The Effect of Brown Sugar Addition on Total Acid, pH, and Antioxidant Activity of Young Coconut Water (Cocos nucifera L,) Probiotic Drinks

Fahdah Haniyah and Eni Purwani(✉)

Department of Nutritional Sciences, Faculty of Health Sciences, Universitas Muhammadiyah Surakarta (UMS). Jl. A. Yano Tromol Pos, 1 Pabelan, Kartasura, Surakarta, Central Java 57162, Indonesia

ep142@ums.ac.id

Abstract. Coconut water is a clear liquid from the coconut endosperm, containing sugars, vitamins, and minerals. Fermentation of young coconut water with *Lactobacillus casei* can generate a probiotic drink that can maintain digestive tract health and increase immunity. As a carbon during fermentation, the amount and type of sugar affect the chemical properties of probiotic drinks. Adding a certain amount of brown sugar to a probiotic drink can affect the total acid, pH, and antioxidant activity. This study aimed to determine the effect of brown sugar addition on the total acid, pH, and antioxidant activity of young coconut water probiotic drinks. The design of this study employed a single-factor Completely Randomized Design (CRD) method with brown sugar additions (0%, 5%, 10%, and 15%). Determination of total acid by titrimetric method, pH measurement by pH meter, and antioxidant activity by DPPH method. Total acid was analyzed using One-Way ANOVA and LSD with a significance level of $p < 0.05$. pH and antioxidant activity were analyzed using Kruskal-Wallis and Dunnett T3 with a significance of $p < 0.05$. The results showed that brown sugar addition affected total acid ($p = 0.001$), pH ($p = 0.014$), and antioxidant activity ($p = 0.003$). This study concluded that the highest and the lowest total acid and antioxidant activity were 15% and 10% brown sugar addition, respectively. Furthermore, the highest and the lowest pH value was at 0% and 15% brown sugar addition, consecutively. The pH and total acid values of this study followed SNI 2981:2009.

Keywords: Young Coconut Water · Probiotic Drink · Brown Sugar · Total Acid · pH · Antioxidant Activity

1 Introduction

Coconut water is mainly consumed in tropical countries [1]. Coconut water *(Cocos nucifera L.),* frequently referred to as young coconuts, is obtained from a coconut fruit seller [2]. Coconut water is a clear liquid acquired from the endosperm of the coconut and can generate various processed foods [3]. Coconut water contains sugars, vitamins,

B. Ichsan et al. (Eds.): ICHWB 2022, AHSR 61, pp. 120–132, 2023.
https://doi.org/10.2991/978-94-6463-184-5_12

amino acids, enzymes, volatile aromatic compounds, and significant amounts of isotonic electrolytes in the form of potassium and other minerals. The use of coconut water in the food and health sector can be a rehydration solution intravenously or orally in an emergency [4].

The substances in young coconut water include carbohydrates (4.00%), water (95.5 0%), fat (<0.1 0%), ash (0.40%), and protein (0.1 0%). Vitamin C and B complexes are also discovered in coconut water, as much as 2.2 0–3.40 mg per 100 ml for vitamin C. Several minerals include magnesium, nitrogen, sulfur, phosphorus, chlorine, iron, and potassium [5]. Carbohydrates, which compose 4,00% young coconut water in the form of simple sugars (fructose and glucose) are essential nutrients to provide microorganisms with the source of carbon [6].

Young coconut water is a part of the coconut that produces various valuable products with antioxidant potential because it contains phenolic compounds and flavonoids with high levels [7]. Considering the health advantages of coconut water, functional food products such as coconut water probiotic drinks are still limited [8–10]. With its high content of nutrients such as sugar, fat, and protein, microorganisms can benefit from young coconut water to produce functional products for its growth, and the microorganisms can process this coconut water to generate probiotic drinks [11].

Probiotic drinks are functional food products fermented by live LAB (Lactic Acid Bacteria) that produce lactic acid [12]. LAB-fermented drinks can increase and maintain the balance of microflora found in the intestines with the ability to live in the stomach to provide health benefits for the human gastrointestinal tract as a large number of microflora in the intestines [13]. Probiotics are microorganisms that can improve the health of those who consume them in an appropriate and sufficient quantity [14]. *Lactobacillus* species have been successful in generating young coconut water probiotic drinks [1, 9, 15, 16]. *Lactobacillus casei* is one of the LAB (Lactic Acid Bacteria), which is beneficial as a starter in producing fermented drinks from the main ingredient of fruit juice. These bacterial species have the effectiveness of developing into probiotic bacteria and the ability to resist bile salts and stomach acids in the GI system [17].

Young coconut water fermented using LAB probiotics can produce new fermented drink products that provide hydration and probiotic benefits for individuals [9]. The health benefits humans can obtain from probiotic drinks are that they can maintain the health of the digestive tract because they maintain the balance of various intestinal types of microorganisms to remain normal and increase immunity by improving intestinal microflora. In addition, it lowers the risk of colon cancer and overcomes diarrhea by inhibiting the growth of pathogenic bacteria in the intestine [18].

Fermenting coconut water will develop the product into a healthier drink, extend its shelf life, and stabilize its storage and distribution. However, more than the then-nutrition in coconut water is required for the starter culture to start the fermentation process. Adding skimmed milk powder, sucrose, and flavorings will provide the starter culture with the necessary nutrients and enhance the product's taste and general acceptance [19]. Sugar and nitrogen in small amounts are necessary to develop and grow Lactic Acid Bacteria (LAB) with a medium of protein and sugar [6]. Nutritional substrates transform into the raw components required by probiotic bacteria. Simple carbohydrates are the most fundamental bacteria to produce fermented drinks [20].

There is a fermentation process that requires the addition of sucrose, which is a source of carbon for microorganisms. As a result of fermentation, probiotic drinks will have a highly sour taste if not provided with sucrose. Ingredients addition with specific types and amounts in probiotic drinks can benefit health and affect the chemical nature of probiotic drinks. The treatment of young coconut water in probiotic drinks with brown sugar addition became an updated topic in this study because brown sugar can create a distinctive sweet taste and become a source of carbon required by lactic acid bacteria. The chemical composition of brown sugar in every 100 g compose of 76 g of carbohydrates, 76 mg of potassium, 11 g of water content, 3 g of protein, 37 g of phosphorus, and 10 g of fat [21].

The addition of brown sugar will affect the total acid, pH, and antioxidant activity of the young coconut water probiotic drinks. Sugar, which acts as a source of LAB nutrition by breaking down sugar, is converted into ATP during fermentation. The breakdown of sugar will increase acidity due to the production of lactic acid and decrease the level of acidity and the amount of sugar [22].

Based on the background, this study aimed to determine the effect of brown sugar addition on the total acid, pH, and antioxidant activity of young coconut (*Cocos nucifera L.)* water probiotic drinks. The use of young coconut water and brown sugar addition is expected to be an innovative probiotic drink with health benefits for humans.

2 Materials and Methods

2.1 Materials

The coconut water probiotic drinks administered ingredients including young coconut water, MRS (De Man Ragosa and Sharpe (MRS) agar), and MRS broth, *Lactobacillus casei FNCC 0040* oblique culture obtained from the Microbiology Laboratory, Center for Food and Nutrition Studies, Universitas Gadjah Mada (UGM), skimmed milk, brown sugar (Javanese sugar), and granulated sugar. Materials for total acid analysis were 0.10 N NaOH and Phenolphthalein (PP) indicators. The pH test applied a buffer of 4.00 and a pH of 7.00, while the antioxidant activity test applied 1.1 – diphenyl – 2 picrylhydrazyl (DPPH) solution, aluminum foil, and methanol. The equipment employed in this study were incubators, glassware, analytical balances, and equipment for analysis, such as pH meters and a UV-Visible spectrophotometer.

2.2 Preparation of Bacterial Culture Starter

Preparing a ready-to-use liquid starter of *Lactobacillus casei FNCC 0040* was initiated by taking one ose of the oblique culture of *Lactobacillus casei FNCC 0040* bacteria into five ml of sterile MRS broth. Afterward, it was incubated for 17 h at 37 °C to obtain a broth culture of *Lactobacillus casei.* The broth culture was taken relatively 10% from 50 ml of the working medium and mixed into the active medium of young coconut water. The subsequent stage is incubation for 12 h at 37 °C.

2.3 Development of Young Coconut Water

The young coconut water probiotic drink process making was initiated by preparing 800 mL, which is clean of impurities. The initial content of total dissolved solids (-Brix) and pH in young coconut water was 4.80% and 5.80, subsequently. All contents transferred the young coconut water into each sterile glass beaker at as much as 100 ml. Therefore, coconut water was mixed with adding 0%, 5%, 10%, and 15% brown sugar and 3% skim milk related to the volume of young coconut water, followed by stirring until reaching homogeneous and pasteurized for ± 10 min at a temperature of 70 °C. After the mixture reached a temperature of 31–35 °C, it fermented at 37 ˚C for 48 h.

2.4 Total Acid

The titrimetric methods analyzed the total acid test of young coconut water probiotic drinks. Sample was added three drops of PP indicator to 10 ml of sample. The probiotic drinks were titrated with NaOH solution until a pink color was formed and did not change again for 30 s. The amount of NaOH applied at the end of the titration is calculated. Total acid obtained value in the form of percent total acid calculated by Eq. (1).

$$Total\ Acid = \left(\frac{V_1 \times N \times B \times 100\%}{V_2 \times 1000}\right) \times 100 \quad (1)$$

In addition, V_2 is the sample volume (ml), V_1 is the volume of NaOH (ml), B is the molecular weight of lactic acid (90), and N is the normality of NaOH (0.1N).

2.5 pH Measurement

The pH value of young coconut water probiotic drinks was measured using a pH meter [23] with slight modifications. Before measurement, the pH meter is standardized with pH 4.00 and 7.00 buffers at 25 °C. The pH value was measured using a calibrated electrode and inserted into the young coconut water probiotic drink as a sample.

2.6 Antioxidant Activity

Antioxidant activity testing of young coconut water probiotic drinks was analyzed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) method [24] with a slight modification. DPPH solution is 0.10 mM in methanol, and 2.40 mL of this solution mixing with 1.60 mL of the sample in methanol. Solution maintained the reaction mixture in the dark and closed room for 30 min. The absorbance of the cross was measured spectrophotometrically with a wavelength of 517 nm. Furthermore, 1.6 mL of methanol was applied instead of the sample for the blank. The experiment was repeated twice at each concentration, and the inhibition percentage obtained was applied as the antioxidant activity value from Eq. (2) [25].

$$\%RSA - \left(\frac{As - Ab}{Ab}\right) \times 100 \quad (2)$$

Furthermore, As is the absorbance of the sample, and Ab is the absorbance from the blank.

2.7 Research Design

The research design employed in this study was a completely randomized design (CRD) method, which was composed of a single factor, namely the concentration of brown sugar, which consisted of four additional treatments of adding brown sugar of 0%, 5%, 10%, and 15%. Twice repeated the treatment. Accordingly, the repetition of the analysis was conducted twice.

2.8 Statistical Analysis

The results of total acid, pH value, and antioxidant activity were obtained from statistical analysis and the normality test applied Shapiro-Wilk. Total acid had a normal data distribution. Thus, the data were tested using One-Way ANOVA and evaluated for significant differences from each treatment using the LSD test with a significance (p-value) <0.05. The pH value and antioxidant activity had an abnormal data distribution. Accordingly, the data were tested using Kruskal Wallis and evaluated for significant differences from each treatment using the Dunnet T3 test with a significance (p-value) < 0.05. All data were analyzed using IBM SPSS Statistics 22.

3 Results and Discussion

3.1 Total Acid

Total acid analysis shows the content of lactic acid solution [26]. The normality test using Shapiro Wilk obtained a p-value of 0.448, indicating that the total acid data distribution was normal, and subsequently applied One-Way ANOVA test. The test results are in Table 1, with a p-value of 0.001. Based on the results, adding brown sugar with different concentrations significantly affects the total acid of the young coconut water fermented drink.

The results of the LSD test revealed that the addition of 0% and 5% brown sugar had a significant difference from the addition of 10% and 15% brown sugar. The addition of 10% brown sugar significantly differed from the addition of 15% brown sugar. Simultaneously, adding 5% and 0% brown sugar was not significantly different.

Table 1. Mean of Total Acid in Young Coconut Water Probiotic Drink with Brown Sugar Addition.

Brown Sugar Addition (%)	Mean of Total Acid ± SD	P Value
0	0.83 ± 0.14a	0.001
5	0.97 ± 0.05^{a}	
10	1.17 ± 0.10^{b}	
15	1.42 ± 0.09^{c}	

Note: different letters indicate significant differences between treatments

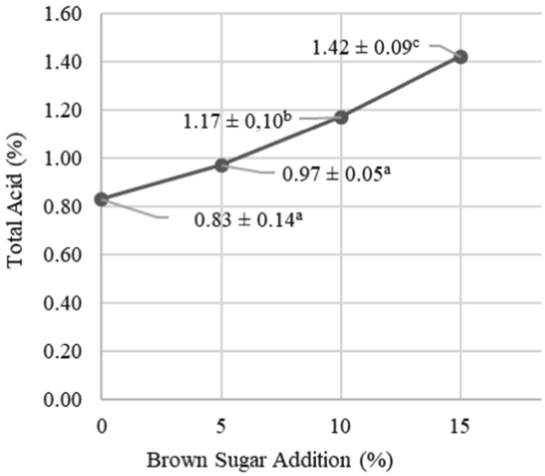

Fig. 1. Graph of Total Acid in Young Coconut Water Probiotic Drink with Brown Sugar Addition. Different Letters Show Significant Differences ($p < 0.05$).

After fermentation, the total acid value of the young coconut water probiotic drink increased from 0.83% to 1.42%. The highest total acid value was in the young coconut water probiotic drink at the addition of 15% brown sugar (1.43%), and the lowest was at the addition of 0% brown sugar (0.83%). The total acid content in coconut water probiotic drinks with brown sugar concentrations of 0%, 5%, 10%, and 15%, according to SNI 2981:2009 in yogurt, a type of probiotic drink, is between 0.50–2.00%. Figure 1 presented the magnitude of the change in the total acid of the probiotic drink. The total acid value in the probiotic drink increased in line with the brown sugar addition.

The total acid in this study is in line with previous research by Yanuar and Sutrisno regarding the production of young coconut water probiotic drinks with different concentrations of sucrose. It indicated that the higher the addition of sucrose, the higher the acid produced by fermented drinks, and the higher the total acid values ranging from 0.27% to 0.83% [11]. This increase in total acid was due to lactic acid bacteria, which hydrolyzed glucose and broke it down into pyruvic acid, broken down by the enzyme lactic dehydrogenase into lactic acid. Higher substrate availability causes lactic acid bacteria to utilize the substrate as a substance to increase and produce more lactic acid [27].

Lactobacillus casei bacteria, a starter in the production of probiotic drinks, is one of the bacteria that includes heterofermentative lactic acid bacteria that produce lactic acid. Therefore, the total acid contained in the product is a result of LAB fermentation. Lactic acid is also a fermented product of glucose in addition to CO2, acetic acid, and ethanol [28]. The availability of the substances required in the fermentation medium will increase the number of bacteria, resulting in the breakdown of sugar to a feasible extent [29]. Higher nutritional content in probiotic drinks will improve the growth of *Lactobacillus casei*. Bacterial activity in breaking down nutrients to be converted into lactic acid can increase the total acid value [30].

Table 2. Mean of pH in Young Coconut Water Probiotic Drink with Brown Sugar Addition.

Brown Sugar Addition (%)	Mean of pH Value ± SD	P Value
0	$3.78 \pm 0.32^{a, b}$	0.014
5	3.55 ± 0.06^{b}	
10	$3.38 \pm 0.10^{a, b}$	
15	3.35 ± 0.06^{a}	

Note: different letters indicate significant differences between treatments

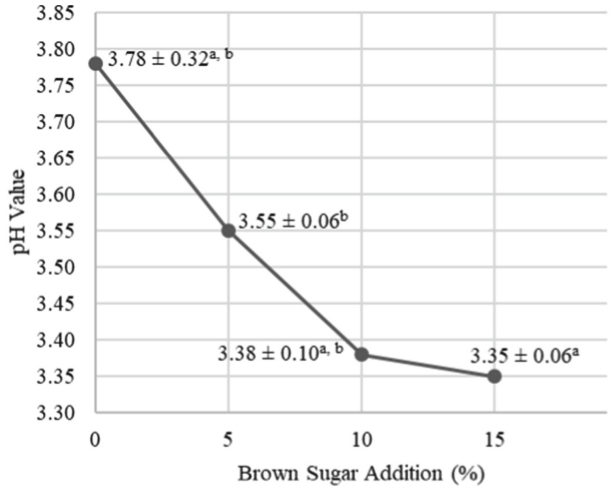

Fig. 2. Graph of pH Value in Young Coconut Water Probiotic Drink with Brown Sugar Addition. Different Letters Show Significant Differences ($p < 0.05$).

3.2 pH Value

The pH value is the level of acidity value in a product, indicating the level of wettability or acidity that the product has [31]. Therefore, the purpose of conducting a pH analysis of probiotic drinks is to find the pH value contained in fermented young coconut water drinks with brown sugar addition.

Using Shapiro Wilk, the tested pH value data for normality obtained a p-value of 0.002, indicating an abnormal pH data distribution. Afterward, it was tested statistically with the Kruskal-Wallis test. Table 2 shows the p-value (0.014), indicating that brown sugar addition significantly affects the pH value of the young coconut water fermented drink. The results of the Dunnett T3 test showed that the addition of 0% brown sugar had no significant difference from 5%, 10%, and 15% brown sugar addition. 10% brown sugar addition had no significant difference from 0%, 5%, and 15% brown sugar addition. However, between 5% and 15% brown sugar addition, there was a significant difference.

After fermentation, the pH value obtained from the pH test results increased from 3.35 to 3.75. The pH value of the young coconut water probiotic drink with the highest

Table 3. Mean of Antioxidant Activity in Young Coconut Water Probiotic Drink with Brown Sugar Addition.

Brown Sugar Addition (%)	Mean of Antioxidant Activity ± SD	P Value
0	16.41 ± 5.41^{a}	0.003
5	41.94 ± 7.58^{b}	
10	79.19 ± 2.04^{c}	
15	91.29 ± 3.54^{d}	

Note: different letters indicate significant differences between treatments

value was 0% brown sugar addition, at 3.75, and the lowest was the brown sugar with the addition of 15%, at 3.35. The pH value of young coconut water probiotic drinks added with brown sugar at concentrations of 0%, 5%, 10%, and 15% corresponds to the quality of fermented milk products, which are probiotic drinks with a suitable pH in the range of 3.00 – 4.60 [32]. Research conducted by Giri on the production of coconut water fermented drinks showed that the pH value of young coconut water fermented drinks using *Lactobacillus casei L4* after 48 h of fermentation was 3.20 [9].

Regarding the changes in the pH value of the product in Fig. 2, there was a decrease in the level of acidity along with brown sugar addition in the fermentation medium. Previous research from Yanuar and Sutrisno regarding the production of young coconut water probiotic drinks added to skim milk and a concentration of different types and amounts of sugar showed that the higher the sucrose concentration, the more decreased pH value, and a pH value of around 3.83 to 4.10 is obtained [11]. Another study from Ziska Taufik and Supriadi about probiotic drinks made from fermented coconut water added a variety of sucrose addition showed lower pH results as sugar addition increased [33].

The production of lactic acid by lactic acid bacteria in the fermentation process results in a decrease in pH [34]. The decreased pH value is directly proportional to the total acid, which has increased [33]. Increased total acid levels will cause a decrease in the level of acidity of probiotic drinks. The reduction in pH value can be affected by the availability of sucrose in the medium. The higher the sugar contained in the product, the more LAB (Lactic Acid Bacteria) increased. Thus, the amount of lactic acid would increase. These bacteria produce lactic acid, which causes the pH to decrease [35]. In the alteration of lactic acid by LAB, the release of H^+ ions occurs. This accumulated acid produces H^+ and $CH_3CHOHCOO$- ions. If there are more H^+ ions, the level of acidity of the solution will also be lower [30].

3.3 Antioxidant Activity

Free radical inhibition activity is essential to prevent various diseases, including cancer. The normality test for antioxidant activity data using Shapiro-Wilk obtained a p-value of 0.035, indicating that the antioxidant activity distribution was not normal. The analysis results using Kruskal-Wallis in Table 3 obtained a p-value of 0.003, which stated that adding brown sugar significantly affected the antioxidant activity of the probiotic drink

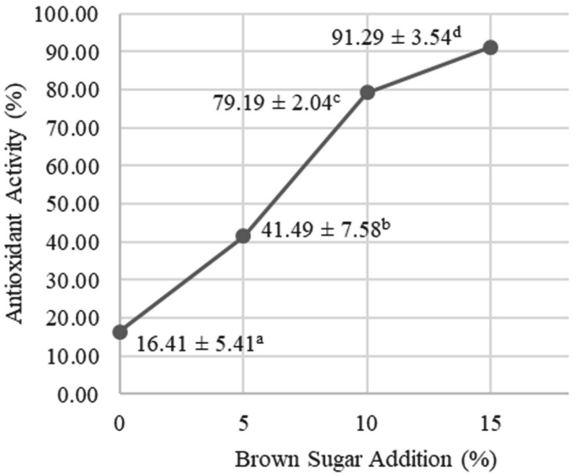

Fig. 3. Graph of Antioxidant Activity in Young Coconut Water Probiotic Drink with Brown Sugar Addition. Different Letters Show Significant Differences ($p < 0.05$).

fermented from young coconut water with brown sugar addition. The results of the Dunnett T3 test showed that there was a significant difference between each treatment with brown sugar additions of 0%, 5%, 10%, and 15%.

The results of the antioxidant activity analysis after fermentation increased from 16.41% to 91.29%. The highest value found for antioxidant activity in the young coconut water probiotic drink was with the addition of 15% brown sugar, at 91.29%, and the lowest was with the addition of 0% brown sugar, at 16.41%. The changes in the value of antioxidant activity in the product are shown in Fig. 3. There was an increase in radical inhibitory activity with the addition of brown sugar. The increase in antioxidant activity is directly proportional to the amount of lactic acid [33]. If the acid level is higher, the ability to inhibit free radicals will also be higher [38]. $CH_3CHOHCOOH$, or lactic acid, which is the product of lactic acid bacteria, has the function of contributing protons to radical molecules [39].

This finding is in line with previous research by Ziska, Taufik, and Supriadi regarding drinks made from fermented young coconut water with variations in various amounts of sugar added. The fermentation process and sugar addition could increase probiotic drinks' antioxidant activity [33]**.** In contrast to the research conducted by Nurhartadi et al. regarding the effect of incubation and sugar concentration on the characteristics of a probiotic drink produced by cheese in the form of whey, which showed that the higher the addition of sucrose, the more the antioxidant activity decreased due to vitamin C produced by *Bifidobacteria.* L-dehydroascorbate, which has active properties such as vitamin C, can be further changed to L-diketogulonic acid, which does not have active properties such as vitamin C [40].

DPPH produces a violet/purple color in methanol solutions and fades to a yellow color, which indicates the presence of antioxidants [24]. The stronger the radical inhibitory activity, the more the purple color decays in the solution [39]. The intensity of the color in the reduced DPPH solution indicates a reaction between the hydrogen

released by the test material and the DPPH radical molecule with the hydrogen released by the test material to form a yellow 1.1-diphenyl-2-picrylhydrazine compound [41]. Young coconut water contains active ingredients that can act as antioxidants, such as vitamin C, L-Arginine, and several minerals (Cu, Zn. Mn) [42].

The availability of sucrose and fermentation time can cause the pH of the product to be low. The low pH value will cause an increase in antioxidant activity [43]. During fermentation, the production of lactic acid will increase. The formation of lactic acid also results in increased antioxidant activity. In lactic acid probiotic drinks, AHAs (α-hydroxy acids) contain antioxidants [39]. In addition to the increase in lactic acid, there are secondary metabolites in the metabolism of LAB, which cause increased radical inhibitory activity. Lactic acid bacteria can produce vitamin E and vitamin C as forms of antioxidant compounds [38].

4 Conclusion

The conclusion from this study results is that adding brown sugar to the probiotic drink of young coconut water can increase total acids and antioxidant activity and significantly affect total acids and antioxidant activity. The higher the addition of brown sugar, the lower the pH value, significantly affecting pH. The most elevated total acid and antioxidant activity was 15% brown sugar addition, and the lowest was 10%. The highest pH value was at 0% brown sugar addition, and the lowest was at 15%. Young coconut water probiotic drinks in all treatments of brown sugar addition correspond to a suitable pH value for probiotic drinks, which is at a minimum of 3.00. Total acid following SNI 2981:2009 for probiotic yogurt drinks is at a maximum of 2.00%. In addition, it has a radical inhibiting activity which increases with brown sugar addition. Therefore, young coconut water and brown sugar addition are innovative probiotic drinks with human health benefits.

Acknowledgment. The authors express gratitude for this research's support from the Department of Nutrition Science, Faculty of Health Sciences, Universitas Muhammadiyah Surakarta.

Authors' Contributions. All authors contributed to this research, methodology, and writing.

References

1. F. C. Prado, J. D. D. Lindner, J. Inaba, V. Thomaz-Soccol, S. K. Brar, and C. R. Soccol, "Development and evaluation of a fermented coconut water beverage with potential health benefits," *J. Funct. Foods*, vol. 12, pp. 489–497, 2015, doi: https://doi.org/10.1016/j.jff.2014.12.020
2. E. Mela, Mustaufik, A. Maksum, N. G. Tbet, "Diversifikasi Produk Pangan Berbasis Air Kelapa," *AGRITECH*, vol. 22, no. 2, 2020
3. G. Zhang, W. Chen, W. Chen, and H. Chen, "Improving the quality of matured coconut (*Cocos nucifera Linn.*) water by low alcoholic fermentation with *Saccharomyces cerevisiae*: antioxidant and volatile profiles," *J. Food Sci. Technol.*, vol. 55, no. 3, pp. 964–976, 2018, doi: https://doi.org/10.1007/s13197-017-3004-y

4. J. W. Yong, L. Ge, Y. F. Ng, S. N. Tan, "The chemical composition and biological properties of coconut water (*Cocos nucifera L.*)," *Molecules*, vol. 14, no. 12, pp. 5144–5164, 2009
5. P. Utami, *Buku Pintar Tanaman Obat*. Jakarta: PT. Agromedia Pustaka, 2008
6. M. Z. Anwar, "Pembuatan Minuman Probiotik Air Kelapa Muda (*Cocos nuciferra L*) Dengan Starter *Lactobacillus Casei Subsp. Casei R-68*," *JOM Faperta*, vol. 5, no. 1, 2018
7. A. Manivannan, R. Bhardwaj, S. Padmanabhan, P. Suneja, K. B. Hebbar, S. R. Kanade, "Biochemical and nutritional characterization of coconut (*Cocos nucifera L.*) haustorium," *Food Chem.*, vol. 238, pp. 153–159, 2018. doi: https://doi.org/10.1016/j.foodchem.2016.10.127
8. F. Camargo Prado, J. De Dea Lindner, J. Inaba, V. Thomaz-Soccol, S. Kaur Brar, C. R. Soccol, "Development and evaluation of a fermented coconut water beverage with potential health benefits," *J. Funct. Foods*, vol. 12, pp. 489–497, 2015. doi: https://doi.org/10.1016/j.jff.2014.12.020
9. S. S. Giri, V. Sukumaran, S. S. Sen, S. C. Park, "Use of a potential probiotic, *Lactobacillus casei L4,* in the preparation of fermented coconut water beverage," *Front. Microbiol.*, vol. 9, no. AUG, pp. 1–9, 2018. doi: https://doi.org/10.3389/fmicb.2018.01976
10. A. Singh, A. Bhardwaj, V. Sharma, "Fermentation of Tender Coconut Water by Probiotic Bacteria *Bacillus coagulans*," *Int. J. Food Stud.*, vol. 7, pp. 100–110, 2018
11. S. E. Yanuar and A. Sutrisno, "Minuman Probiotik dari Air Kelapa Muda dengan Starter Bakteri Asam Laktat *Lactobacillus casei*," *J. Pangan dan Agroindustri*, vol. 3, no. 3, pp. 909–917, 2015
12. I. A. P. Pranayanti and A. Sutrisno, "Pembuatan Minuman Probiotik Air Kelapa Muda (Cocos nucifera L.) dengan Starter *Lactobacillus casei* strain Shirota," *J. Pangan dan Agroindustri*, vol. 3, no. 2, pp. 763–772, 2015
13. A. M. Tangapo and S. M. Mambu, "Edukasi Mengenai Pentingnya Konsumsi Probiotik Untuk Peningkatan Kesehatan Pada Kelompok Wanita di Kelurahan Banjer Kecamatan Tikala Kota Manado," *VIVABIO J. Pengabdi. Multidisiplin*, vol. 1, no. 3, pp. 13–17, 2019. doi: https://doi.org/10.35799/vivabio.1.3.2019.26723
14. C. Hill *et al.*, "Expert consensus document: The international scientific association for probiotics and prebiotics consensus statement on the scope and appropriate use of the term probiotic," *Nat. Rev. Gastroenterol. Hepatol.*, vol. 11, no. 8, pp. 506–514, 2014. doi: https://doi.org/10.1038/nrgastro.2014.66
15. P. R. Lee, C. X. Boo, and S. Q. Liu, "Fermentation of coconut water by probiotic strains *Lactobacillus acidophilus L10 and Lactobacillus casei L26*," *Ann. Microbiol.*, vol. 63, no. 4, pp. 1441–1450, 2013. doi: https://doi.org/10.1007/s13213-013-0607-z
16. M. I. Watawana, N. Jayawardena, C. B. Gunawardhana, and V. Y. Waisundara, "Enhancement of the antioxidant and starch hydrolase inhibitory activities of king coconut water (*Cocos nucifera var. aurantiaca*) by fermentation with kombucha 'tea fungus,'" *Int. J. Food Sci. Technol.*, vol. 51, no. 2, pp. 490–498, 2016. doi: https://doi.org/10.1111/ijfs.13006
17. F. Nurainy, S. Rizal, S. Suharyono, and E. Umami, "Karakteristik Minuman Probiotik Jambu Biji (*Psidium guajava*) pada Berbagai Variasi Penambahan Sukrosa dan Susu Skim," *J. Apl. Teknol. Pangan*, vol. 7, no. 2, pp. 47–54, 2018. doi: https://doi.org/10.17728/jatp.2510
18. Widodo *et al.*, "Bakteri Asam Laktat Strain Lokal: Isolasi sampai Aplikasi sebagai Probiotik dan Starter Fermentasi Susu." Gadjah Mada University Press, Yogyakarta, 2017.
19. E. Bandalan and L. Galvez, "Optimization of Coconut Water Beverage Fermented with *Lactobacillus acidophilus*," *Ann. Trop. Res.*, vol. 202, pp. 196–202, 2016. doi: https://doi.org/10.32945/atr38117.2016
20. T. A. Akbar, "Analisa Kadar Sakarin Pada Minuman Yoghurt." Universitas Muhammadiyah Surabaya, 2012
21. L. J. Utama, A. C. Sembiring, and A. Nur, *Konsep Dasar Ekonomi Pangan Dan Gizi*. Bandung: Media Sains Indonesia, 2021

22. N. D. Putriningtyas and S. Wahyuningsih, "Potensi yogurt kacang merah (Phaseolus vulgaris L) ditinjau dari sifat organoleptik, kandungan protein, lemak dan flavonoid," *J. Gizi Indones. (The Indones. J. Nutr.*, vol. 6, no. 1, pp. 37–43, 2017, doi: https://doi.org/10.14710/jgi.6.1.37-43
23. A. Sofyan, A. Y. Ikhsani, E. Purwani, L. E. N. Hasanah, and F. Febriyadin, "The effect of suweg (*Amorphophallus paeoniifolius*) flour and incubation temperature on characteristics of yogurt with the addition of Bifidobacterium bifidum as probiotic," *Mater. Today Proc.*, vol. 63, pp. S507–S512, 2022. doi: https://doi.org/10.1016/j.matpr.2022.04.538
24. M. M. Rahman, M. B. Islam, M. Biswas, and A. H. M. Khurshid Alam, "In vitro antioxidant and free radical scavenging activity of different parts of Tabebuia pallida growing in Bangladesh," *BMC Res. Notes*, vol. 8, no. 1, pp. 1–9, 2015. doi: https://doi.org/10.1186/s13104-015-1618-6.
25. A. L. Freire, C. L. Ramos, P. N. da Costa Souza, M. G. B. Cardoso, and R. F. Schwan, "Nondairy beverage produced by controlled fermentation with potential probiotic starter cultures of lactic acid bacteria and yeast," *Int. J. Food Microbiol.*, vol. 248, pp. 39–46, 2017. doi: https://doi.org/10.1016/j.ijfoodmicro.2017.02.011
26. F. Jaya, *Ilmu, Teknologi, dan Manfaat Kefir*. Malang: Universitas Brawijaya Press, 2019.
27. Y. Yunus and E. Zubaidah, "Pengaruh konsentrasi sukrosa dan lama fermentasi terhadap viabilitas *L. casei* selama penyimpanan beku velva pisang ambon," *J. Pangan dan Agroindustri*, vol. 3, no. 2, pp. 303–312, 2015
28. I. W. A. Widyantara, K. A. Nocianitri, and N. M. I. H. Arihantana, "Pengaruh Lama Fermentasi Terhadap Karakteristik Minuman Probiotik Sari Buah Sirsak (*Annona muricata Linn*)," *J. Ilmu dan Teknol. Pangan*, vol. 9, no. 2, pp. 151–160, 2020. doi: https://doi.org/10.24843/itepa.2020.v09.i02.p05
29. A. Triyono, "Mempelajari Pengaruh Maltodekstrin dan Susu Skim Terhadap Karakteristik Yoghurt Kacang Hijau (*Phaseoulus radiatus L.*)," *Semin. REKAYASA Kim. DAN PROSES*, 2010
30. K. Khotimah and J. Kusnadi, "Antibacterial Activity of Probiotic Date Fruit (Phoenix dactilyfera L.) Beverages Using *Lactobacillus plantarum* and *Lactobacillus casei*," *J. Pangan dan Agroindustri*, vol. 2, no. 3, pp. 110–120, 2014
31. R. A. Sultan, Lahming, and A. Sukainah, "Karakteristik Minuman Probiotik Kombinasi Sari Buah Nenas (*Ananas comosus L.*) dan Pepaya (carica papaya L.)," *Pap. Knowl. . Towar. a Media Hist. Doc.*, vol. 3, no. April, pp. 49–58, 2015
32. Susrini, *Pengantar Teknologi Pengolahan Susu*. Malang: Universitas Brawijaya, 2003
33. R. Ziska, A. Taufik, and D. Supriadi, "Uji Aktivitas Antimikroba Dan Antioksidan Dari Minuman Probiotik Hasil Fermentasi Air Kelapa (*Cocos nucifera)*," *J. Farm. Galen.*, vol. 4, no. 1, pp. 14–19, 2017
34. E. K. Wardani, S. Zulaekah, and E. Purwani, "Pengaruh Penambahan Sari Buah Nanas (Ananas Comosus) terhadap Jumlah Bakteri Asam Laktat (BAL) dan Nilai pH Soyghurt," *J. Kesehat.*, vol. 10, no. 1, p. 68, 2017. doi: https://doi.org/10.23917/jurkes.v10i1.5494
35. M. F. Sari and R. H. Catarina, "Perbandingan Karakteristik Minuman Probiotik Semangka (Citrullus lanatus) Dengan Variasi Jenis Semangka Merah Dan Kuning Menggunakan Starter *Lactobacillus casei* Strain Shirota," *Biota J. Ilm. Ilmu-Ilmu Hayati*, vol. 5, no. November 2019, pp. 25–33, 2020. doi: https://doi.org/10.24002/biota.v5i1.2945
36. M. Abountiolas and C. do Nascimento Nunes, "Polyphenols, ascorbic acid and antioxidant capacity of commercial nutritional drinks, fruit juices, smoothies and teas," *Int. J. Food Sci. Technol.*, vol. 53, no. 1, pp. 188–198, 2018. doi: https://doi.org/10.1111/ijfs.13573
37. R. K. Wardani, S. Subariyatun, S. W. Azhari, and A. Sofyan, "Functional Properties of Instant Yellow Rice of Gembili Tubers (Dioscorea esculenta) to Improve Food Security," pp. 25–33, 2021

38. E. P. Oktaviani, L. M. E. Purwijantiningsih, and F. S. Pranat, "Kualitas dan Aktivitas Antioksidan Minuman Probiotik Dengan Variasi Ekstrak Buah Naga Merah (*Hyloreceus polyrhizus*)," *J. Teknobiologi*, vol. 1, no. 1, pp. 1–15, 2014
39. A. P. Kusumaningrum, "Kajian Total Bakteri Probiotik Dan Aktivitas Antioksidan Yoghurt Tempe," Universitas Sebelas Maret, 2011
40. E. Nurhartadi, A. Nursiwi, R. Utami, and E. Widayani, "Pengaruh waktu inkubasi dan konsentrasi sukrosa terhadap karakteristik minuman probiotik dari whey hasil samping keju," *J. Teknol. Has. Pertan.*, vol. 9, no. 2, pp. 73–83, 2018
41. D. Andriani and L. Murtisiwi, "Uji Aktivitas Antioksidan Ekstrak Etanol 70% Bunga Telang (*Clitoria ternatea L*) dari Daerah Sleman dengan Metode DPPH," *Pharmacon J. Farm. Indones.*, vol. 17, no. 1, pp. 70–76, 2020. doi: https://doi.org/10.23917/pharmacon.v17i1.93211
42. D. R. Pramana, "Pengaruh Air Kelapa Muda (*Cocos nucifera L.*) Terhadap Kadar Hematokrit Darah Studi Eksperimental pada Tikus Jantan Galur Wistar (*Rattus norvegicus)* yang diinduksi Plumbum (Pb)." Universitas Islam Sultan Agung, 2019
43. D. I. Kartikasari and F. C. Nisa, "Pengaruh Penambahan Sari Buah Sirsak dan Lama Fermentasi Terhadap Karakteristik Fisik dan KImia Yoghurt," *J. Pangan dan Agroindustri* vol. 2, no. 4, pp. 239–248, 2014