

Analysis of Vitamin C Levels in Kombucha Tea From Rujak Fruit Skin Waste Using Iodimetric Titration Method

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Abstract. Kombucha tea is a drink produced through a fermentation process of a mixture of tea and sugar using kombucha culture or also called SCOOBY (Symbiotic Culture of Bacteria and Yeast). The basic ingredients for making kombucha tea are granulated sugar solution and tea bags, in this study it was varied using fruit skin. Vitamin C is known as an important antioxidant found in many fruits, including in rujak fruit skin which is often considered waste. The purpose of this study was to determine the levels of vitamin C in kombucha tea fermented from fruit skin waste. This study used qualitative and quantitative methods. The qualitative method uses the Benedict test and the quantitative method uses the calculation of the percentage of vitamin C content using the iodimetric titration method. The results of the qualitative analysis of the Benedict test were positive for vitamin C with a change in the color of the sample solution to green-yellowish and the analysis of the percentage of vitamin C content of kombucha tea can vary depending on the base ingredients used, fermentation time, and storage temperature.

Keywords: Fruit peel waste, Kombucha tea, Iodimetric titration, Vitamin C

1. BACKGROUND

Vitamin C is a very important substance and is rich in benefits for the human body. Vitamin C also acts as an antioxidant that can repair skin cells and tissues damaged by free radicals. In the human body, the formation of vitamin C by the body is very little so that additional vitamin C is needed from external factors, namely by consuming foods and drinks containing vitamin C (Ruayati et al., 2019).

Nowadays, public awareness is increasing in terms of health. One of them is by consuming food and drinks that not only consider aspects of taste and nutrition, but also healthy functional aspects. One of the drinks that produces health benefits is functional drinks (Pratama & Pato, 2015).

Functional beverages are foods that naturally or have been processed, contain one or more compounds that, based on scientific studies, are considered to have certain physiological functions that are beneficial to health. *Kombucha tea* is one example of a functional beverage. This tea is made from sweet tea that is brewed and fermented using *kombucha culture*. *Kombucha culture* is a collection of bacteria and fungi that form gelatinoid substances that grow along the container. Microorganisms in *kombucha* produce enzymes that can convert

sugar content into various nutritional acids, alcohol compounds and vitamins that have many benefits. (Firdaus, et al., 2020).

Currently, *kombucha tea drinks* are widely varied in their preparation methods because the taste produced by *kombucha fermentation* is influenced by the length of fermentation time, the amount of tea, sugar and the mixture of basic ingredients added to the tea medium during the fermentation process. Fruit skin is one of the basic ingredients in making *kombucha tea*. Fruit skin waste is organic waste that is naturally easy to decompose. Although easily decomposed, organic waste is wet waste that can rot or disintegrate, causing an odor that will cause air pollution (Yulistia & Chimayati, 2021). In fact, this waste or waste can be used to make something more useful. This is because fruit skin contains pectin, potassium, vitamin C and antioxidant compounds (Marjenah et al., 2018).

Analysis of vitamin C levels usually uses titration with iodine. The iodimetric titration method (direct titration) is a titration based on a reduction-oxidation (redox) reaction that uses an I2 solution to oxidize the analyte. Iodine is not a very strong oxidizer, so only substances that are strong enough as reductants can be titrated. Vitamin C acts as a reducing agent (reductant) and I2 acts as an oxidizing agent (oxidant). Vitamin C reacts with iodine to form dehydroascorbic acid, with iodine acting as an oxidizing agent and starch acting as an indicator (Rahayuningsih et al., 2022) . Control of fruit peel waste is considered to be underutilized by the wider community. Overall, organic waste management is still a big problem because it is often disposed of directly into landfills without prior processing, this can trigger problems such as causing unpleasant odors and polluting the environment (Faj, et al., 2023) . Based on this background, the author is interested in conducting research with the title "Analysis of Vitamin C Levels in *Kombucha Tea* from Rujak Fruit Peel Waste Using the Iodimetric Titration Method".

Literature Review And Hypothesis Development

Kombucha tea is a tea mushroom that originally came from East Asia and then spread to Europe. Kombucha tea was discovered more than 2000 years ago and is thought to have originated from the North China region. In Indonesia, kombucha was first known in the Sulawesi region, which was brought by airmen to treat chronic diseases. Kombucha is a drink made by fermenting tea and sugar with kombucha culture or also called SCOBY (Symbiotic Culture of Bacteria and Yeast). Inside the SCOBY kombucha there are several groups of bacteria and yeast. The bacterial group has Acetobacter pasteurianu, Acetobacter aceti , Acetobacter xylinum, glucono oxydans. The yeast group has substances such as saccharomyces bisporus, saccharomyces ludwigii, saccharomyces cerevisiae, Zygosaccharomyces sp and *Torolupsis sp* . *Kombucha* fermentation is carried out in an optimum time of 7-14 days and should be carried out in a sterile container made of glass, because containers made of metal can react with the acid contained in *kombucha*. The ideal temperature for *kombucha fermentation* is between 27° C - 30° C, this is because the growth and metabolic activity of microorganisms in *kombucha* can grow optimally at a temperature of 30° C (Nasution & Nasution, 2022).

General content of *kombucha according to* (Lestari & Sa'diyah, 2020) is: vitamins B1, B2, B3, B6, B12, B15, vitamin C, folic acid, glucuronic acid, gluconic acid, acetic acid, chondroitin sulfate, hyalunic acid, lactic acid, acetaminophen, essential amino acids, enzymes, antibiotics.

Fruit peel waste is organic waste that is naturally easy to decompose. Although easily decomposed, organic waste is wet waste that can rot or disintegrate, causing odors that will cause air pollution (Yulistia & Chimayati, 2021). In fact, this waste or waste can be used to make something more useful.

Based on research by Zulkifli et al., (2012), fruit skin contains two to six times more phenolic compounds and two to three times more flavonoid compounds compared to the fruit flesh. In addition, several substances such as anthocyanins, flavonols, kaempferol and xanthone glycerides in fruits were initially found from the skin. Some fruit skins can be used as mixtures or basic ingredients in making drinks.

a. Apple Peel

Apple peel waste can be used as raw material for food and beverages, the amount of apple peel is quite large, which is an estimate of 1/3 of unpeeled apples. So that apple peel waste is widely used as processed because the nutritional content in apple peel is still quite intact, such as carbohydrates, protein, fat, calcium, phosphorus, iron, vitamin B, vitamin C and water (Muflihunna et al., 2022).

b. Papaya Skin

Papaya skin has a nutritional content that is almost similar to the fruit flesh and contains antibacterial compounds such as alkaloids, tannins, steroids, saponins, and flavonoids (Liling et al., 2020).

c. Pineapple Skin

The chemical compounds contained in pineapple skin that are beneficial for health are bromelain, flavonoids, tannins, oxalates, and phytates, the largest components of which are bromelain and flavonoids of the dihydroflavonone group. The compounds contained in this fruit skin have stronger antibacterial activity against gram-positive (Anggreini et al., 2020).

d. Guava Skin

Guava skin contains more antioxidant compounds than the fruit flesh, and this fruit skin also has antioxidant activity including alkaloids, flavonoids, saponins, tannins, and terpenoids (Gunawan et al., 2021).

Vitamin C is also called ascorbic acid which is in the form of white crystals, easily oxidized by air so that the color becomes brown (Prambudi, 2019). Vitamin C is the most unstable vitamin and is easily damaged during processing and storage. The rate of damage increases due to the work of metals, especially copper, iron, and enzyme activity. Exposure to oxygen, prolonged heating in the presence of oxygen, and exposure to light all damage the vitamin C content in food (Techinamuti & Pratiwi, 2003).

Physical properties of vitamin C:

- a. Description: White or slightly yellow crystals or powder that gradually darken under the influence of light. Stable in dry conditions in air. Rapidly oxidized in solution. Melts at a temperature of $\pm 190^{\circ}$ C (Ministry of Health of the Republic of Indonesia, 1995).
- b. Solubility: Easily soluble in water, somewhat difficult to dissolve in ethanol, insoluble in chloroform, ether and benzene (Ministry of Health of the Republic of Indonesia, 1995).

Furthermore, the chemical properties of vitamin C according to the Indonesian Ministry of Health (1995) are:

a. In water that is acidic to litmus paper, the reducing agent is easily oxidized due to the presence of ethanol groups on the C2 and C3 atoms which easily release 2 H atoms.

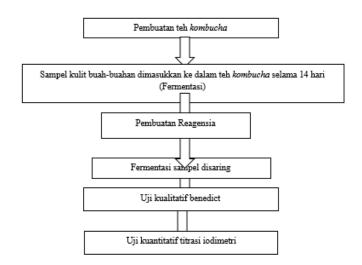
Benefits of Vitamin C

- a. According to (Hasanah, 2018), the function of vitamin C in the body is to form collagen between cells, repair bones and teeth, and prevent boils and bleeding.
- b. The benefits of vitamin C in the body are stored in white blood cells to maintain a strong immune response (Hughes, 2010).
- c. Vitamin C acts as an antioxidant and free radical inhibitor. Free radicals are stimulated by increased ultraviolet radiation from the sun, ultraviolet light penetrates the skin as a reactive substance. The effects of these free radicals are quickly seen during the process of skin contraction and deformation.

Vitamin C helps the body neutralize these free radicals by reducing or protecting against UV rays. Vitamin C is absorbed into cells and remains on the skin for 30 to 36 hours, making it useful as a sunscreen (Pakaya, 2014).

Vitamin C plays an important role in the human body, when the human body lacks vitamin C, symptoms of disease appear such as canker sores, muscle pain, weight loss, and lethargy (Safnowandi, 2022) . The signs of a body lacking vitamin C according to Hasanah, (2018) include:

- a. Dry hair or split ends
- b. Dry, rough skin
- c. Gums bleed and itch easily
- d. Teeth easily decay
- e. Wounds take longer to heal
- f. Experiencing repeated nosebleeds
- g. Pain or swelling in the joints
- h. Anemia



Gambar 1 Alur Penelitian

2. THEORETICAL STUDY

This section describes the relevant theories underlying the research topic and provides a review of several previous studies that are relevant and provide references and foundations for this research to be conducted. If there is a hypothesis, it can be stated implicitly and does not have to be in an interrogative sentence.

3. RESEARCH METHODS

This research is an experimental research by adding apple, guava, pineapple, and papaya skins into *kombucha tea solution*. The design of this research is *laboratory-based*. The population in this study is fruit skins. The samples of this research are apple, guava, pineapple,

and papaya skins and the symbiotic bacteria and fungi are Acetobacter xylinum and Saccaharomyces cerevisiae.

This study consists of five stages, namely preparation of raw materials for fruit skin samples, fermentation of samples with *kombucha tea*, qualitative benedict test, quantitative iodimetry titration test and data analysis. The type of data analysis used is quantitative data, namely the results of measuring the levels of vitamin C in samples converted into Microsoft Excel. Variables are behaviors or characteristics that give different values to something (objects, humans, etc.). The independent variable in this study is *kombucha tea* from rujak fruit skin waste. The dependent variable in this study is the level of vitamin C in *kombucha tea* from rujak fruit skin waste

The tools used are glass jar, 1 test tube, 5 stirring rods, 3 sheets of filter paper, 1 funnel, 1 dropper pipette, 1 analytical balance, 6 brown bottles, 6 Erlenmeyer flasks, 1 stand and burette, measuring flask, measuring cup, volume pipette, spirit lamp, tripod, tripod base, and tube clamp. Materials: 1 tea bag, 250 g granulated sugar, kombucha *SCOBY*, apple, guava, pineapple, and papaya skins. Aquadest, 1% starch indicator, I2 $_{0.1000 \text{ N}}$, Na2S2O3 0.0100 N, Na-Citrate, CuSO4 $_{.5H20}$, KIO3 $_{0.0100}$ N, KI 10% and H2SO4 $_{10\%}$.

4. RESULTS AND DISCUSSION

In the qualitative test of the sample, the results found a color change to green indicating that the sample was positive for vitamin C. Continued with the quantitative test, using iodimetric titration.

 $_{of}$ the $_{Na2S2O3}$ solution after being standardized with the primary standard solution KIO3 $_{obtained}$ a result of 0.0398 N (table 1)

	Table 1 Vol	ume of Na 2 S 2	O 3 Titrant Against K	IO 3
	Burette scale read	ling (50mL)		
No			Volume Used	Normality
	Vol. Beginning	Vol. End	-	
1	35 mL	37.5 mL	2.5 mL	
2	37.5 mL	40 mL	2.5 mL	0.0398 N
3	40 mL	42.6 mL	2.6 mL	
	Average		2,533 mL	

 $_{12}$ solution after being standardized with Na2S2O3 solution _{obtained a result} of 0.1313 N which can be seen in table ₂ below.

	Burette	scale	Volu	
No	reading (5	50mL)	me	Normality
			Used	-
	Vol.	Vol.	-	
	Beginning	End		
1	0 mL	3 mL	3 mL	
2	0 mL	3 mL	3 mL	0.1313 N
3	0 mL	3 mL	3 mL	-
	Averag	ge	3 mL	

	Table 2 Volume	of Titrant I	2 Against Na	$2S_{2}O_{3}$
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The results of measuring vitamin C levels in kombucha tea samples from rujak fruit skin waste are presented in Table 3.

Sample	Titration Volume (ml)	Sample Volume (ml)	% Vitamin
Tea	2	10	2,3124
Kombucha	2	10	2,3124
from waste rujak fruit skin	2	10	2,3124

Table 3. Results of measuring vitamin C levels

Quantitative analysis of vitamin C levels in *kombucha tea* from rujak fruit skin waste produced results of 2.3124% in 10 mL of sample solution.

Discussion

Qualitative analysis of vitamin C in samples was carried out using Benedict's reagent, the test is said to be positive if a green-yellowish color is formed. The Benedict test uses chemical reagents containing cupric sulfate, sodium carbonate, and sodium citrate. The presence of sodium carbonate and sodium citrate makes the Benedict's reagent a weak base. Vitamin C is a strong reducing agent that can reduce Cu $^{2+ ions}$ from the Benedict's reagent into Cu $^{+ ions}$ that precipitate into Cu $_2$ O.

One method that can be used to determine the levels of vitamin C is the iodimetric titration method. The iodimetric titration process consists of 3 stages, namely standardization of Na $_2$ S $_2$ O $_3$ solution with KIO $_3$, standardization of I 2 solution with Na $_2$ S $_2$ O $_3$ and titration of the sample with I $_2$ (Sari et al., 2021). The purpose of standardizing Na $_2$ S $_2$ O $_3$ is to determine the normality of the Na $_2$ S $_2$ O $_3$ solution because the Na $_2$ S $_2$ O $_3$ solution is not a primary standard solution so the solution must be standardized first with the primary standard KIO $_3$. Standardization of I $_2$ with Na $_2$ S $_2$ O $_3$ is to determine the normality of I $_2$ which will be used to determine vitamin C levels (Anggreini et al., 2020).

The basic principle of the iodimetry method is to reduce vitamin C. Vitamin C (acorbic acid) is a strong reducing agent and can be simply titrated with a standard iodine solution. The end point of this titration is the formation of a complex purple iodamylum.

The normality of Na2S2O3 obtained _{from} standardization with KIO3 solution _{is} 0.0394 N while the normality of I2 _{from standardization} with Na2S2O3 solution _{is 0.1313} N. In the sample titration process using I2 solution, the volume of iodine required for titration is proportional to the vitamin C content. The greater the volume of iodine required, the greater the vitamin C content in the sample (Sari et al., 2021)

5. CONCLUSION AND SUGGESTIONS

Based on the research that has been done, it was concluded that the vitamin C content contained in kombucha tea from rujak fruit skin waste 10 mL is 2.3124%. It is recommended to conduct further research on the vitamin C content in kombucha tea from fruit skin waste, for example by using different methods to obtain more accurate analysis of vitamin C levels, different concentrations of mixed ingredients, different basic ingredient mixtures or determining the content of other compounds found in kombucha tea from rujak fruit skin waste.

6. THANK-YOU NOTE

The author would like to thank Poltekkes Kemenkes Medan and also the Food and Beverage Health Chemistry Laboratory of the Medical Laboratory Technology Department of Poltekkes Kemenkes Medan for providing the opportunity to conduct this research.

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