

The Addition of Red Dragon Fruit and Lemon Peels for the Improvement of Fermented Beverage Products

Sofi Nabila¹, Arie Srihardyastutie^{1*}, Sasangka Prasetyawan¹, Aulanni'am¹, Rurini Retnowati¹

¹Chemistry Departement, Faculty of Mathematics and Natural Science, Brawijaya University, Malang, 65145, Indonesia

*Corresponding author: arie_@ub.ac.id

Abstract

Coconut water is part of the coconut fruit, that has bioactive compounds that are thought to own high inhibitor activity to the presence of tannin compounds. However, this compound is antinutritional because it can interfere with macromolecule out there within the body. Considering chance of tannins, these parts are often reduced by fermentation techniques using lactic acid bacteria. This analysis aims to review the addition of dragon fruit and lemon peels in fermentation coconut water using lactic acid bacteria in order to reduce tannin levels and its potential capability as antioxidants. Coconut water fermented drink was prepared by fermentation technique using lactic acid bacteria with the addition of variations in mass of dragon fruit peel and lemon peel consisting of 5 treatments. The optimum conditions for fermentation of coconut water were determined from the decrease in tannin levels, total acid, and the highest total lactic acid bacteria. The optimum conditions for fermentation of coconut water were then tested using an HPLC instrument. Finally, the antioxidant activity was tested using the FRAP (Ferric Reducing Antioxidant Power) method. The results showed that the addition of dragon fruit and lemon peels in fermented coconut water can reduce tannin levels, can increase total acid levels and the number of lactic acid bacteria formed in coconut water fermented drinks. The best proportion was obtained in the treatment of ratio of mass 1:2 resulted in the highest decrease in tannin content of 83.94 mg/100 g followed by a total acid value of 1.13% and the number of lactic acid bacteria was 8.4×10^7 CFU/mL. The HPLC chromatogram showed that fermented coconut water using lactic acid bacteria contained organic acid compounds such as lactic acid, acetic acid, and ascorbic acid. The highest antioxidant activity test was 82.37 mmgAAE/g. This study confirmed that the addition of dragon fruit and lemon peels affected the degradation of tannins and increased antioxidant activity during coconut water fermentation using lactic acid bacteria.

Keywords

Coconut Water, Fermentation, Tannin, Organic Acids, Antioxidant

Received: 30 August 2022, Accepted: 13 Desember 2022

<https://doi.org/10.26554/sti.2023.8.1.100-107>

1. INTRODUCTION

Indonesia is part of the coconut fruit producing countries with a total production of around 15.2 milayrd coconuts per year (Zulaikhah, 2019). Of this amount, the use of coconut that has been utilized most of them only focus on the meat part, while the use of by-products such as coconut water is still limited and often directly disposed of as waste. Even though coconut water is 25% of the coconut fruit which contains complete nutrition including carbohydrates, vitamins, minerals, protein and dietary fiber. The carbohydrate content in coconut water is quite high, so it has the potential as a growth medium for food-producing bacteria.

Utilization coconut water as fermentable sugar has been widely used in the process of making nata de coco (Nurdyansyah and Hasbullah, 2018). In addition, coconut water also contains several bioactive compounds that have the potential

as antioxidants such as tannin compounds. Siti Thomas Zulaikhah (2019) added that coconut water can reduce oxidative stress, which is characterized by a decrease in MDA levels in mice. Coconut water is rich in tannin compounds which significantly reduce free radical formation and have antioxidant effects (Mahayothee et al., 2016). The high content of tannin compounds in coconut water can bind to proteins in the body, so it can interfere with the work of digestive enzymes.

Considering the risk of tannins, one of the efforts to reduce the antinutritional content is through modified microbiological (fermentation) method (Kinanti et al., 2014). In some cases, the fermentation process is also expected to affect the bioaccessibility of other compounds that can increase antioxidant activity and as a method for product development (Verni et al., 2019). The use of lactic acid bacteria (LAB) has been widely used in fermented food products known as food class microorganisms

(microorganisms that do not pose a health risk). Fermented coconut water using lactic acid bacteria culture has benefits such as reducing nutrient absorption, increasing nutritional value and ability of antioxidant activity (Ziska et al., 2017). Several studies have shown that lactic acid bacteria have properties that can degrade tannins in food because they produce tannase enzymes so that can break down tannins into simpler compounds (Shang et al., 2019).

Among the lactic acid bacteria, strains of the *Lactobacillus plantarum* group had tannase activity (Jiménez et al., 2014). Biochemical pathways for tannin degradation by *Lactobacillus plantarum* involve the action of tannase and gallic decarboxylase to hydrolyze the galloil ester bond of tannins to produces gallic acid and glucose (Ananda et al., 2019). Based on research results Midik et al. (2020) showed that the fermentation of coconut water using lactic acid bacteria *L. plantarum* FNCC 0027 produced organic acid compounds as the main product can cause a decrease in the pH value of the substrate, causing a sour taste. One of the way to reduce the acidity level is to add natural sweeteners such as honey. Honey is known as a potential prebiotic that contains oligosaccharides to promote lactic acid bacteria growth (Mohan et al., 2021).

In the fermentation process in produce lactic acid, the growth of lactic acid bacteria can be affected several factors, one of which is the addition of other ingredients. Several studies have shown that the addition of other ingredients in the fermentation process can affect the levels of lactic acid produced (Nurfuzianti, 2021). Based on the results of the study Nurdyansyah and Hasbullah (2018) stated that the concentration of lactic acid produced fermentation with the addition of leather flour substrate bananas, have a fairly high total acid content, this caused by the more sugar there is on the media obtained from the substrate used by lactic acid bacteria as a carbon source to produce organic acids and energy through glycolysis process. In addition to honey, in the manufacture coconut water fermented drink is also added with fruit skin. Fermented fruit peels also have potential as a probiotic carrier that can be a breeding ground for bacteria, especially lactic acid bacteria.

Red dragon fruit peel is part of the dragon fruit that is rarely used and often disposed of as waste. This is caused by people who consume fruits, so it has an impact on to the high volume of waste from the rest of the fruit (Nurdyansyah and Hasbullah, 2018). Problem the waste from the fruit peel has not yet been resolved optimally. Therefore, a processed utilization pattern is needed dragon fruit skin waste food. Several studies have revealed that the skin of red dragon fruit contains vitamin C, flavonoids, tannins, alkaloids, steroids, and saponins (Hendra et al., 2019). Most of the research related to bioactive peptides through fermentation has been carried out by lactic acid bacteria which have proteolytic system. Dragon fruit peel fermentation with lactic acid bacteria can increase inhibition ROS formation. This shows that lactic acid bacteria are able to produce exopolysaccharides (EPS) to increase survival of bacteria and acts as an antioxidant during the fermentation

process (Verni et al., 2019).

Based on the description above, this study aims to study the addition of dragon fruit peel and lemon peel in coconut water fermentation using lactic acid bacteria in reducing tannin levels and their potential as antioxidants.

2. EXPERIMENTAL SECTION

2.1 Tools and Materials

The instruments used in the research were spectrophotometer UV-VIS and HPLC (High Performance Liquid Chromatography). The reagents were Folin-Denis reagent, CaCO_3 , FeCl_3 0.1%, TCA 10%, NaOH 0.1 N, $\text{K}_3\text{Fe}(\text{CN})_6$ 1%, oxalic acid, phenoptlaein1%. The medium for Lactic Acid Bacteria cultivation was MRS media (*de Mann Rogosa Sharpe*) (OXOID). The material used was coconut water, red dragon fruit peels, and lemon peels were collected from Pandaan market, East Java, Pasuruan. The plant specimen was identified and authenticated by plant taxonomist of the Biology Laboratory, Departement of Biology, Brawijaya University. In this study, commercial lactic acid bacteria with the brand Yakult were used. Probiotic Yakult used in this study contained *Lactobacillus plantarum* and *Lactobacillus casei*.

2.2 Methods

2.2.1 Preparation Lactic Acid Bacteria Starter

Inoculum preparation of lactic acid bacteria culture was carried out aseptically in a laminar air flow. Starter cultures of lactic acid bacteria obtained commercially were cultured in 100 g of coconut water with the addition of 10 g of honey which had been sterilized at 70°C for 10 minutes. Then the sterile coconut water was cooled to 40°C and added 10% of commercially obtained lactic acid bacteria colony inoculants. Then immediately cover immediately with plastic and tied with a rubber band. Incubation was carried out at room temperature for 24 h. After 24 h, the starter culture of lactic acid bacteria is ready to be used as a starter culture for further fermentation.

2.2.2 Fermentation of Coconut Water with Addition of Dragon Fruit and Lemon Peel

The substrate was prepared by diluting honey in 10% (v/v) coconut water and sterilized at 70°C for 10 minutes. The sterilized mixture was then cooled to 40°C and inoculated with 10% (v/v) lactic acid bacteria. Before fermenting coconut water with the addition of variations in the mass of dragon fruit and lemon peel, optimization of fermentation time was carried out to obtain the best conditions for bacteria during subsequent fermentation. The optimum time obtained is 48 hours. Next, the dragon fruit peel and lemon peel were added with the following ratio: P₁ (without the addition of fruit peel), P₂ (30 g of dragon fruit peel), P₃ (30 g of lemon peel), P₄ (20 g : 10 g), P₅ (10 g : 20 g). Fermentation was carried out at room temperature under anaerobic conditions. The results of the fermentation were then analyzed for tannin content, total acid, total LAB, antioxidant activity and the resulting organic acid content test.

2.2.3 Tannin Determination

The tannin content of the filtrate fermented coconut water was determined according to Ananda et al. (2019). The fermented filtrate was centrifuged at 3000 rpm for 10 minutes, then 400 μ L of the centrifuged results was taken and 0.5 mL of Folin-Denis reagent was added. Then the solution was allowed to stand for 3 minutes then added 1 mL of saturated Na_2CO_3 solution and vortexed until homogeneous. Then, 10 mL distilled water was added to the solution and the absorbance was measured after 30 minutes at a wavelength of 760 nm using a spectrophotometer, the reagent blank was prepared in the same way without the sample solution. The tannin content was determined based on the standard tannic acid curve of the tannic acid solution and expressed as milligrams per 100 g sample.

2.2.4 Total Acid Analysis

The total acid test was carried out using the titration method which was expressed as a percentage of lactic acid which refers to Zofia et al. (2020) the fermented sample of 10 mL taken, then melted and put into an erlenmeyer, then phenolphthalein indicator was added as much as 2-3 drops then titrated using 0.1 N NaOH. Titration was stopped until a color change occurs constantly pink. Calculation of total acid can use the following formula:

$$\frac{V1 \times N \times B \times 100\%}{V2 \times 1000} \quad (1)$$

2.2.5 Lactic Acid Bacteria Measurement

The total calculation of lactic acid bacteria is used on the Total plate count (TPC) method (Sakul et al., 2020). Then poured the MRS agar medium were incubated at room temperature for 48 hours. Total Lactic acid bacteria was calculated which ranged from 30 to 300 colonies.

2.2.6 Identification of Organic Acids by HPLC

Identification of organic acid fermented filtrate using HPLC refers to Izquierdo Llopert et al. (2020). The fermented filtrate was centrifuged for 10 minutes at room temperature. then the analytes were separated in smart RP 18 column (column length 15 cm and diameter 4.6 mm). The mobil phase used two solvent in an isocratic elution. The mobile phase rate was 1 mL/min, the running time was 15 minutes, the temperature was 25°C, the volume for injection was 100 μ L, and the wavelength at 210 nm.

2.2.7 Antioxidant Activity

Antioxidant activity was measured using the FRAP method (Zhong and Shahidi, 2015). 1 mL of the fermented filtrate placed in a 10 mL volumetric flask and dissolved with distilled water to the limit. Then 1 mL of phosphate buffer and 1 mL of $\text{K}_3\text{Fe}(\text{CN})_6$ were added and then incubated at 50°C for 20 minutes. 1 mL of 10% TCA was added and then centrifuged at 3000 rpm for 10 minutes at room temperature. 1 mL was taken from the top layer, then 0.5 mL of 0.1% FeCl_3 was added.

Then the absorbance was measured at a wavelength of 700 nm. The FRAP value was expressed in mg equivalent of ascorbic acid/ g extract.

2.2.8 Statistic Analysis

Study data were replicated five times each and analyzed using SPSS 16 one way analysis of variance (ANOVA). If there was a significant difference between treatments, it will be continued with Duncans test at $\alpha = 0.05$.

3. RESULT AND DISCUSSION

3.1 Effect of Addition of Dragon Fruit Peel and Lemon Peel to Tannin Levels, Total Acid Content and Total Lactic Acid Bacteria

Before fermenting coconut water with the addition of variations in mass of dragon fruit peel and lemon peel, optimization of the fermentation time was carried out to get the best conditions for bacteria during the next fermentation. Bacterial growth is influenced by acid levels in the fermentation system. The longer the fermentation, the more lactic acid produced (Azwar et al., 2022). Based on the determination of total acid against time, the optimum time of fermentation was found at 48 hours. This is in accordance with the study Midik et al. (2020) which showed that the optimum growth of lactic acid bacteria *Lactobacillus plantarum* in coconut water media was at 48 hours. Determination of this optimum time is used to determine the fermentation time in the manufacture of fermented coconut water products with the addition of dragon fruit peel and lemon peel. Based on the results of the research, fermentation of coconut water with the addition of dragon fruit peel and lemon peel using lactic acid bacteria showed a significant effect on tannin levels, total acid and total LAB produced during the fermentation process (Figure 1).

The results of the analysis of total tannin levels, total acid, and total lactic acid bacteria on the addition of variations in the mass of dragon fruit peel and lemon peel can be seen in Table 1.

3.1.1 Tannin levels

Seen in Figure 1 shows that the value of the tannin content of fermented has decreased. Previously, the initial tannin content in coconut water was quite high at 346.30 mg/100 g. After being fermented using lactic acid bacteria and the addition of fruit peel, there was a decrease in tannin levels ranging from 83-130 mg/100 g. Based on statistical tests showed that the fermentation treatment with the addition of a mass variant of the fruit peel had a significant effect on the tannin content of coconut water. Although, based on the Tukey test, there was no significant difference in the addition of dragon skin (30 g), lemon (30 g) and treatment with a ratio (2:1). The addition of dragon fruit peel and lemon peel in coconut water fermentation contributes to increasing nutrition so that it can be used as a carbon source to increase the activity of lactic acid bacteria. The growth of lactic acid bacteria causes the tannin content in food to decrease. Based on their molecular

Table 1. Tannin Levels, Total Acid, and Total Lactic Acid Bacteria of Fermented Coconut Water added with Dragon Fruit Peels and Lemon Peels

Treatment	Tannin Level (mg/100g)	Total Acid (%)	Total Lactic Acid Bacteria (Log/ml)
P1	134.22 ± 3.65 ^b	0.25 ± 0.072 ^a	7.90 ± 2.91 ^a
P2	105.06 ± 2.65 ^c	0.44 ± 0.069 ^b	7.74 ± 2.23 ^a
P3	95.39 ± 3.25 ^c	1.00 ± 0.08 ^c	8.76 ± 2.3 ^b
P4	104.82 ± 3.32 ^c	0.92 ± 0.056 ^c	7.92 ± 2.58 ^a
P5	83.94 ± 3.71 ^d	1.13 ± 0.058 ^d	8.92 ± 2.16 ^c

Information:
*Data shown as mean of 5 replicates ± standard deviation. Data are shown as mean±standard deviation
*Different superscripts in the same column show very significant effect (P<0.05)

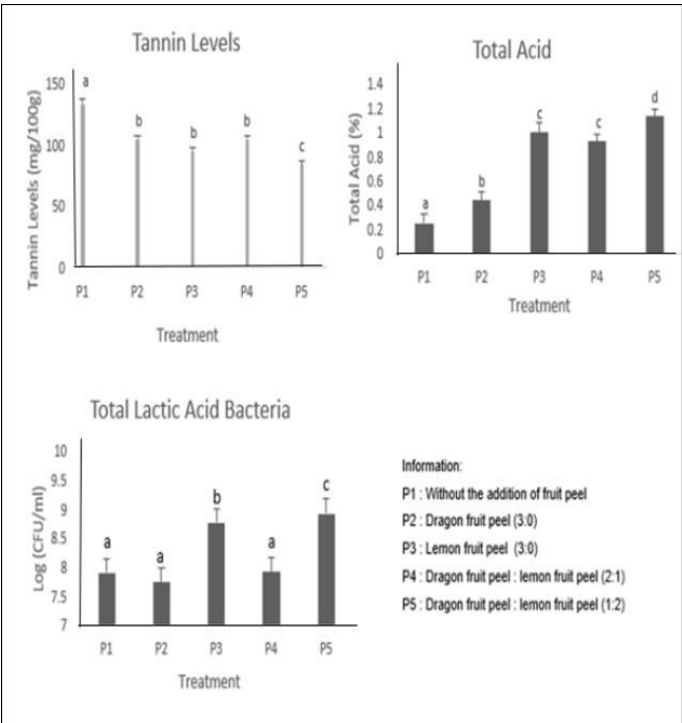


Figure 1. Effect of Addition Fruit Peels to Tannin Levels, Total Acid, and Total Lactic Acid Bacteria

structure, tannins are polyphenolic compounds found in plants that have free radical scavenging activity, where their derivatives are attached to the core of polyols such as catechins found in coconut water. This shows that coconut water has antioxidant activity. Based on research Hassan et al. (2018) shows that coconut water has a fairly high content of tannin compounds. The greater the tannin content, the greater the antioxidant activity. However, this is consistent with Jiménez et al. (2014) which showed that the higher the molecular mass of tannins, the stronger the antinutritional effect. One of the characteristics of tannin compounds as antinutrients is that they are able to form complex-protein bonds which are responsible for inhibiting enzyme digestion (Gemedede and Ratta, 2014). Given the risk

of tannins, it is recommended to reduce the molecular mass of tannins through fermentation techniques using lactic acid bacteria. This is supported by Matsuda et al. (2016) who said that lactic acid bacteria are a group of bacteria that have tolerance for tannins. The highest decrease in tannin levels was found in the coconut water fermentation treatment with the addition of dragon fruit peel and lemon peel with a ratio (1:2) of 83.94 mg/100 g. This decrease was caused by the increasing activity of lactic acid bacteria so that the more the amount of degraded tannins (Shang et al., 2019). Lactic acid bacteria can produce tanase enzymes that play a role in the hydrolysis of tannins. Tannase or tannin acyl hydrolase is an enzyme that catalyzes the hydrolysis reaction of esters found in hydrolyzed tannins and gallic acid esters (Ananda et al., 2019). The activity of the tanase enzyme produced by lactic acid bacteria depends on the difference in the optimal substrate for hydrolyzing tannins. The hydrolysis of tannins by tannase bacteria can release glucose which can act as a carbon source for microorganisms. This is supported by Shang et al. (2019) which shows that the substrate in the fermentation process will affect the survival of lactic acid bacteria.

3.1.2 Total Acid Content

The effect of Effect of addition mass variance of dragon fruit peel and lemon peel on total acid (Table 1). The total acid content ranged from 0.255-1.13%. From the current data, the increase in total acid levels showed an increase in total acid levels, but not significant enough which was indicated by the results of statistical data analysis that did not significantly affect the treatment of adding lemon peel (30 g) and the addition of dragon fruit peel and lemon peel with a ratio (2:1). The coconut water fermentation treatment with the addition of dragon fruit peel and lemon peel with a ratio (1:2) had the highest total acid content of 1.13%. The addition of fruit skin to coconut water fermentation media acts as a carbon source that can optimize coconut water fermentation using lactic acid bacteria (Sakul et al., 2020). This is supported by Le et al. (2021) which states that dragon fruit peel contains inulin and FOS (fructooligosaccharide) compounds which contain carbohydrates, protein and dietary fiber which act as carbon sources

for microorganisms. Meanwhile, dragon fruit skin also contains anthocyanin compounds, tannins and vitamin C. The content of tannin compounds in coconut water and dragon fruit peel can be hydrolyzed by lactic acid bacteria to form glucose and gallic acid (Ananda et al., 2019). During the fermentation process by lactic acid bacteria, there is a breakdown of glucose into lactic acid through the glycolysis process, which converts glucose into pyruvate and converts it into lactic acid (Zofia et al., 2020). In addition, the fermentation process using lactic acid bacteria also produces vitamins (Kang et al., 2020). One of the vitamins that can be produced by lactic acid bacteria is ascorbic acid (Vitamin C) which can be found in the skin of lemons. This is due to the lemon peel contains a fairly high ascorbic acid compound. The formation of lactic acid, gallic acid and ascorbic acid by lactic acid bacteria is the result of the metabolism of lactic acid bacteria from the fermented substrate which accumulates in the total acid formed during the fermentation process. The total acid calculated is the amount of acid produced from the metabolism of lactic acid bacteria.

3.1.3 Total Lactic Acid Bacteria

Lactic acid bacteria is a parameter that acts as an indicator of the success of fermented products. Based on Table 1, it can be seen that the growth of lactic acid bacteria in the coconut water fermentation treatment with the addition of dragon fruit peel and lemon peel increased, but not significantly, which was indicated by statistical data which showed no significant difference in the treatment without the addition of fruit skin, the addition of fruit peel treatment. dragon (30 g), and the addition of dragon fruit peel and lemon peel in a ratio (2:1). The addition of fruit rind mass added to coconut water fermentation has an effect on increasing the number of lactic acid bacteria which is influenced by the availability of substrates in the fermentation media (Anwar, 2018). Glucose in the substrate is one of the important nutrients used for the growth of LAB as an energy source, so the presence of glucose can trigger the growth of LAB colonies in large numbers quickly (Rizal et al., 2016). The results of the analysis of total lactic acid bacteria in fermented coconut water with the addition of variations in the mass of dragon fruit peel and lemon peel in this study showed a range of 7.74 log colonies/mL to 8.92 log colonies/mL or equivalent to 108 colonies/mL. The Indonesian National Standard SNI 7552: 2009 states that the minimum requirement for the total value of lactic acid bacteria in fermented beverages is 10^6 col/mL (Rizal et al., 2016). Based on the results of the study, the total value of lactic acid bacteria in coconut water fermented drinks has met the standards of lactic fermented drinks. The highest total lactic acid bacteria was seen in the coconut water fermentation treatment with the addition of dragon fruit peel and lemon peel in a ratio (1:2) which was 8.4×10^7 CFU/mL. The content of glucose in the substrate has a positive effect on the growth of lactic acid bacteria which can be converted into lactic acid (Parasathi et al., 2020). The amount of LAB has a positive correlation with the amount of acid produced. Microbes will make adjustments to certain substrates, so that

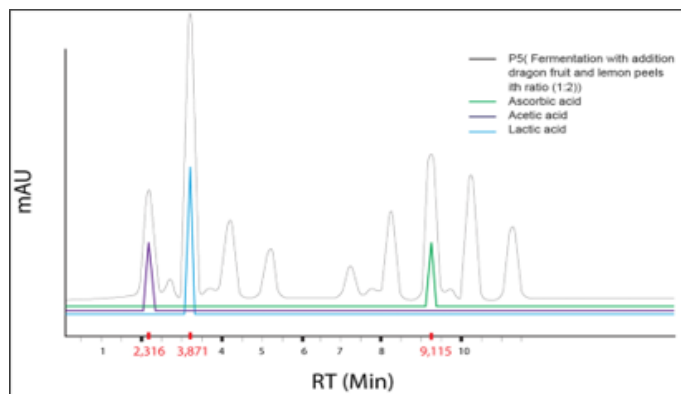


Figure 2. Chromatogram of Organic Acids Fermented by Coconut Water with the Addition of Dragon Fruit Peel and Lemon Peel

the cell growth process increases along with the increase in the amount of nutrients present in the substrate, this also affects the amount of lactic acid which will increase (Zofia et al., 2020). This is supported by Zhuo et al. (2021) who said that the addition of a high mass of lemon peel causes the substrate reshuffle process to produce energy for bacterial cell proliferation. After fermentation, the lemon peel fiber will be degraded into simple carbohydrate prebiotics such as galactose and xylose.

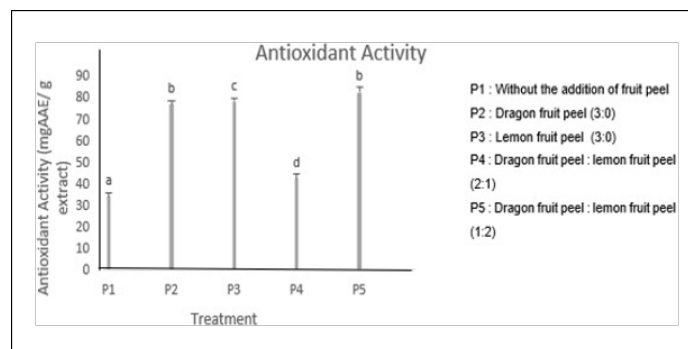
3.2 Effect of Addition of Dragon Fruit Peel and Lemon Peel to Identification of Organic Acid Compounds

Based on the results of the main parameters, namely a decrease in tannin levels, an increase in total acid and total lactic acid bacteria obtained from fermenting coconut water with the addition of dragon fruit peel and lemon peel. Based on the assessment of the decrease in the highest tannin level, it shows that the treatment of fermented coconut water with the addition of dragon fruit peel and lemon peel in a ratio (1:2) using lactic acid bacteria indicates the treatment chosen as the best treatment. This best treatment was chosen due to the presence of tannin degradation activity by the tannase enzyme produced by lactic acid bacteria to reduce the content of antinutrients contained in the fermentation media. Lactic acid bacteria used in this study were obtained commercially (*L.plantarum* and *L.casei*) which are a group of heterofermentative bacteria, which can convert glucose in coconut water into organic acid compounds (lactic acid and acetic acid) under anaerobic conditions (Ananda et al., 2019). The presence of organic acid compounds fermented using lactic acid bacteria can be identified using HPLC (Figure 2). To determine the presence of organic compounds, it is necessary to standardize the organic acids analyzed using HPLC (Andersson and Hedlund, 1983).

Based on Figure 2 shows that the filtrate from fermented coconut water contains organic acid compounds consisting of lactic acid, acetic acid and ascorbic acid. It is based on the correspondence of the peaks of organic acids at a given retention time in the sample chromatogram when compared

Table 2. The Retention Time of Organic Acids Analyzed by HPLC

RT (standard)	RT (sample)	Concentration ($\mu\text{g/mL}$)	Component
2.316	2.316	324.69	Acetic Acid
3.871	3.871	884.62	Lactic Acid
9.115	9.115	447.38	Ascorbic Acid

**Figure 3.** Antioxidant Activity from Fermented Coconut Water with Addition of Dragon and Lemon Fruit Peel using Lactic Acid Bacteria

to the standard chromatogram. The retention time (t_r) of the mixture of organic acids is shown in Table 2. Lactic acid is the acid that is formed with the highest concentration based on Table 2. The formation of lactic acid by lactic acid bacteria is the main metabolic product of lactic acid bacteria (Anwar, 2018). In addition to lactic acid, the fermentation results also showed the formation of quite high ascorbic acid compounds (Table 2). The presence of ascorbic acid compounds in the fermentation results can be correlated with their potential as a source of antioxidants. The content of high concentrations of ascorbic acid can increase antioxidant activity.

3.3 Effect of Addition of Dragon Fruit Peel and Lemon Peel to Antioxidant Activity

The antioxidant activity of fermented coconut water samples was tested using the FRAP method. This method is one of the analyses to determine the antioxidant activity to convert Fe^{3+} to Fe^{2+} . The addition of dragon and lemon fruit peel in coconut water fermentation using lactic acid bacteria has an effect on antioxidant activity produce Figure 3.

Dragon fruit peel and lemon are sources of phenolic compounds that have high antioxidant activity. Flavonoid compounds have antioxidant activity caused by the presence of electron donors with -OH groups attached to the carbon of the aromatic ring (Raharjo and Haryoto, 2019). The use of lactic acid bacteria during the fermentation process can increase the potential to inhibit the formation of reactive oxygen species (ROS) (Medina López et al., 2022). The potential of LAB in increasing the antioxidant activity of foodstuffs is caused by a decrease in pH and the effect of enzymatic analysis during

fermentation (Zhao et al., 2021). The decrease in pH was caused by the high levels of acid produced during the fermentation process. The high acid content indicates the potential to increase antioxidant activity. The acid activity produced during the fermentation process contributes to the metabolism of polyphenols present in fruit peels fermented with LAB to ferulic acid which has the ability to inhibit lipid peroxidation (Verni et al., 2019). Based on the determination of the presence of organic acid compounds. The results showed that the best treatment for fermenting coconut water with the addition of dragon fruit peel and lemon peel using a ratio (1:2) showed that the organic acids detected using HPLC contained high concentrations of ascorbic acid compounds. The high content of ascorbic acid in fermented products is closely related to its antioxidant activity. Vitamin C functions as an antioxidant because it. Effectively capture free radicals, especially ROS or reactive oxygen compounds (Silalahi et al., 2018). As a free radical scavenger, vitamin C can react with superoxide anions, hydroxyl radicals, oxygen singlet and lipid peroxides.

The value of antioxidant activity in this study was expressed in mg ascorbic acid equivalent/g extract (AAE). Ascorbic acid is used as a comparison solution which acts to counteract various extracellular free radicals. This is because vitamin C can reduce Fe^{3+} ions from the active site of the prolyl enzyme hydroxylase to form Fe^{2+} so that enzymes are in their active form. Based on Figure 3, the highest antioxidant activity was shown in the fermentation treatment with the addition of dragon fruit peel and lemon with a ratio (1:2) of 82.37 mg AAE/g extract. then proceed with the fermentation treatment with the addition of lemon peel extract 78.24 mg AAE/g. This is supported by research Tkacz et al. (2020) which showed that the effect of fermentation using *Lactobacillus plantarum* was effective in increasing the antioxidant activity of flavonols. This is related to the results of the study that fermented coconut water with the addition of dragon fruit and lemon peel was more effective and had higher antioxidant activity.

4. CONCLUSION

Based on the research that has been done, then it can be concluded that the addition of mass variants of dragon fruit peel and lemon peel have a significant influence on the highest decrease in tannin levels, the highest increase in total acid and total lactic acid bacteria, respectively, occurred in the coconut water fermentation treatment with the addition of dragon fruit peel and lemon peel using lactic acid bacteria, namely 83.94 mg/100 g, 1.13% and 8.4×10^7 CFU/mL. While the optimum

conditions of the results of the decrease in the highest tannin content have a significant effect on the antioxidant activity produced is 82.37 mg AAE/g extract.

5. ACKNOWLEDGMENT

I would like to thank the supervising lecturer who has provided direction until the research is completed. I also thank you for the invaluable assistance given by several restaurants that have provided fruit peel waste as sampels. We are very grateful for their generous, knowledgeable and constructive assistance.

REFERENCES

- Ananda, T. D., A. Srihardyastutie, S. Prasetyawan, and A. Safitri (2019). Effect of Mixed Inoculums Volume and pH on Anti Nutritional Level in Cabbage Fermentation using *Saccharomyces cerevisiae* and *Lactobacillus plantarum*. *Materials Science and Engineering*, **546**(6); 062004
- Andersson, R. and B. Hedlund (1983). HPLC Analysis of Organic Acids in Lactic Acid Fermented Vegetables. *Zeitschrift für Lebensmittel-Untersuchung und Forschung*, **176**(6); 440–443
- Anwar, . P. U., M. Z. (2018). Making Probiotic Drink of Young Coconut Water (*Cocos nucifera* L) with Starter *Lactobacillus casei* sub R-68. *JOM Faperta*, **5**(1); 1–12
- Azwar, A., H. Hisbullah, A. Irgi, W. Julyadi, A. Adisalamun, M. Mukhlisshien, R. N. R. Nasrullah, A. Abubakar, M. Z. M. Zani, and J. A. J. Ali (2022). Design of Control Loop Pairing in a UCT Bioreactor System. *Jurnal Serambi Engineering*, **7**(1); 2577–2586
- Gemed, H. F. and N. Ratta (2014). Antinutritional Factors in Plant Foods: Potential Health Benefits and Adverse Effects. *International Journal of Nutrition and Food Sciences*, **3**(4); 284–289
- Hassan, R. M., N. A. F. Zulrushdi, A. M. Yusoff, N. Ibrahim, and N. M. Nor (2018). Phytochemical Properties and Antimicrobial Activity of Natural Colorant Extracted from Mesocarp and Exocarp of *Cocos nucifera*. *Journal of Nutritional Biology*, **4**(2); 236–243
- Hendra, R., L. Masdeatresa, R. Abdulah, and Y. Haryani (2019). Antibacterial Activity of Red Dragon Peel (*Hylocereus polyrhizus*) Pigment. *Journal of Physics: Conference Series*, **1351**(1); 012042
- Izquierdo Llopart, A., A. Carretero, and J. Saurina (2020). Organic Acid Profiling by Liquid Chromatography for the Characterization of Base Vines and Sparkling Wines. *Food Analytical Methods*, **13**(10); 1852–1866
- Jiménez, N., M. Esteban Torres, J. M. Mancheño, B. de Las Rivas, and R. Muñoz (2014). Tannin Degradation by A Novel Tannase Enzyme Present in Some *Lactobacillus plantarum* Strains. *Applied and Environmental Microbiology*, **80**(10); 2991–2997
- Kang, W., L. Pan, C. Peng, L. Dong, S. Cao, H. Cheng, Y. Wang, C. Zhang, R. Gu, and J. Wang (2020). Isolation and Characterization of Lactic Acid Bacteria from Human Milk. *Journal of Dairy Science*, **103**(11); 9980–9991
- Kinanti, P. S. K., B. S. Amanto, and W. Atmaka (2014). Kajian Karakteristik Fisik dan Kimia Tepung Sorghum (*Sorghum bicolor* L) Varietas Mandau Termodifikasi yang Dihasilkan dengan Variasi Konsentrasi dan Lama Perendaman Asam Laktat. *Jurnal Teknosains Pangan*, **3**(1); 135–144 (in Indonesia)
- Le, T. L., N. Huynh, and P. Quintela-Alonso (2021). Dragon Fruit: A Review of Health Benefits and Nutrients and its Sustainable Development under Climate Changes in Vietnam. *Czech Journal of Food Sciences*, **39**(2); 71–94
- Mahayothee, B., I. Koomyart, P. Khuwijitjaru, P. Siri Wongwilaichat, M. Nagle, and J. Müller (2016). Phenolic Compounds, Antioxidant Activity, and Medium Chain Fatty Acids Profiles of Coconut Water and Meat at Different Maturity Stages. *International Journal of Food Properties*, **19**(9); 2041–2051
- Matsuda, M., Y. Hirose, M. Kanauchi, S. Hatanaka, and A. Totoka (2016). Purification and Characteristics of Tannase Produced by Lactic Acid Bacteria, *Lactobacillus plantarum* H78. *Journal of the American Society of Brewing Chemists*, **74**(4); 258–266
- Medina López, S. V., C. M. Zuluaga Domínguez, J. P. Fernández Trujillo, and M. S. Hernández-Gómez (2022). Non-conventional Hydrocolloids' Technological and Functional Potential for Food Applications. *Foods*, **11**(3); 401
- Midik, F., M. Tokatlı, S. Bağder Elmacı, and F. Özçelik (2020). Influence of Different Culture Conditions on Exopolysaccharide Production by Indigenous Lactic Acid Bacteria Isolated from Pickles. *Archives of Microbiology*, **202**(4); 875–885
- Mohan, A., N. Gutierrez Maddox, T. Meng, N. He, Y. Gao, Q. Shu, and S. Y. Quek (2021). Manuka Honey with Varying Levels of Active Manuka Factor (AMF) Ratings as An Anaerobic Fermentation Substrate for *Limosilactobacillus reuteri* Dpc16. *Fermentation*, **7**(3); 128
- Nurdyansyah, F. and U. H. A. Hasbullah (2018). Optimasi Fermentasi Asam Laktat oleh *Lactobacillus casei* Pada Media Fermentasi yang Disubstitusi Tepung Kulit Pisang. *Journal of Biology*, **11**(1); 64–71 (in Indonesia)
- Nurfuzianti, R. (2021). Pengaruh Proses Fermentasi Terhadap Kandungan Asam Laktat pada Makanan Fermentasi. *Parapemikir: Jurnal Ilmiah Farmasi*, **10**(2); 1–6 (in Indonesia)
- Parasthi, L. Y. E., D. N. Afifah, C. Nissa, and B. Panunggal (2020). Total Lactic Acid Bacteria and Antibacterial Activity in Yoghurt with Addition of *Ananas comosus* Merr. and *Cinnamomum burmannii*. *Amerta Nutrition*, **4**(4); 257–264
- Raharjo, D. and H. Haryoto (2019). Antioxidant Activity of Mangrove *Sonneratia caseolaris* L using the FRAP Method. *International Summit on Science, Technology, and Humanity*
- Rizal, S., M. Erna, F. Nurainy, and A. R. Tambunan (2016). Karakteristik Probiotik Minuman Fermentasi Laktat Sari Buah Nanas dengan Variasi Jenis Bakteri Asam Laktat. *Jurnal Kimia Terapan Indonesia*, **18**(01); 63–71 (in Indonesia)
- Sakul, S., D. Rosyidi, L. E. Radiati, and P. Purwadi (2020).

- The Effect of Different Starter Cultures on the Fermentation of Yogurt Added with Aqueous Extract of White Oyster Mushroom (*Pleurotus ostreatus*). *Jurnal Ilmu dan Teknologi Hasil Ternak (JITEK)*, **15**(1); 46–51
- Shang, Y.-F., H. Cao, Y.-L. Ma, C. Zhang, F. Ma, C.-X. Wang, X.-L. Ni, W.-J. Lee, and Z.-J. Wei (2019). Effect of Lactic Acid Bacteria Fermentation on Tannins Removal in Xuan Mugua Fruits. *Food Chemistry*, **274**; 118–122
- Silalahi, J., D. Nadarason, and Y. Silalahi (2018). The Effect of Storage Condition on Antioxidant Activity of Probiotics in Yoghurt Drinks. *Asian Journal of Pharmaceutical and Clinical Research*, **11**(12); 2455–3891
- Siti Thomas Zulaikhah, S. T. Z. (2019). Health Benefits of Tender Coconut Water (TCW). *International Journal of Pharmaceutical Sciences and Research*, **10**(2); 474–480
- Tkacz, K., J. Chmielewska, I. P. Turkiewicz, P. Nowicka, and A. Wojdyło (2020). Dynamics of Changes in Organic Acids, Sugars and Phenolic Compounds and Antioxidant Activity of Sea Buckthorn and Sea Buckthorn-apple Juices During Malolactic Fermentation. *Food Chemistry*, **332**; 127382
- Verni, M., V. Verardo, and C. G. Rizzello (2019). How Fermentation Affects the Antioxidant Properties of Cereals and Legumes. *Foods*, **8**(9); 362
- Zhao, Y. S., A. S. Eweys, J. Y. Zhang, Y. Zhu, J. Bai, O. M. Darwesh, H. B. Zhang, and X. Xiao (2021). Fermentation Affects the Antioxidant Activity of Plant-based Food Material Through the Release and Production of Bioactive Components. *Antioxidants*, **10**(12); 2004
- Zhong, Y. and F. Shahidi (2015). Methods for the Assessment of Antioxidant Activity in Foods. In *Handbook of Antioxidants for Food Preservation*. Elsevier, pages 287–333
- Zhuo, L. C., A. S. K. Yong, R. Shapawi, and Y. H. Lin (2021). Effects of Fermented Lemon Peel Supplementation in Diet On Growth, Immune Responses, and Intestinal Morphology of Asian Sea Bass, Lates Calcarifer. *Aquaculture Reports*, **21**; 100801
- Ziska, R., A. Taufik, and D. Supriadi (2017). Uji Aktivitas Antimikroba dan Antioksidan dari Minuman Probiotik Hasil Fermentasi Air Kelapa (*Cocos nucifera*). *Jurnal Farmasi Galenika*, **4**(1); 14–19 (in Indonesia)
- Zofia, N.-Ł., Z. Aleksandra, B. Tomasz, Z.-D. Martyna, Z. Magdalena, H.-B. Zofia, and W. Tomasz (2020). Effect of Fermentation Time on Antioxidant and Anti-ageing Properties of Green Coffee Kombucha Ferments. *Molecules*, **25**(22); 5394
- Zulaikhah, S. T. (2019). Health Benefits of Tender Coconut Water (TCW). *International Journal of Pharmaceutical Sciences and Research*, **10**(2); 474–80