



Total antioxidant and Total oxidant status of Fermented Food Kombucha Tea

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ABSTRACT: Fermented foods are widely used today. The properties of fermented foods, which have an important place in human nutrition and health, vary depending on the food product used. Fermented foods, which stand out with their nutritional properties, also have significant effects on health. In our study, the total antioxidant status, total oxidant status and oxidative stress index of kombucha tea as a fermented beverage were determined. In this context, kombucha tea produced as a result of fermentation was used as a material. TAS, TOS and OSI values of the tea were measured with Rel Assay kits. As a result of the analyzes, the TAS value of kombucha tea was determined as 9.458 ± 0.116 mmol/L, TOS value as 10.507 ± 0.141 μ mol/L and OSI value as 0.111 ± 0.001 . In this context, it was determined that kombucha tea, which has positive effects on the digestive system, can be used as an antioxidant source.

Keywords: Antioxidant, fermented foods, functional foods, oxidant, health.

INTRODUCTION

Fermented foods are the transformation of a food product into different properties through enzymatic effects. In this process, the controlled growth of health-friendly microorganisms that preserve the nutritional properties of the food is ensured. Fermented foods have a very old history. They have become quite popular today due to their positive effects on the digestive system. The fermentation technique used to obtain fermented foods was used to preserve food and beverages in human history. In this context, it is widely applied to make foods with different tastes and textures preferred foods through fermentation (Şanlıer et al., 2019; Ilango and Antony, 2021; Yu et al., 2021; Dahiya and Nigam, 2022; Leeuwendaal et al., 2022). In our study, the total antioxidant level, total oxidant level and oxidative stress index of the fermented food known as kombucha were determined.

Kombucha is an important tea consisting of bacteria and fungi. It is of Asian origin. The fungus consists of a gelatinous membrane and is disc-shaped. Kombucha is based on the principle of the growth of the fungus in a mixture of tea and sugar. Over time, the fungus spreads throughout the mixture and tends to grow thicker. It contains organic acids, active enzymes, and amino acids. It is classified as a non-alcoholic beverage because it contains less than 0.5% alcohol. By increasing the brewing time and using high amounts of sugar and yeast, an alcohol content of 1.0% or 1.5% can be achieved. The brewing period is an average of 10 days. This tea is widely known for its soothing effect on the digestive system (Kallel et al., 2012; Jayabalan et al., 2014; Neffe-Skocińska et al., 2017). It is recommended that the production stages be at room temperature. It is important to purify the environment so as not to affect the fermentation process. A clean cloth should be covered over the container during the fermentation phase to ensure that it is more hygienic. The consumption of kombucha produced under these conditions is very important for the digestive system (Chakravorty et al., 2016; Villarreal-Soto et al., 2018; Nyhan et al., 2022).

MATERIALS AND METHODS

Kombucha fermentation process

In our study, sucrose sweetened black tea is the most common substrate for kombucha production. The production stage of kombucha tea is shown in Figure 1. According to this method, 50 g of sucrose is dissolved in 1 liter of boiling water. Then black tea leaves are added. Then, after 5-10 minutes, the leaves are removed using a filter. Then, it is left to cool for a certain period of time. Then, kombucha culture and 20 g of sucrose are added. Then, a clean cloth is covered on the jar for 2 weeks of fermentation and fermentation is waited in a dark environment.

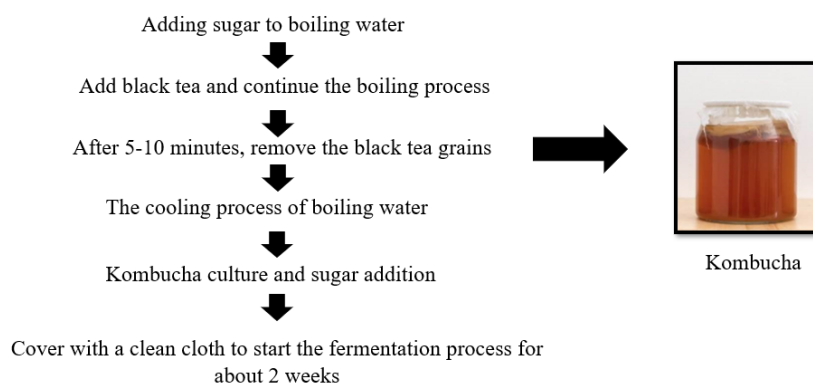


Figure 1. Kombucha fermentation process

Total antioxidant and oxidant status

Total antioxidant (TAS) and oxidant (TOS) values of kombucha tea were determined using Rel assay kits. The manufacturer's protocol was followed for the analyses. Rel Assay Diagnostics-TAS Assay Kit was used to determine antioxidant status. The kit contains Reagent 1 (Buffer), Reagent 2 (Colored ABTS Radical Solution), Standard 1 (1.00 mmol Trolox Equiv./L) and Standard 2 (1.00 mmol Trolox Equiv./L). 200 μ L of Reagent 1 was added to the plate wells. Then 12 μ L of kombucha tea was added. The first absorbance was determined at 660 nm. Then 30 μ L of Reagent 2 was added. Incubation was performed at 37 $^{\circ}$ C for 5 min. Then the second absorbance was determined at 660 nm (Erel, 2004).

Rel Assay Diagnostics-TOS Assay Kit was used for the oxidant status test. The kit contains Reagent 1 (Assay buffer), Reagent 2 (Prochromogen solution), Standard 1 (Blank solution: distilled water) and Standard 2 (stock stabilized standard solution (SSSS): 800mM H₂O₂ Equiv./L). In this context, standard 2 was diluted 40 times using distilled water. Then, 5 μ L of standard 2 was taken into Eppendorf and 1mL of distilled water was added. It was then vortexed. 5 μ L of this solution was taken into Eppendorf and 1mL of water was added. In this context, 20 μ Molar H₂O₂ was prepared. This solution was prepared again each time. Then, 200 μ L of Reagent 1 was first placed in the well on the plate and 30 μ L of kombucha tea was added on it. Then the first absorbance was read at 530 nm. Then 10 μ L of Reagent 2 was added and incubated at 37 $^{\circ}$ C for 5 min. Then the second absorbance was read at 30 nm (Erel, 2005).

For OSI (Oxidative stress index) determination, the unit of the total oxidant value was equalized to the unit of the total antioxidant value. Then the total oxidant value was proportioned to the total antioxidant value and the percentage was obtained (Sevindik, 2021).

RESULTS AND DISCUSSION

Antioxidant activity

Free radicals are compounds with oxidative properties produced as a result of metabolic activities (Uysal et al., 2023). As the levels of these compounds increase, serious cellular damage can occur (Mushtaq et al., 2020). In order to prevent this cellular damage, the antioxidant defense system steps in and suppresses oxidant compounds (Mohammed et al., 2021). When oxidant compounds come against the antioxidant defense system, oxidative stress occurs (Eraslan et al., 2021). Serious diseases such as diabetes, cancer, cardiological disorders, neurodegenerative diseases, and obesity can occur as a result of oxidative stress. Supplemental antioxidants serve to reduce or suppress the effects of oxidative stress (Bal et al., 2023; Mohammed et al., 2023; Sevindik et al., 2023; Yaz et al., 2023). In our study, the antioxidant potential of kombucha tea was determined. The findings are shown in Table 1.

Table 1. TAS, TOS and OSI values of Kombucha tea

Solvent	TAS (mmol/L)	TOS (μ mol/L)	OSI (TOS/(TASX10))
Kombucha tea	9.458 \pm 0.116	10.507 \pm 0.141	0.111 \pm 0.001

Values are given as mean \pm standard deviation. (n=3)

As a result of literature research, no findings were found regarding TAS, TOS and OSI values of kombucha tea. The antioxidant potential of kombucha tea has been reported using different methods (Chu and Chen, 2006; Malbaša et al., 2011;

Lobo et al., 2017; Ahmed et al., 2020; Massoud et al., 2022). In our study, antioxidant and oxidant status of kombucha tea was determined using Rel Assay kits. TAS, TOS and OSI values of non-fermented but natural plant and mushroom species have been reported in the literature. The antioxidant potential of kombucha tea used in our study can be evaluated by comparing with these studies. In this context, TAS values of *Marrubium globosum*, *Centaurea rigida*, *Echium italicum*, *Cantharellus cibarius*, *Clavariadelphus truncatus*, *Macrolepiota procera* and *Salvia absconditiflora* were reported as 7.677, 3.522, 4.896, 5.268, 2.415, 2.823 and 7.350 mmol/L, respectively. TOS values were reported as 12.387, 15.424, 12.255, 6.380, 3.367, 10.349 and 8.501 μ mol/L, respectively. OSI values were reported as 0.162, 0.440, 0.250, 0.121, 0.140, 0.367 and 0.116, respectively (Sevindik, 2018; Sevindik, 2019; Akgul et al., 2016; Akgul et al., 2020; Kına et al., 2021; Pehlivan et al., 2021; Uysal et al., 2021). Compared to these plant and mushroom species, the TAS value of kombucha tea used in our study was determined to be higher than *M. globosum*, *C. rigida*, *E. italicum*, *C. cibarius*, *C. truncatus*, *M. procera* and *S. absconditiflora*. The TAS value is an indicator of the entirety of endogenous antioxidant compounds. It was determined that the kombucha tea used in our study had higher antioxidant potential compared to natural plant and mushroom species. TOS value is an indicator of the totality of endogenous oxidant compounds. The TOS value of the kombucha tea used in our study was determined to be lower than *M. globosum*, *C. rigida*, *E. italicum*, and higher than *C. cibarius*, *C. truncatus*, *M. procera* and *S. absconditiflora*. In this context, it is thought that kombucha tea has normal oxidant compound levels. The OSI value shows the percentage suppression of endogenous oxidant compounds by endogenous antioxidant compounds. The OSI value of the kombucha tea used in our study was found to be lower than *M. globosum*, *C. rigida*, *E. italicum*, *C. cibarius*, *C. truncatus*, *M. procera* and *S. absconditiflora*. This result shows that kombucha tea has a significant potential in suppressing oxidant compounds. As a result, it has been determined that kombucha tea, which has significant positive effects on the digestive system, also has a high antioxidant potential.

CONCLUSION

In this study, the total antioxidant status, total oxidant status and oxidative stress index of kombucha tea were determined. As a result of the analyzes, it was determined that the antioxidant potential of kombucha tea was high. In this context, it was determined that kombucha tea, which is a fermented food, has antioxidant potential and can be an antioxidant source in this context.

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CONFLICT OF INTEREST

No conflict of interest was declared by the authors.

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