Original Article

JMPI p-ISSN : 2442-6032 e-ISSN : 2598-9979

DOI : 10.35311/jmpi.v10i1.519

Characteristics of Four Variants of Kombucha Tea as Candidate of Functional Health Drink

Nooryza Martihandini*, Imat Rochimat

Departement of Pharmacy, Politeknik Kesehatan Kemenkes Tasikmalaya

Citation: Martihandini, N., & Rochimat, I. (2024). Characteristics of Four Variants of Kombucha Tea as Candidate of Functional Health Drink. *Jurnal Mandala Pharmacon Indonesia,* 10(1), 227-237. https://doi.org/10.35311/jmpi.v10i1.519

Submitted: 26 April 2024
Accepted: 12 Juni 2024
Published: 30 Juni 2024

*Corresponding Author:
Nooryza Martihandini
Email: nooryzamartihandini@gmail.com

ABSTRAK

Daun teh diketahui memiliki kandungan senyawa fenolik tinggi yang dapat menetralkan radikal bebas penyebab berbagai penyakit degeneratif. Proses fermentasi infusa daun teh dengan SCOBY (*Symbiotic Culture of Bacteria and Yeast*) akan menghasilkan minuman kombucha dengan aktivitas antioksidan yang lebih tinggi. Penelitian ini bertujuan untuk membuat dan mengevaluasi minuman teh kombucha. Metode penelitian menggunakan desain eksperimental laboratorium. Minuman kombucha disiapkan dari 4 varian teh berbeda yaitu teh hitam, teh hijau, teh putih dan teh oolong. Masing-masing infusa teh ditambahkan sukrosa dan difermentasi selama 14 hari dengan kultur SCOBY. Kombucha dievaluasi karakteristiknya meliputi organoleptic, aktivitas antioksidan, uji kandungan total senyawa fenolik, total asam, total gula, dan nilai pH. Hasil penelitian menunjukkan bahwavarian teh yang digunakan mempengaruhi karakteristik kombucha yang dihasilkan. Kombucha teh hitam memiliki aktivitas antioksidan paling tinggi dengan nilai persentase penghambatan radikal sebesar 82.38 ± 0.08%.Kadar fenolat total dari teh kombucha berada pada rentang 0,040 – 0,082%. Proses fermentasi menghasilkan asam organik dan mengurangi kandungan gula dalam kombucha. Berdasarkan hasil penelitian ini dapat disimpulkan bahwa kombucha memiliki aktivitas menangkal radikal bebas sehingga memiliki potensi untuk dikembangkan sebagai minuman fungsional kesehatan.

Kata Kunci : Antioksidan, Karakteristik, Fermentasi, Teh Kombucha

ABSTRACT

Tea leaves contain high levels of phenolic compounds which can neutralize free radicals causing various degenerative diseases. The fermentation process of tea leaf infusion using SCOBY (Symbiotic Culture of Bacteria and Yeast) produce a kombucha drink having higher antioxidant activity. This study aimed to produce and evaluate kombucha drinks from four types of tea leaves The research method used a laboratory experimental design. Kombucha drinks were prepared from 4 different tea variants, namely black tea, green tea, white tea and oolong tea. Each tea infusion was added with sucrose and fermented for 14 days with SCOBY culture. Then, kombuchas were evaluated for its characteristics in term of organoleptics, antioxidant activity, total phenolic compound content, total acid, total sugar, and the pH value. The results showed that the variant of tea influenced characteristics of kombucha. The black tea kombucha had the highest antioxidant activity with the percentage of radical inhibition of 82.38 ± 0.08%. The total phenolic contents of tea kombuchas were in the range of 0.040 –0.082 %. Fermentation process produced organic acid and reduced sugar content in all kombuchas. It can be concluded that kombuchas showed the radical scavenging activity hence it can potentially developed as functional health drink.

Keywords : Antioxidant, Characteristics, Fermentation, Kombucha Tea

INTRODUCTION

Unhealthy lifestyles, stress and environmental pollution lead the excessive synthesis of reactive oxygen species in the body. Disruption of homeostasis caused by free radicals triggers oxidative stress and damages the structure of human organs. This process initiates the degenerative diseases. In order to maintain a balance between the production and elimination of reactive oxygen species, we need antioxidant intake from daily food. One of the main antioxidants widely distributed in dietary intake is phenolic compounds. This compounds have been known for their benefits in slowing the aging process and reducing the risk of degenerative diseases (Jakubczyk *et al*., 2020).

Tea leaves have high level of polyphenol compounds. This natural ingredients contain flavan-3-ol group of polyphenols, mainly catechin, epigalocatechin and epigalocatechin-3-gallate. Several studies proved that the polyphenol and catechin content of tea leaves had antioxidant activitygenerating various pharmacological effects such as anticancer, anti-inflammatory, antidiabetic, antihypertensive, antiobesity, antiplatelet, anticholesterol, and neuroprotective (Zanwar et al., 2013).

Tea is a popular drink consumed by more than two thirds of the world's population with various types, such as black tea (78%), green tea (20%) and oolong tea (2%). Generally, tea drink is made by brewing using hot water(Gaur & Agnihotri, 2014; Massoud et al., 2022).Tea derived from *Camellia sinensis* leaves can be categorized into 4 types based on the degree of fermentation, namely black tea, green tea, white tea and oolong tea. Green tea and white tea do not undergo fermentation, black tea undergoes fermentation, and oolong tea undergoes a partial fermentation process. Every types of tea have different appearances, aromas, flavors, and colors (Namal Senanayake, 2013).

Green tea has a significantly higher polyphenol content compared to black and oolong teas. The component of green tea is mainly dominated by catechins with a composition of 25-35% of the dry weight. All tea variants contain epicatechin, epigalocatechin, epicatechin gallate and epigalocatechin gallate. The primary catechin compounds in black tea are epicatechin gallate and epigalocatechin gallate, while in green and oolong tea is epigalocatechin gallate (Namal Senanayake, 2013). The presence of hydroxyl groups and aromatic rings of polyphenol compounds can bind and neutralize oxygen free radicals in the body such as superoxide anions, peroxide radicals and singlet oxygen causing various degenerative diseases (Değirmencioğlu et al., 2021).

Currently, consumer trend is towards a healthy lifestylemarked by the increasing of consumer awareness and interest in health and functional foods. Several factors increase the consumer demand for functional foods such asthe increasing of income, the life expectancy, the level of adoption of healthy lifestyles, and thtechnological advances(Pratiwi, 2018). Kombucha, a fermented tea drink, has known as functional drink due to the high content of beneficial compounds. Kombucha is obtained using a symbiotic culture of bacteria and yeast. This drink is prepared by combining tea, sugar, and SCOBY and then fermented at room temperature for 7-14 days. Previous study showed that fermentation process increased the phenolic content of tea so that the antioxidant activity of kombucha was higher than unfermented tea drinks (Massoud et al., 2022). The chemical composition of kombucha varies depending on the type of tea and fermentation conditions. SCOBY use tea leaves and sugar as a source of nitrogen (caffeine, theanine, and theophylline) and a source of carbohydrates. The phenolic compounds, organic acids (especially glucuronic, acetic, gluconic, lactic acid), vitamins, micronutrients and cell metabolites contributed for the functional properties of kombucha (Değirmencioğlu et al., 2021; Massoud et al., 2022). Those components make kombucha tea as a functional drink potentially improving health and reducing the risk of degenerative diseases (Linnarto et al., 2019).

This research aimed to develop a kombucha tea drink from black tea, green tea, white tea and oolong tea substrates. The secondary metabolites of tea kombucha were evaluated qualitatively using phytochemical screening. Then, the kombucha was evaluated to determine the characteristic of each type kombucha in term ofantioxidant activity, total phenolic content, total acid and total sugar. The evaluation also aims to explore the potential of each type of kombucha as a functional drink.

RESEARCH METHODS

Materials

Four tea variants consisting of black tea, green tea, white tea and oolong tea were obtained from East Java & Co. The starter and SCOBY culture were purchased from Rumah Fermentasi. Other materials were sucrose (Rose Brand), Folin-Ciocalteu reagent (Sigma Aldrich),

DPPH (Sigma Aldrich), gallic acid (Sigma Aldrich), NaOH (Merck), FeCl3 (Sigma aldrich), chloroform (Merck) methanol (Emsure), wagner reagent (DPH), anhydrous acetic acid (Isolab), sulfuric acid P (Emsure). The tools used in this study were UV-Vis spectrophotometer (Agilent Cary 60), ph Meter (Hanna), and analytical balance (Sartorius).

Manufacturing of Tea Kombucha

Tea leaves infusion were prepared from black tea, green tea, white tea and oolong tea. One hundred grams of sugar were mixed with 8 grams of tea and 1 L of hot water (90°C) for 10 minutes in a sterile flask. After cooling (30°C), the tea infusion was filtered and put into a sterile glass bottle. The tea infusion was added with 10% SCOBY culture under aseptic conditions. Then the bottle was covered with a clean cloth and tied tightly. Fermentation was carried out by incubating the kombucha culture at room temperature for 14 days. After 14 days, the obtained kombucha was filtered and analyzed.

Phytochemical Screening

1. Flavonoid

Samples were added 5 ml of methanol and filtered. Then two drops of concentrated HCl and Magnesium powder were poured into the mixture. The mixture was shaken vigorously. The presence of flavonoids is figured out by the formation of a dark orange color.

2. Saponin

A total of 2 grams of sample was extracted with 5 mL aquadest and filtered using cotton. The filtrate was shaken vigorously for 2 minutes. Then, two drops of HCl 2 N was added. The sample containing saponin are characterized by the formation of stable foam with high intensity for 10 minutes.

3. Polyphenol

Two grams of samples were added with methanol. The filtered solution was transferred into a new test tube, then it mixed with solution of FeCl3 5%. The presence of polyphenols is indicated by the formation of a bluish black or green color.

4. Alkaloid

The samples in a test tube were extracted with ammoniacal choroform. Then the samples were added with Wagner reagent. A positive result is indicated by the presence of a bwonish precipitate.

5. Steroid and Triterpenoid

The sample was extracted with 5 mL ethanol. Then, it was filtered and heated to form dry extract. After that, it was extracted with the mixture of kloroform-aquadest (1:1). The obtained extract was added with one drop of concentrated H_2SO_4 and one drop anhydrous acetic acid. Extracts containing steroids are indicated by a color change to green or purple. A positive results for triterpenoid is indicated by the formation of red or brown color.

The Total Phenol Content Assay

The total phenolic content of kombucha was determined by the Folin-Ciocalteu method. Samples were diluted with methanol and centrifuged at 6000 rpm for 5 minutes. The obtained supernatant was mixed with Folin-Ciocalteu reagent and solution of Na_2CO_3 7,5%. The mixture was incubated at 45°C for 15 minutes. Absorbance of samples was measured at a wavelength of 765 nm using UV-Visual spectrophotometry.

The Antioxidant Activity Assay

The antioxidant activity of tea kombucha was measured by a spectrophorometric method using DPPH radicals (2,2-Diphenyl-1Pikrylhydrazyl). Absorbance was measured at 515 nm. The measurement results were expressed as % DPPH inhibition.The sample was diluted into 5 variations of graded concentrations using methanol. Then, the diluted sample were added 1 mL DPPH solution. The solutions were homogenized and incubated at 37°C for 30 minutes. The absorbances of the solution were determined using a spectrophotometer at a wavelength of 515 nm. After that, the inhibition ability of each sample concentration (% inhibition) was calculated using followed equation:

% Inhibition :

$$\frac{A_{control} - A_{sample}}{A_{control}} \times 100\%$$

Determination of Total Acid Content

A sample of tea kombucha tea was weighed at 1 gram, then put into a 25 mL volumetric flask and methanol added to the tare limit. The solution was homogenized and 3 drops of PP indicator were added. After that, the sample was titrated with 0.1 N NaOH until the end point of the titration.

Determination of Total Sugar Content

A sample of tea kombucha was added with $CaCO_3$, ethanol and aquadest. The cooled mixture was added with saturated Pb acetate and then poured into measuring flask filled with dinatrium oxalate. Then the mixture was added with aquadest. The filtered mixture was poured into test tube and mixed with aquadest and anthrone reagent. The sample was homogenized and heated for 12 minutes. The absorbance of the cooled sample was measured using UV-Vis spectrophotometry at a wavelength of 630 nm.

RESULTS AND DISCUSSION

Kombucha was made using 4 different tea variants purchased from East Java and Co. The tea variants are classified based on the degree of fermentation, consisting of black tea, green tea, white tea and oolong tea. Green tea and white tea do not undergo fermentation, black tea undergoes fermentation, and oolong tea undergoes a partial fermentation process. The four types of tea have different appearances, aromas, flavors, and colors(Namal Senanayake, 2013). The majority (78%) of tea variant produced in the world is black tea. Meanwhile, green tea production is around 20%, which is widely consumed in Asia, North America and the Middle East. Less than 2% of oolong tea is produced and consumed majorly in Southern China and Taiwan(Gaur & Agnihotri, 2014).

Figure 1. Tea variants for manufacturing kombucha: A. black tea; B. green tea; C. White tea; D. oolong tea

Green tea is a rich natural source of polyphenols, especially epigallocatechin-3-gallate (EGCG). This tea is produced by drying and steaming fresh tea leaves to deactivate the enzyme polyphenol oxidase. Black tea contains tearubigin and teaflavin which are produced through the enzymatic oxidation of tea leaf polyphenols. Oolong tea is made through partial oxidation of tea leaves (Gaur & Agnihotri, 2014). White tea comes from young, curled tea leaves which are evaporated and dried to prevent oxidation. White tea contains higher levels of phenolic compounds, caffeine and theogaline (3-galloyquinic acid) than other teas (Piyasena et al., 2023).

The chemical contents of teas produce the differences in taste and flavour. Polyphenol, amio acids and caffeine develop the taste of tea. On the other hand, volatile compounds (terpenoids, alcohols, and carbonyl compounds) develop the aroma of tea. The higher contents of catechins and amino acids generate the higher astringency and bitternes. The taste of green tea is a mixture of bitternes, astringency and slight sournes. The oolong tea has a unique taste generated by non oxidized catechins, thearugibins, caffeine, amino acid and sugars. The oolong tea has stronger sweetness but lower astringency than green tea. The astringency taste of black tea produced

primarily by cathecins, theaflavins and flavonol glycosides (Namal Senanayake, 2013).

Kombucha is produced by fermenting the tea infusion containingsucrose with SCOBY culture under aerobic conditions. SCOBY culture is a symbiotic system between fungi and acetatic acid bacteria. The SCOBY culture composition depends on climatic and geographical conditions (Değirmencioğlu et al., 2021). This research used the SCOBY culture obtained from Rumah Fermentasi.

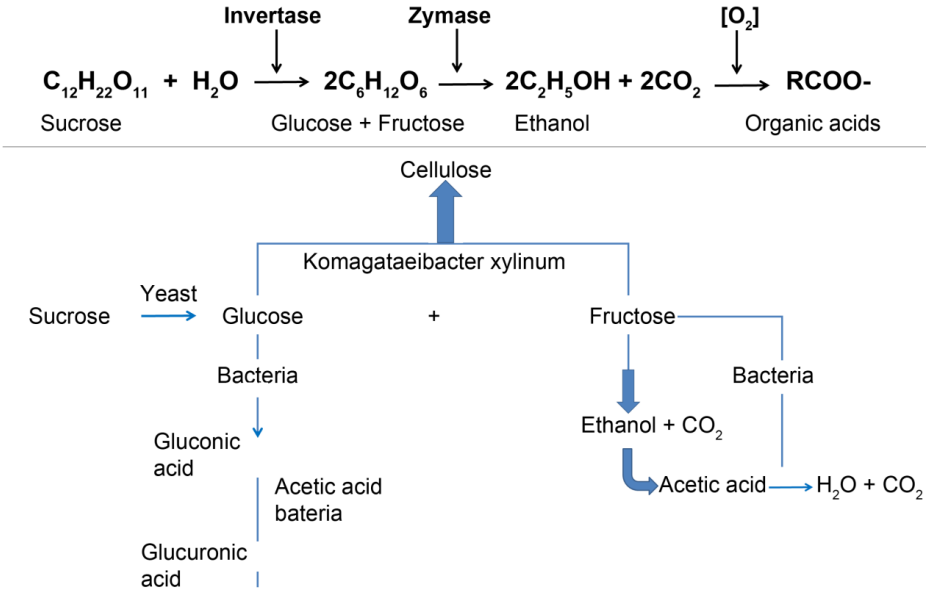

Figure 2. Fermentation route of kombucha

This study used tea infusion produced by brewing tea leaves at temperature of 90°C. Previous study proved that temperature range of 80° - 100° C and brewing time range 5 -10 minutes produced greater antioxidant activity due to the higher contents of bioactive compounds such as gallic acid, epigallocatechin and caffein(Pérez-Burillo et al., 2018; Sharpe et al., 2016). Similar result was obtained by Zimmermann and Gleichenhagena that brewing at 100°C produced the highest contents of flavanols(Zimmermann & Gleichenhagen, 2011).

Important substrates for producing kombucha are tea and sucrose. Sucrose is the major carbon source in the fermentation process by SCOBY culture. Hydrolysis reaction by the yeast invertase enzyme found in the SCOBY changes sucrose into glucose and fructose. Then the glucose is converted into ethanol and carbon dioxide by yeast. Ethanol is then converted into acetic acid by acetobacteria. The production of organic acids causes the kombucha drink to have a sour taste and low pH(Massoud et al., 2022). SCOBY also uses tea leaves as a source of nitrogen (caffeine, theanine, and theophylline).

All of the obtained kombuchas after fermentation for 14 days had the sour taste and lighter color than unfermented teawith different grade of brown color as presented on the Figure 3. The white tea kombucha was the lightest color among the other kombuchas. Futhermore, the oolong tea kombucha had the darkest color. The change of thearubigin into theaflavin during fermentation results in a change in the color of the kombucha from dark to lighter in line with the length of fermentation time. Fermentation also generated a new SCOBY biofilm layer on the surface of the drink separating from the parent SCOBY. The layer of the new SCOBY was thicker as the fermentation time increases. The layer had a dense polymeric structure composed of cellulose produced by strains such as *Komagataeibacter xylinus* or *Gluconacetobacter spp*. Other species that are part of the kombucha consortium can also produce cellulose biofilms such as *Aerobacter*, *Agrobacterium*, *Azotobacter* and *Rhizobium* (Villarreal-Soto et al., 2019). This cellulose

biofilm is produce from various carbon sources, including glucose, ethanol, sucrose, and glycerol by synthesis of uridine diphosphate-glucose (UDPGlc), which is the cellulose precursor (Coelho et al., 2020).

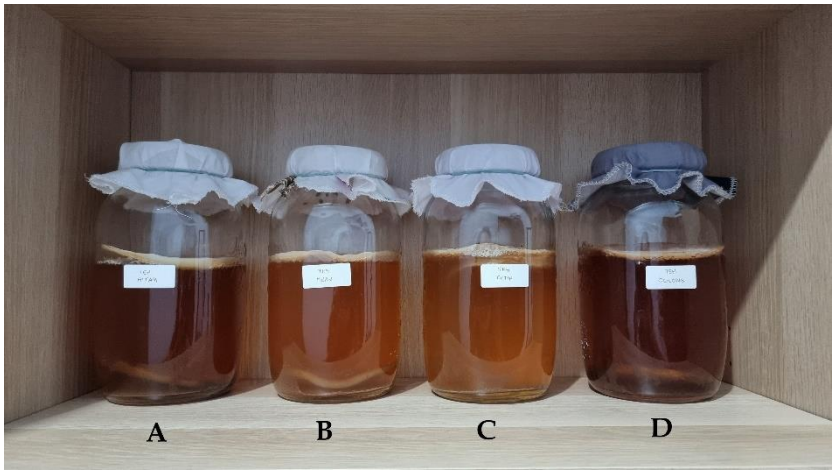

Figure 3. Kombucha from different tea variants: A. Black tea; B. Green tea; C. White tea; D. Oolong tea

Phytochemical screening was conducted to show the chemical compounds of kombucha qualitatively as illustrated on Table 1. Tea has various chemical compounds, such as polyphenols, minerals, vitamins, amino acids, methylxanthin, pigments, carbohydrates, organic acids, fats and volatile substances.Previous study showed that black tea, green tea and white tea contained phenolic compounds, flavonoid, saponin, terpenoid and steroid (Leslie & Gunawan, 2019).

Based on the phytochemical screening presented on Table 1, all kombucha variants contained polyphenol, flavonoid, and saponin.Various polyphenolic compounds are contained in tea leaves. The polyphenol components of tea act as antioxidants (Chakravorty et al., 2019). Flavanols (catechins) and flavonols are the primary secondary metabolites found in tea with concentration of around 30% of the dry weight of fresh tea leaves. Flavanols are the dominant phenolic compounds in tea consisting of (+) catechin, epigalocatechin-3-gallate (EGCG), epicatechin-3-gallate (ECG), epigalocatechin (EGC) and epicatechin (EC)(Piyasena et al., 2023).

Table 1.Phytochemical screening of kombuchas

No.	Secondary Metabolite	Black Tea Kombucha (BTK)	Green Tea Kombucha (GTK)	White Tea Kombucha (WTK)	Oolong Tea Kombucha (OTK)
1	Polyphenol	+	+	+	+
2	Flavonoid	+	+	+	+
3	Saponin	+	+	+	+
4	Steroid / Triterpenoid	+	-	-	-
5	Alkaloid	-	-	-	-

There is an increase of polyphenols including flavonoids during fermentation of kombucha tea. The increasing content of polyphenolic compounds of kombucha is related to various reactions during fermentation, such as oxidation of phenolic compounds by enzymes and hydrolysis reactions by microbes that trigger the

formation of catechins, flavonoids and other antioxidant compounds (Jakubczyk et al., 2020). The study found that the variant of tea influenced the total phenolic content of kombucha. Table 2 showsthe results of total phenolic assay discoveredthat white tea kombucha had the highest polyphenol content with the concentration of 0.082%.The order of the kombuchas from the highest total phenolic content was WTK > GTK > OTK > BTK.

Table 2. The total phenolic content of kombuchas

No.	Type of Kombucha	Total Phenolic Content (%)
1	BTK	0.040 ± 0.001%
2	GTK	0.080 ± 0.002%
3	WTK	0.082 ± 0.003%
4	OTK	0.059 ± 0.001%

The antioxidant activity of tea is mainly attributedby phenolic compounds. Phenolic compounds has antioxidant activity due to donating electrons to free radicals. The antioxidant activity of kombuchas was measured using DPPH method. DPPH is a stable free radical having intense purple color caused by the delocalization of electron. Phenolic compounds easily react with DPPH molecules by transferring electron or donating hydrogen atom. This reaction generate formation of hydrazine (DPPH-H), a reduced form of the DPPH molecules. Futhermore, the formation of hydrazine then lead the discoloration of the DPPH solution so that it can be measured by UV-visual spectrophotometry. Figure 4 shows the reaction of antioxidant compound with DPPH (Gulcin & Alwasel, 2023).

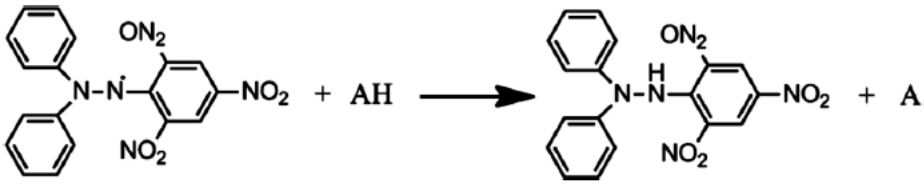

Figure 4. Reaction of antioxidant to scavenge DPPH radical

Bacterial and yeast enzymes were produced during fermentation so that complex compounds undergo breakdown into small polyphenol molecules under acidic environment of kombuchas (E. Ivanišová et al., 2019). *Gluconacetobacter sp.*in SCOBY cultureshad function togenerate theaflavin compounds, flavonol glycosides, ascorbic acid, glucuronic acid, glucaric acid, and saccharic acid-1,4-lactone. Oxidation and fermentation process of phenolic compounds cause an increase in antioxidant capacity. The fermentation process in making tea kombuchaincrease the content of phenolic compounds and produce glucuronic acid which can increase the antioxidant activity of kombucha (Değirmencioğlu et al., 2021). Catechins are broken down by the activity of bacteria and yeast in SCOBY cultures into simpler compounds that increase kombucha's antioxidant capacity. Moreover, fermentation with SCOBY also induce the breakdown of the tea cell wall structure to produce various antioxidant compounds.

Table 3 illustrates the results of antioxidant activity test expressed as percent inhibition. Kombuchas had the inhibition capacity in the range of 65,63 – 82,38%. This study found that black tea kombucha had the highest percent inhibition so that it's antioxidant activity was the highest among the other variants kombucha.

Table 3. The antioxidant activity of kombuchas

No.	Type of Kombucha	% Inhibition
1	BTK	82.38 ± 0.08%
2	GTK	78.02 ± 0.32 %
3	WTK	65.74 ± 0.68 %
4	OTK	65.63 ± 0.13 %

The results of antioxidant activity assay showed that the order of the kombucha from the highest percentage of inhibition was BKT > GTK > WTK > OTK. Similar result of previous research was observed that green tea kombucha had the higher antioxidant activity than white tea kombucha (Jakubczyk et al., 2020). These results revealed that kombucha had capacity to scavenge free radical. Many research reported that free radical contributed to rise degenerative diseases. Polyphenolic compoundsin kombucha contribute to the antioxidant activity. This compounds have capacity to neutralize endogeneous free radical such as anion superoxide, nitric oxide radical, hydroxyl radical,hydrogen peroxide, oxygen singlet radicals by scavenging activity (Jadid et al., 2017).

Previous study observed that antioxidant activity of kombucha was attributed not only by polyphenol but also by some metabolites such as organic acids and vitamins produced during fermentation (Eva Ivanišová et al., 2020). The fermentation process during manufacturing kombucha produces breakdown of tea cell wall and trigger production of antioxidant compounds which can act to terminate free radical, chelate metals, quenche oxygen and donor hydrogens (Jakubczyk et al., 2020).

Microorganisms in SCOBY cultures process various substances contained in tea and sugar into various metabolites. Organic acids are compounds produced by fermentation of sucrose as substrate for SCOBY microorganism. The results of total acid content of kombuchas are presented on Table 4. The organic acids of kombucha are acetic acid, glucuronic acid, gluconic acid, tartaric acid, malic acid, citric acid, lactic acid, succinic acid and malonic acid. The evaluation showed that the sequence of total acid content from the highest was OTK > GTK > BTK > WTK. This result was similar with the previous study reporting that green tea kombucha had higher acidity than black tea kombucha (Wang et al., 2022).

Table 4. The total acid content of kombuchas

No.	Type of Kombucha	Total Acid Content (%)
1	BTK	0.92 ± 0.02 %
2	GTK	1.04 ± 0.01 %
3	WTK	0.80 ± 0.01%
4	OTK	1.23 ± 0.02%

Acetic acid is the main acid in kombucha drinks (Coelho et al., 2020). Acetic acid is obtained from the oxidation of glucose and fructose derived from sucrose by acetic acid bacteria (Khaerah & Akbar, 2019).The formation of these organic acids causes the sour taste and aroma of kombucha product. Dissolved acetic acid release free protons causing a decrease the pH of solution. Reducing the pH value during fermentation support the life of *Acetobacter xylinum* bacteria in the kombucha culture to carry out its metabolic activities. Gluconic acid is the other major organic acid found in kombucha. This organic acid has beneficial effect for kombucha due to giving refreshing, mild and soft taste. The organic acid content increase with the prolonged fermentation time. Glucuronic acid produced during fermentation of

kombucha is a beneficial detoxifier since it binds the toxic materials in the liver hence these compounds are easily excreted by the kidney (Coelho et al., 2020).

Sucrose is a carbon source for microorganism of SCOBY. The sugar content of kombucha will change with fermentation time. Yeast hydrolyze sucrose into reducing sugars, glucose and fructose.The yeast then converts reducing sugars in an oxygen-free environment into ethanol. This process causes the decreasing of sugar content with increasing fermentation time (Jakubczyk et al., 2020).The low sugar content reduce the calorie content of the kombucha drink so that the kombucha is safer for consumption by diabetes mellitus sufferers.The total sugar content of kombuchas is summarized in the Table 5. The order of the total sugar content of kombucha from the lowest was BTK<GTK<WTK<OTK.

Table 5. The total sugar content of kombuchas

No.	Type of Kombucha	Total Sugar Content (%)
1	BTK	3.95%
2	GTK	4.72%
3	WTK	4.84%
4	OTK	5.37%

CONCLUSION

Kombuchas were successfully produced from four variants of tea, consisting of black tea, green tea, white tea and oolong tea. The type of tea influenced the characteristics of kombucha in term of antioxidant activity, the total phenolic content, the total acid content and the total sugar content. This study found that black tea kombucha had the highest antioxidant capacity among the other variant kombuchas. On the other hand green tea kombucha and white tea kombucha had the higher phenolic content than the other type kombucha. Therefore, the selection of variant of tea needs to be considered to produce good quality kombucha.

ACKNOWLEDGMENTS

The authors gratefully acknowledge Politeknik Kesehatan Kemenkes Tasikmalaya for supporting this research through DIPA Politeknik Kesehatan Kemenkes Tasikmalaya 2023.

REFERENCES

Chakravorty, S., Bhattacharya, S., Bhattacharya, D., Sarkar, S., & Gachhui, R. (2019). Kombucha: A promising functional beverage prepared from tea. In *Non-alcoholic Beverages: Volume 6. The Science of Beverages*. Elsevier Inc. https://doi.org/10.1016/B978-0-12-815270-6.00010-4

Coelho, R. M. D., Almeida, A. L. de, Amaral, R. Q. G. do, Mota, R. N. da, & Sousa, P. H. M. d. (2020). Kombucha: Review. *International Journal of Gastronomy and Food Science*, *22*(July), 100272. https://doi.org/10.1016/j.ijgfs.2020.100272

Değirmencioğlu, N., Yıldız, E., Sahan, Y., Güldas, M., & Gürbüz, O. (2021). Impact of tea leaves types on antioxidant properties and bioaccessibility of kombucha. *Journal of Food Science and Technology*, *58*(6), 2304–2312. https://doi.org/10.1007/s13197-020-04741-7

Gaur, S., & Agnihotri, R. (2014). Green tea: A novel functional food for the oral health of older adults. *Geriatrics and Gerontology International*, *14*(2), 238–250. https://doi.org/10.1111/ggi.12194

Gulcin, İ., & Alwasel, S. H. (2023). DPPH Radical Scavenging Assay. *Processes*, *11*(8). https://doi.org/10.3390/pr11082248

Ivanišová, E., Meňhartová, K., Terentjeva, M., Godočíková, L., Árvay, J., & Kačániová, M. (2019). Kombucha tea beverage: Microbiological characteristic, antioxidant activity, and phytochemical composition. *Acta Alimentaria*, *48*(3), 324–

331. https://doi.org/10.1556/066.2019.48.3.7

Ivanišová, Eva, Meňhartová, K., Terentjeva, M., Harangozo, Ľ., Kántor, A., & Kačániová, M. (2020). The evaluation of chemical, antioxidant, antimicrobial and sensory properties of kombucha tea beverage. *Journal of Food Science and Technology*, *57*(5), 1840–1846. https://doi.org/10.1007/s13197-019-04217-3

Jadid, N., Hidayati, D., Hartanti, S. R., Arraniry, B. A., Rachman, R. Y., & Wikanta, W. (2017). Antioxidant activities of different solvent extracts of Piper retrofractum Vahl. using DPPH assay. *AIP Conference Proceedings*, *1854*. https://doi.org/10.1063/1.4985410

Jakubczyk, K., Kałduńska, J., Kochman, J., & Janda, K. (2020). Chemical profile and antioxidant activity of the kombucha beverage derived from white, green, black and red tea. *Antioxidants*, *9*(5). https://doi.org/10.3390/antiox9050447

Khaerah, A., & Akbar, F. (2019). Aktivitas Antioksidan Teh Kombucha dari Beberapa Varian Teh yang Berbeda. *Prosiding Seminar Nasional LP2M UNM*, 472–476.

Leslie, P. J., & Gunawan, S. (2019). Uji Fitokimia dan Perbandingan Efek Antioksidan Pada Daun Teh Hijau , Teh hitam , dan Teh putih (Camellia sinensis) dengan Metode DPPH (2 , 2-difenil-1- pikrilhidrazil). *Tarumanagara Medical Journal*, *1*(2), 383–388.

Linnarto, F. P., Gunawan, K. P., Setiadi, M., Ashyari, R. A., & Lukman, S. (2019). Teh Putih sebagai Alternatif Minuman Fungsional untuk Gaya Hidup Sehat: Peluang Komersialisasi di Indonesia. *Indonesian Business Review*, *2*(1), 139–159. https://doi.org/10.21632/ibr.2.1.139-159

Massoud, R., Jafari-Dastjerdeh, R., Naghavi, N., & Khosravi-Darani, K. (2022). All aspects of antioxidant properties of kombucha drink. *Biointerface Research in Applied Chemistry*, *12*(3), 4018–4027. https://doi.org/10.33263/BRIAC123.40184027

Namal Senanayake, S. P. J. (2013). Green tea extract: Chemistry, antioxidant properties and food applications - A review. *Journal of Functional Foods*, *5*(4), 1529–1541. https://doi.org/10.1016/j.jff.2013.08.011

Pérez-Burillo, S., Giménez, R., Rufián-Henares, J. A., & Pastoriza, S. (2018). Effect of brewing time and temperature on antioxidant capacity and phenols of white tea: Relationship with sensory properties. *Food Chemistry*, *248*, 111–118. https://doi.org/10.1016/j.foodchem.2017.12.056

Piyasena, K. G. N. P., Abayarathne, A. A. B., Weerakoon, N. C., Edirisinghe, E. N. U., Jayasinghe, W. S., Napagoda, M. T., Hettiarachchi, L. S. K., & Abeysinghe, I. S. B. (2023). Chemical and biological characteristics of Sri Lankan white tea. *Food and Humanity*, *1*(August), 966–972. https://doi.org/10.1016/j.foohum.2023.08.019

Pratiwi, A. P. (2018). *Optimasi Formulasi Teh Putih, Maltodekstrin, Dan Natrium Bikarbonat Terhadap Karakteristik Effervescent Teh Putih*. http://repository.unpas.ac.id/38585/%0Ahttp://repository.unpas.ac.id/38585/1/syarat wisuda %281%29.pdf

Sharpe, E., Hua, F., Schuckers, S., Andreescu, S., & Bradley, R. (2016). Effects of brewing conditions on the antioxidant capacity of twenty-four commercial green tea varieties. *Food Chemistry*, *192*, 380–387. https://doi.org/10.1016/j.foodchem.2015.07.005

Villarreal-Soto, S. A., Beaufort, S., Bouajila, J., Souchard, J. P., Renard, T., Rollan, S., & Taillandier, P. (2019). Impact of fermentation conditions on the production of bioactive compounds with anticancer, anti-inflammatory and

antioxidant properties in kombucha tea extracts. In *Process Biochemistry* (Vol. 83). Elsevier Ltd. https://doi.org/10.1016/j.procbio.2019.05.004

Wang, X., Wang, D., Wang, H., Jiao, S., Wu, J., Hou, Y., Sun, J., & Yuan, J. (2022). Chemical Profile and Antioxidant Capacity of Kombucha Tea by the Pure Cultured Kombucha. *Lwt*, *168*(September), 113931. https://doi.org/10.1016/j.lwt.2022.113931

Zanwar, A. A., Badole, S. L., Shende, P. S., Hegde, M. V., & Bodhankar, S. L. (2013). Antioxidant Role of Catechin in Health and Disease. In *Polyphenols in Human Health and Disease* (Vol. 1). Elsevier Inc. https://doi.org/10.1016/B978-0-12-398456-2.00021-9

Zimmermann, B. F., & Gleichenhagen, M. (2011). The effect of ascorbic acid, citric acid and low pH on the extraction of green tea: How to get most out of it. *Food Chemistry*, *124*(4), 1543–1548. https://doi.org/10.1016/j.foodchem.2010.08.009