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Penentuan Nilai pH dan Kadar Vitamin C pada Kimchi dan Acar yang Difermentasi Secara Spontan oleh Bakteri Laktat Asam

[Determination of pH value and Vitamin C Levels in Spontaneous Fermented Kimchi and Pickles by Acid Lactic Bacteria]

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ABSTRACT

Fermentation is the process of chemical transformation in an organic substrate through the activity of enzymes produced by microorganisms. Food fermentation provides some nutritional benefits. It also helps to maintain and increase the nutritional value of food such as vitamin C. Vitamin C, which satisfies the nutritional needs of a balanced diet, is found in abundance in fruits and vegetables. However, there is currently no preservation method that can maintain the amounts of ascorbic acid that are initially found in fruits and vegetables. This study aims to determine the effect of Vitamin C on pickle and kimchi fermentation process on some vegetable (red chilli, cucumber, water pumpkin and chinese cabbage). The result of analysis showed red chili pickle and cucumber pickle with no salt addition have vitamin C of 42.50 ± 2.07 mg/100gr and 16.13 ± 1.06 mg/100gr, respectively, while with 2.5% salt addition has vitamin C of 29.30 ± 4.14 mg/100gr and 8.80 ± 0.70 mg/100gr, respectively. While water pumpkin kimchi and Chinese cabbage kimchi with no salt addition have vitamin C of 20.50 ± 1,65 mg/100gr and 20.50 ± 0.71 mg/100mg, while kimchi and Chinese cabbage with 2.5% salt addition have vitamin C of 25.81 ± 2,30 mg/100gr and 34.12 ± 1.48 mg/100gr, respectively. Therefore, it showed that the addition of salt ingredient on red chili and cucumber pickle might reduce the vitamin C levels, while it might increase the vitamin C levels of water pumpkin and Chinese cabbage kimchi.

Keywords: fermentation, kimchi, pickle, vegetable, vitamin C

ABSTRAK

Fermentasi adalah proses transformasi kimia pada substrat organik melalui aktivitas enzim yang dihasilkan oleh mikroorganisme. Fermentasi makanan memberikan beberapa manfaat gizi. Fermentasi juga membantu mempertahankan dan meningkatkan nilai gizi makanan seperti vitamin C. Vitamin C, yang memenuhi kebutuhan gizi dari pola makan seimbang, ditemukan dalam jumlah banyak pada buah-buahan dan sayuran. Akan tetapi, saat ini belum ada metode pengawetan yang dapat mempertahankan jumlah asam askorbat yang awalnya ditemukan pada buah-buahan dan sayuran. Penelitian ini bertujuan untuk mengetahui pengaruh Vitamin C pada proses fermentasi acar dan kimchi pada beberapa sayuran (cabai merah, mentimun, labu air, dan sawi putih). Hasil analisis menunjukkan acar cabai rawit merah dan acar mentimun tanpa penambahan garam memiliki kadar vitamin C masing-masing sebesar 42,50 ± 2,07 mg/100gr dan 16,13 ± 1,06 mg/100gr, sedangkan dengan penambahan garam 2,5% memiliki kadar vitamin C masing-masing sebesar 29,30 ± 4,14 mg/100gr dan 8,80 ± 0,70 mg/100gr. Sedangkan kimchi labu air dan kimchi sawi tanpa penambahan garam memiliki kadar vitamin C masing-masing sebesar 20,50 ± 1,65 mg/100gr dan 20,50 ± 0,71 mg/100mg, sedangkan kimchi dan sawi dengan penambahan garam 2,5% memiliki kadar vitamin C masing-masing sebesar 25,81 ± 2,30 mg/100gr dan 34,12 ± 1,48 mg/100gr. Hal ini menunjukkan bahwa penambahan bahan garam pada cabai merah dan acar mentimun dapat menurunkan kadar vitamin C, sedangkan penambahan bahan garam pada acar labu air dan acar sawi dapat meningkatkan kadar vitamin C pada acar labu air dan acar sawi putih.

Kata kunci: fermentasi, kimchi, acar, sayur, vitamin C

Introduction

One of food processing which applied to extend their shelflife and maintain the nutritional content is process of fermentation. Fermentation is process that modifiy the chemical molecule into an organic substrate through the activity of enzymes produced by microorganisms (Kusuma et al., 2020). Fermentation occurs when the decomposition of carbohydrate and amino acid compounds in a processed product requires no oxygen or anaerobic (Megawati, 2017). Some of fermetation food product are pickle, sauerkraut, kimchi, yogurt, kefir, or cabbage acid originating in the Americas, as well as salt vegetable processing originating from Indonesia. Fermentation is carried out because it can increase economic value and health benefits, extend storage life, and process other derivatives (Anggraeni et al., 2021). The prevention of degradation that occurs in vegetables is by applying the fermentation process in so that the physical and chemical characteristics of the quality of vegetables will not be reduced (Aliya et al., 2016). Vegetables are one of the food ingredients that have a short shelflife and have nutritional value that is susceptible to damage, such as vitamin C.

Based on the source of microorganisms, fermentation is divided into two types, namely, spontaneous and non-spontaneous. Spontaneous fermentation is a process of fermenting food that does not add a starter to a microorganism, thus its easy to develop. Microorganisms will actively develop and transform the fermented food into the desired product (Pradipta, 2017). Spontaneous fermentation processes will produce a different end product and end quality or uncertainty caused by the number of active microbes of different kinds (Bayuwati, 2019). This spontaneous fermentation techniques can be easily applied to vegetable as the solution for maintaining the shelf life of their nutritional value and self life. A highly varied group on vegetable spontaneous fermenting known as lactic acid bacteria (LAB).

LAB is a necessary bacterium in a variety of vegetable fermentations that is naturally present in the vegetables themselves. Exploiting these bacteria, combined with the proper salt and temperature supply, will produce a good quality fermentation product (Surbakti & Hasanah, 2019). LAB has an ability to produce lactic acid from sugar. Lactate bacteria generally react negatively to catalase and do not form spores. Several genera of lactic acid bacteria, namely Enterococcus, Streptococcus, Leuconostoc, Lactobacillus, Lactococcus, and Pediococcus (Ruiz-Rodríguez et al., 2017). LAB strains are also referred to as rapidly proliferating bacteria that are capable of a variety of metabolic processes. Numerous advantageous substances, including organic acids, antimicrobials, and special enzymes that may convert complex organic molecules into straightforward useful chemicals, are produced as a result of metabolic activities (Von Wright & Axelsson, 2019). Therefore, the key to the advantages and applications of LAB is its quick growth features and metabolic activity. The famous LAB fermentation in vegetable are pickle and kimchi. Generally, pickled and kimchi is served as an appetizer and a unique seasoning that contributes to the core flavor on food.

Pickle is a food-processed product that uses preservation methods by applying fermentation processes. The fermentation process occurs when sugar is converted into acid, which can occur due to the activity of lactic acid bacteria (BAL). Pickle is one of the complex foods that consists of the fermentation of vegetable, red garlic, pepper, and salt solution, but if consumed excessively, it will cause digestive disorders. The cholesterol content in vegetables tends to be low as well as contains complex calories and nutritional values. If consumed, it will help to increase and regulate the amount of fluid needed by the body, help to control diabetes, reduce the risk of sputum, and help to lose weight (Bayuwati, 2019). Kimchi is a traditional fermenting vegetables with lactic acid bacteria (LAB). The fermentation of kimchi involves a variety of bacteria, however during the salting of the cabbage and the fermentation process, LAB take over while the spoilage bacteria are inhibited. Vegetables, together with other nutritious functional foods like ginger, garlic, and red pepper powder, are the main constituents of kimchi. Since kimchi is thought to be a source of LAB, it can be regarded as a vegetable prebiotic food that offers health advantages. Additionally, the fermentative byproducts from the functional ingredients greatly increase the functionality of kimchi (Park et al., 2014).

Some of the vegetables developed in this study are red chili (Capsicum annuum L.), cucumber (Cucumis sativus), chinese gabbage (Brassica rapa subsp. pekinensis) and water pumpkin (Lagenaria siceraria). Red chili is a fruit and plant slassified as food flavor enhancer. A popular salad ingredient, cucumber is prized for its crisp texture and juiciness. But it lacks flavor, is really watery, and isn't very nourishing (Mariod et al., 2017). Chinese gabbage known as white gabbage is a vegetable that can be consumed in raw form or in processed form in various Indonesian dishes, such as, lodeh vegetables, noodles, stir-fries, pickles, gado-gado, pecel and so on. In addition to being useful as a food ingredient, Chinese gabbage are also useful for treating (therapy) various diseases. Water pumpkin is a plant that is widely known by the public as a type of vegetable. The use of water pumpkin is more often used as a cooking dish by the public. However, many people do not like water pumpkin if it is used as a food dish. In fact, water pumpkin has health benefits. Water pumpkin contains calcium, iron, vitamin C, saponins and polyphenols (Marliana, 2011). Therefore, this study was conducted to determine the method of manufacture and the effect of fermentation on the pH and vitamin C values of pickles and kimchi.

Materials and Methods

Materials

Pickle: The sample used was 200 g of fresh red chili (Capsicum annuum L) and 200 g of cucumber. The ingredients used were white garlic (8 g), salt (2.5%), sugar, acetic acid (1.2%), and lime juice. Kimchi: Chinese cabbage water pumpkin, garlic, red chili powder, ground ginger, onion, rice flour, water, salt, sugar, lime leaves. The materials used for chemical testing were 10% H2SO4 (Merck), 1% amilum (Merck), iodine, NaOH (Merck), distilled water and filter paper.

Equipments

The equipment used in this research was an micropipette (Eppendorf), beaker glass (Pyrex), erlenmeyer (Pyrex), measuring flask (Pyrex), volume pipette (Pyrex), oven (Memmert), analytical balance (Radwag), pH meters (DrGray), thermometers (Gea Medical), spatula, aluminium foil, container, spoons, knives, pans, stoves, chopper, filing paper, and burette.

Preparation of Pickle and Kimchi fermentation

Red chili pickle: The preparation of red chili pickles begins with washing 60 grams of fresh red chili and removing the seeds. Then, the fresh red chili is cut, soak it in 10g/l of limestone water for 30 minutes, rinsed with clean water, and drained. The cut chili is then blanced in hot water at 82°C for 3 minutes. The treatment process involves mixing a solution of 250 mL of water, 2.5% salt, 1% sugar, 1% acetic acid, and 8 g of garlic until homogeneous, followed by the addition of the fresh red chili. The untreated pickle (control) contains no added salt. The mixture is heated at 100°C for 2 minutes before fermentation begins. Fermentation is carried out for 7 days at room temperature.

Cucumber pickle: Cucumbers as much as 200 g are cut into cubes of ± 1.5 cm, then put the cucumbers and the prepared fine spices into a glass jar containing a salt solution of 0% and 2.5% concentration. The space inside the glass jar must be conditioned that there is no air space or anaerob. Cover using aluminum foil before using the glass jar lid, and recoat all sides of the glass jar using aluminum foil. The fermentation process will last for 7 days, at the room temperature.

Water pumpkin kimchi: wash the water pumpkin and peel it. Cut the chayote into thin, long strips. Then add spices such as garlic, chili powder, onion, ginger, sugar and kaffir lime leaves. Then blanching is done at 60^{o}C for 5 minutes. Then soak the kimchi in a 2.5% salt solution for 2 hours at room temperature, and continue with fermentation at a cold temperature of 10^{o}C for 7 days.

pH Analysis of pickle and kimchi

A pH meter was used to measure the pH. The pH meter is first calibrated using a buffer for pH 4 and pH 7. The pH meter electrode was dipped into 10 ml samples to take measurements.

Vitamin C Analysis of Pickle and kimchi

The analysis was followed by Nadhilah et al., with silght modification (Nadhilah et al., 2025). The vitamin C analysis starts with weighing 10 grams of sample, mashing it, and filtering the filtrate using filter paper. Then, the filtrate is put into an erlenmeyer, followed by the addition of 3 drops of 10% H2SO4 solution and 5 drops of 1% amylum solution. After that, the titration was carried out with standard iodine solution until the color change into blue occurred. The titration volume was recorded, and this process was repeated three times. To obtain accurate results, the average titration volume was calculated. The formula for vitamin C content (mg/100 g of material) can be seen in equation 1

$$\text{Vit. C (mg/100 g)} = \frac{\text{iod (ml) x 0.01 x 0.88 x dilution x 100}}{\text{sample (ml)}} \quad \text{.......... (1)}$$

Results and Discussion

pH Analysis of pickle and kimchi

The results of the analysis for the pH level of pickles and kimchi carried out every day until 7 days. During pickle and kimchi fermentation, lactic acid bacteria (LAB) were spountanous grow and lower the pH to a point where competing organism are no longer able to grow (Hayek & Ibrahim, 2013). A pH value less than 7 indicates that the solution is acidic. Pickle has an acidic state with a pH range of less than 4.6 (Rahma, 2019). The decrease in pH levels is caused by an increase in total lactic acid. Based on Table 1, the results of pH analysis on pickle control (no salt) is 5 while red chili pickle and cucumber pickle with 2.5% salt produces a pH of 4 from the first day to the 7th day. It shown that the utilization of 2.5% salt has effect on a acidity during pickle fermentation.

Pickle fermentation generally occurs for 2 to 3 weeks with a stable temperature between 21-24ºC (Fakhira et al., 2023). Pickle fermentation is influenced by several factors, such as the salt addition or acid solution, anaerobic, temperature, lactic acid bacteria activity and competing microorganisms (Mardhatillah et al., 2021). The concentration of salt in fermentation affects the acidity value. The presence of salt and acid from fermentation will inhibit the growth of unwanted microbes, and delay tissue softening caused by enzymes. The proteolytic and spoilage bacteria are intolerant to the media with 2-2.5% salt, thus causing succesfull fermentation (Setiawan et al., 2013). Based on the data, pickles with a 2.5% salt concentration produce a pH value of 4 so that it can be said have a lower pH or higher acidity compared to pickle with no salt. The decrease in pH is due to an increase in total lactic acid. pH 4 in pickle is a good condition for pickle products, because the pH in pickles must be 4.5 or less than 4.5 (Ruma et al., 2020). Based on other studies, the higher salt concentration, which 3-15% salt, might inhibit the total acid produced. The total acid value in sweet potato pickles on the 9th day of fermentation with 3% salt was 0.80, with 9% salt was 0.40, and with 12% salt 0.30 (Setiawan et al., 2013). High salt concentration will inhibit the growth of LAB so the total lactic acid will produce last longer and affects the total acid and pH of the final fermentation product. Another thing that affects pH stability in different salt concentrations is likely influenced by the length of fermentation.. Another thing that affects pH stability in different salt concentrations is likely influenced by the length of fermentation. The longer the fermentation, the lower the pH level in pickles (Ruma et al., 2020).

Table 1. Results of pH testing of pickle and kimchi

Sample	Treatment	pH value (day)						
		1	2	3	4	5	6	7
Red chili pickle	No Salt	5	5	5	5	5	5	5
	Salt 2.5%	4	4	4	4	4	4	4
Cucumber pickle	No Salt	4	4	4	4	4	4	4
	Salt 2.5%	4	4	4	4	4	4	4
Water pumpkin kimchi	No Salt	5	4	4	4	4	4	4
	Salt 2.5%	5	4	4	4	4	4	4
Chinese cabagge kimchi	No Salt	5	4	4	4	4	3.5	3.5
	Salt 2.5%	5	4	4	4	4	3.5	3.5

Based on Table 1, the pH test results indicate that the chinese cabbage kimchi and water pumpkin kimchi samples experienced a decrease from day 1 to day 7.The changes that occurred were similar in both of kimchi without salt and those treated with 2.5% salt. The pH decrease in kimchi is caused by the growth of lactic acid bacteria that produce acid compounds during the fermentation period. Lactic acid bacteria convert the glucose contained in chinese cabbage and water pumpkin into lactic acid. The longer the fermentation process, the more it will decrease the pH level in kimchi (Iwansyah et al., 2019). The decrease in pH values in the control kimchi and the 2.5% salt treatment did not show any difference, indicating that the 2.5% salt concentration added in the kimchi did not affect the change-over on pH value. This is in accordance with Aristiyan et al., who stated that a 2.5% salt concentration for microorganism growth cannot affect the pH level (Aristyan et al., 2014).

The pH value of chinese cabbage kimchi, on the 7th day of fermentation decreased to 3.5. This conditions might be influenced by several factors such as the peak activity of bacteria and the growth of different microorganisms (Kaban et al., 2019). In the early stages of fermentation, the activity of lactic acid bacteria can be higher, producing lactic acid that lowers the pH. After a few days, bacterial activity begins to decrease, and this can slow down the drop in pH. During the fermentation process, variations in the composition of microorganisms can occur. Some types of dominant lactic acid bacteria have specific pH preferences, and when they dominate, the pH becomes more stable. pH is used to determine the level of acidity or alkalinity of a substance and solution. High water content in water pumpkin kimchi will affect the pH; high water content will make the pH of kimchi high. Conversely, low water content will cause the pH of kimchi is lower (Sadek et al., 2009). Chinese gabbage has water content of 90% (Winarsih et al., 2012), and water pumpkin has a water content of 93% (Yuliani et al., 2005). Kimchi chinese cabage also has water content of 8,82-9,33% (Ardiyanto, 2019). Kimchi water content has a water content of 11%. This shows that the water content of chinese cabbage shows a lower water content compared to water pumpkin, so that this makes the final pH of white cabbage kimchi lower.

LAB is divided into two, namely homofermentative and heterogenic. The homofermentative fermentation process produces only one type of component, e.g. lactic acid. In contrast, heterofermentation produces a mixture of various compounds or other components such as acetate, ethanol, carbon dioxide, and lactic acids (Aini et al., 2021). During the fermentation process of kimchi and pickle, heterofermentative lactic acid bacteria will be formed, resulting in various acidic compounds that can affect the pH value. In addition, Lactobacillus bacteria that play a role in the fermentation process can produce high levels of lactic acid, so that if consumed can facilitate the digestive system. Kimchi plays a role in anti-inflammatory, antibacterial, antioxidant, anticancer, anti-obesity, probiotic properties, cholesterol reduction, and anti-aging properties for the body (Handayani, 2023).

Vitamin C Analysis of Pickle and kimchi fermented

Based on the Table 2, it can be seen that the vitamin C content in pickle and kimchi have different value on each sample and treatments. Pickle with no any salt treatment have a higher vitamin C compared to kimchi with 2.5% salt treatment. This is similar with Putri's research, which states that the vitamin C content in the control is higher than the vitamin C content of the citric acid addition treatment (Putri & Setiawati, 2017). The addition of salt or acid will be followed by a decrease in vitamin C levels (Tonthawi & Musfiroh, 2023). The addition of higher acetic acid can oxidize ascorbic acid found in vegetables and fruits. The addition of citric acid can reduce Vitamin C levels due to the oxidation process that breaks the OH and H bonds in the ascorbic acid structure (Savitri, 2019).

Table 2. Vitamin C analysis result of pickle and kimchi

Sample	Treatment	Replicate (mg/100mg)			Vitamin C (mg/100g)
		I	II	III	
Red chili pickle	No Salt	39.6	44	44	42.50 ± 2.07
	Salt 2.5%	35.2	26.4	26.4	29.30 ± 4.14
Cucumber pickle	No Salt	17.5	14.9	16.3	16.13 ± 1.06
	Salt 2.5%	9.5	8.1	9.5	8.80 ± 0.70
Water pumpkin kimchi	No Salt	17.6	21.12	17.6	18.77 ± 1,65
	Salt 2.5%	23.76	29.04	24.64	25.81 ± 2,30
Chinese cabagge kimchi	No Salt	21.5	20	20	20.50 ± 0.71
	Salt 2.5%	35.2	37	33.36	34.12 ± 1.48

Meanwhile, kimchi with 2.5% salt showed a higher vitamin C content compared to kimchi with no any salt. It was found that at a salt concentration of 2.5% on water pumpkin kimchi with a fermentation period of 6 days was 35.20 mg/100 gram, while the vitamin C content without the addition of salt was 20.50 mg/100 gram. The increasing of vitamin C content in kimchi is caused by the activity of lactic acid bacteria which help in the process of vitamin C biosynthesis from the substrate (Nur Kholis, 2018). However, the journal that is the reference in the vitamin C test shows that the vitamin C content will decrease along with the fermentation period and the high salt concentration. The decrease in vitamin C content is caused by it being easily soluble in water and easily damaged during the manufacturing process so that it is very easily lost due to the treatment given or slicing and storage (Hayati et al., 2017).

The cucumber pickle with no salt solution concentration had vitamin C of 16.13 (mg/100 g) and the vitamin C of cucumber pickle with 2.5% salt solution concentration had vitamin C of 8.8 (mg/100g). This shows that the use of salt solutions with high concentrations causes vitamin C levels to decrease due to the immersion of food ingredients in salt solutions so that the water content in the ingredients will also dissolve in the salt solution so that the vitamin C levels in food ingredients decrease (Wulan et al., 2019). The vitamin C content has been mixed with water molecules in the tissue which has hypotonic properties and low density. Salt solutions outside the eastern tissue have hypertonic properties so that they have a high density. If there are solutions with different concentrations, water molecules will pass through the membrane until the two solutions are isotonic or balanced. The osmosis process in hypertonic solutions, some water molecules will dissolve into salt molecules so that the higher the salt concentration given in each feeding treatment, the easier it will be for water molecules containing vitamin C to dissolve into salt molecules so that vitamin C will decrease (Kumar et al., 2013). In addition, the main factors that affect the decrease in vitamin C content are temperature, oxygen, light, metal catalysts, initial concentration of ascorbic acid, ratio of ascorbic acid to dehydroascorbic acid, number of microorganisms, and protection from food packaging (Marian & Tuhuteru, 2019).

Another caused of the decreasing of vitamin C were the easily dissolved and easily damaged during processing and storage. Vitamin C is easily dissolved in water where high salt concentrations. Salt absorbs water and nutrients in vegetable tissue so that liquid comes out of the vegetables. The processing process involves blanching and heating, also might damage the vitamin C (Barani et al., 2023).

Conclusion

The pH value on pickles and kimchi with no salt and 2.5% salt has no significant different. Generally, good pickle and kimchi fermented have pH value of 4 - 4.5. The vitamin C analysis in red chili pickle, cucumber pickle, water pumpkin kimchi and chinese cabagge kimchi has different pattern. Vitamin C on pickle product with 2.5% salt has a decrease value compared to pickle without salt addition. While vitamin C on kimchi product with 2.5% salt has a increase value compared to kimchi without salt addition. This shows that the higher the salt concentration used might lower the vitamin C on pickle, but might increase its vitamin C on kimchi product.

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