7.5% (T3) and 10% (T4) compared to the product of fermented cabbage waste control treatment. The color of fermented cabbage waste with the addition of vitamins and minerals turned in to dark brown. This can be caused by the constituent materials such as the addition of molasses or the addition of vitamin c. Vitamin C (ascorbic acid) is a reducing compound and can also act as a precursor for the formation of no enzymatic brown color [12].

3.1.3. Texture. Texture is an important variable for analyzing surface of various types of products. Texture is an important factor to determine the quality of product besides color and odor. Texture is a characteristic of a substance as a result of a combination of physical properties include size, shape and elements of material formation that can be felt by the senses and taste [8]. Organoleptic test scale with variable texture of fermented cabbage waste on each treatment ranged from wet to crumble. The average value of numerical scale texture of fermented cabbage waste is highest in the fermented cabbage waste with the addition of 7.5% vitamins and minerals (T3) which is 3.50 and the lowest average value in the treatment of fermented cabbage waste with the addition of 10% vitamins and minerals (T4) which is 3.00 (Table 1). The results of statistical calculations showed that fermented cabbage waste with the addition of vitamins and minerals had significant effect ($P < 0.05$) on the texture of fermented cabbage waste. This is due to visual observations that there are texture differences between cabbage waste which gets the treatment with the addition of vitamins and minerals 2.5% (T1), 5% (T2), 7.5% (T3) and 10% (T4) compared to fermented cabbage waste control treatment. The changes that occur in texture variable are not too significant. The average of each treatment is crumble because it had undergone a drying and grinding process. Textures can be affected by materials used such as water and cabbage waste. Changes in texture in a fermented product can be determined by natural components materials such as water, fat, protein and carbohydrates [11].

3.2. Potential Hydrogen (pH)

Table 2. pH value of fermented cabbage waste with the addition of vitamins and minerals

Treatment	Addition (%)	Replicates U1	U2	U3	Average of pH Value
T0	0	4.52	4.39	4.54	4.48$\pm$ 0.08[c]
T1	2.5	4.34	4.52	4.43	4.43$\pm$ 0.09[c]
T2	5	3.98	4.87	5.35	5.07$\pm$ 0.25[b]
T3	7.5	5.63	4.81	5.34	5.26$\pm$ 0.42[b]
T4	10	6.45	6.00	6.02	6.16$\pm$ 0.25[a]

Numbers followed by the same letter are non significant from the real level 5%.

Potential Hydrogen (pH) is one of the important factors to be considered during the fermentation process. The pH of fermented products has decreased because of microbial activity, pH is one of the stability indicator of a fermentation process [13]. The highest pH value was found in the fermented cabbage waste with addition of 10% vitamins and minerals (T4) which is 6.16 and the lowest pH value was found in the fermented cabbage waste with control treatment (T0) and the treatment with addition of 2.5% vitamins and minerals (T1) which is 4.43 (Table 2). The results of variance analysis showed that the addition of vitamins and minerals treatment of fermented cabbage waste products had significantly affected ($P < 0.05$) on the pH value of fermented cabbage waste. These results showed that the higher the concentration addition of vitamins and minerals, the higher the pH produced. This can be caused by the high addition of minerals, so some bacteria such as have a slight growth inhibition. If there is a condition such as high mineral concentrations causes an increase in pH, it can be caused by high mineral additions, and some microbes such as *L. bulgaricus* experienced growth barriers due to high mineral concentrations [14]. Microbes only require only a small amount of minerals even though minerals are important for their growth. Lactic acid bacteria can live at a low pH of 2 - 6.5 [15].

3.3. Lactic Acid Bacteria Count

Table 3. Lactic Acid Bacteria count of fermented cabbage waste with the addition of vitamins and minerals

Treatment	Addition (%)	Replicates U1	U2	U3	Average of Log LAB (10^6 CFU/ml)
T0	0	0	0	0	0.0[c]
T1	2.5	0	0	0	0.0[c]
T2	5	4.52	3.50	6.71	4.9$\pm$ 1.64[ab]
T3	7.5	8.11	1.51	4.23	4.6$\pm$ 3.32[b]
T4	10	8.54	8.12	7.60	8.1$\pm$ 0.47[a]

Numbers followed by the same letter are non significant from the real level 5%.

Lactic acid bacteria (LAB) is bacteria that can convert glucose into lactic acid. Lactic acid bacteria (LAB) are natural microflora that belong to probiotic bacteria and can help fermentation process of fruit, vegetables or other ingredients that can inhibit the growth of pathogenic bacteria in these substances [16]. The higher LAB count was found in the fermented cabbage waste with the addition of 10% vitamins and minerals (T4) which is 8.1 x 106 CFU / ml and the lowest average value was found in the fermented cabbage waste by control treatment (T0) and the treatment of addition 2.5 % vitamins and minerals (T1) which is 0 x 106 CFU / ml (Table 3). The results of variance analysis showed that the addition of vitamins and minerals treatment of fermented cabbage waste product had significant affect (P <0.05) on the total lactic acid bacteria (LAB) of fermented cabbage waste. The results showed that the higher the percentage of vitamin and mineral addition, the higher the total LAB in cabbage waste fermentation. Lactic acid bacteria is influenced by the addition of vitamins and minerals which are a food source for LAB. Lactic acid bacteria growth is influenced by the availability of food sources to grow [17]. Vitamins and minerals are nutrients needed by bacteria to metabolism. Vitamin C, A, B1, B2 and vitamin B3 are vitamins that are needed for microbes [14]. In addition, microbes also need minerals such as micro elements K, Ca, Mg, Cl, Fe, Mn, Co, Cu, Zn and Mo.

4. Conclusion
Based on the results of the study, the addition of vitamins and minerals with different treatments can improve the quality of cabbage waste fermentation seen from the potential aspects of hydrogen and total lactic acid bacteria. But it does not significantly influence the physical quality of organoleptics.

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