

Review

A Review of Kombucha Antioxidant Effects On Atherosclerosis In Preclinical Studies

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ABSTRACT

Background Atherosclerosis is the leading cause of death among cardiovascular diseases in Indonesia, in which death is possible if uncontrolled. The prevalence of the disease may be prevented by consuming foods or drinks containing high levels of antioxidant such as kombucha. Exploration of kombucha properties as an antioxidant that will improve atherosclerosis could be obtained from preclinical studies.

Objective To discuss the antioxidant effect of kombucha on atherosclerosis in preclinical studies.

Method This study is a narrative review with literatures from 2013 to 2022.

Result Kombucha is an antioxidant-rich beverage made from enzymes generated by the SCOBY bacteria, which are fermented in sweet tea for 7-14 days to achieve optimal quantities of polyphenols. In addition to the antioxidants in the blood, the effect will lower the levels of LDL, VLDL, triglycerides, and total cholesterol in the blood. Moreover, it can increase HDL in blood and inhibit lesion forming in the aorta.

Conclusion Polyphenol in kombucha tea has an antiatherogenic effect that may prevent atherosclerosis in preclinical studies.

Keywords: antioxidant, atherosclerosis, kombucha, polyphenol

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INTRODUCTION

According to WHO 2018 non-communicable diseases are the leading causes of death in middle and low-income countries including Indonesia, 35% of them due to the cardiovascular disease.¹ Inadequate blood supply to the heart is the main cause of cardiovascular diseases which may result in death. The accumulation of fat in the arteries, often known as atherosclerosis, is one of the causes. Many causes can contribute to atherosclerosis, but the most frequent is a rise in blood cholesterol levels. An imbalance between free radicals and antioxidants is

created by a sedentary lifestyle.² If atherosclerosis is uncontrolled, it can lead to complications as result, there are a variety of approaches to prevent atherosclerosis, one of which is to consume antioxidant-rich foods or beverages.³

One functional drink that is rich in antioxidants is Kombucha. This kombucha has been consumed in Asia for over two millennia and is claimed to have a variety of health advantages. Kombucha has become popular in Indonesia, particularly among youngsters, because of its distinct flavor and several health advantages.⁴

Kombucha is a sugared, fermented tea that also contains a symbiotic culture of acetic acid bacteria and fungus, often known as symbiotic culture of bacteria and yeast (SCOBY) as its main ingredients.⁵ This beverage is usually fermented for seven to fourteen days at room temperature. According to several studies, kombucha that has been fermented for fourteen days has a high antioxidant content such as phenol, which decreases if it is fermented for more than two weeks.^{5,6} Kombucha's phenolic content has an antihyperlipidemic effect, which can help decrease cholesterol levels in the bloodstream, specifically Low Density Lipoproteins known as LDL and prevent atherosclerosis.^{6,7} This phenol is generated in kombucha tea by the secretion of enzymes from bacteria and fungus in the Kombucha.⁶ The purpose of this review is to explore the anti-atherosclerotic property of kombucha

by lowering cholesterol levels in particular LDL, and its effect on the tunica intima in arteries after 14 days of kombucha intake. These findings were obtained from preclinical studies.

METHODS

The following is a summary of 9 experimental studies conducted over the last 10 years (2013-2022). The narrative approach was constructed to compile this review, which began with a PubMed search using the keywords kombucha tea, polyphenols and atherosclerosis. The studies selection proceeded by categorizing and limiting the content and subjects to kombucha and animal experimental studies. Literatures excluded were the ones with unavailable full text and over 10 years prior to the study.

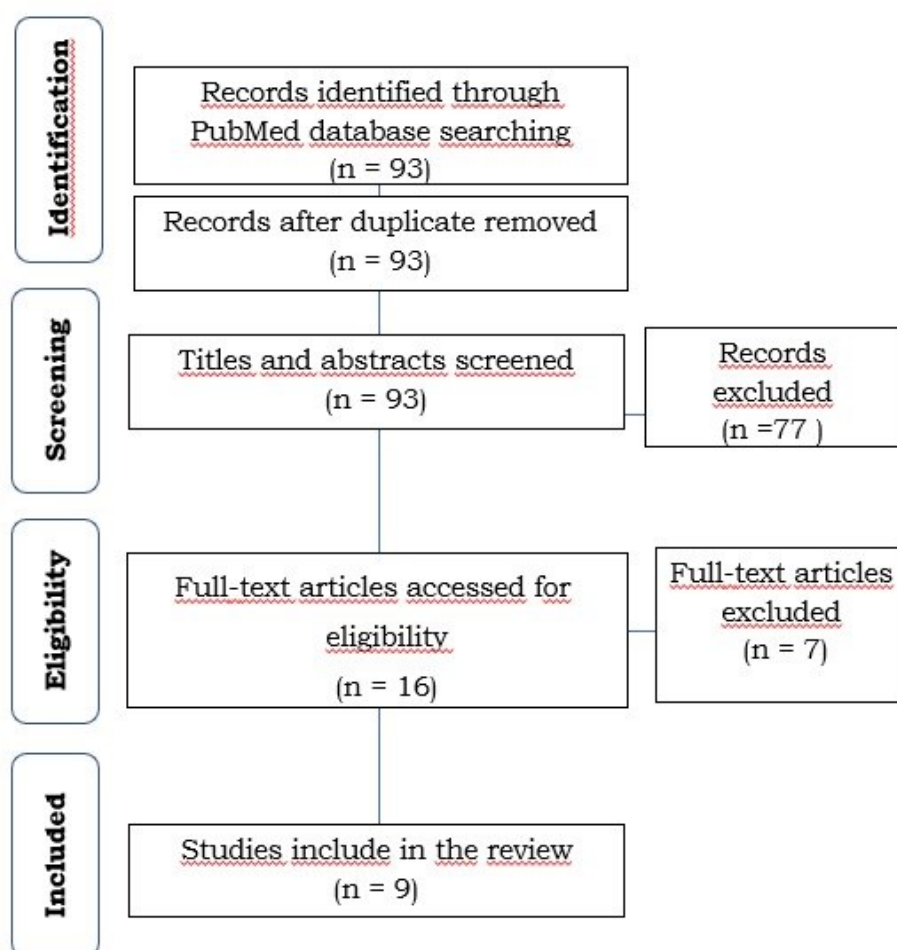


Figure 1. Flowchart of Study Selection

RESULTS AND DISCUSSION

1. Preparations and The Levels of Polyphenols in Kombucha Tea

The main ingredients for kombucha tea were sugared tea and SCOBY. In this review the kombucha was made using black tea and green tea. The SCOBY is then added to the sugared tea and stored in a large jar. Furthermore, this mixture were fermented for 7 to 14 days at room temperature, producing a tea with sweet and sour flavor and slightly alcoholic.^{5,8} There were two layers in this drink: a liquid layer at the bottom and an overtop biofilm.

The polyphenols content measurement employed Folin-Ciocalteu

spectrophotometry by mixing the kombucha and the Folin-Ciocalteu reagent, then it was calculated using the 765nm absorbent^{5,6}. Antioxidant action is caused by a characteristic of the total phenol content, the greater the phenolic content, more free radicals are scavenged; thus, the antioxidant activity increases.⁹ The total phenol content was expressed as gallic acid equivalents (GAE, mM) from the calibration curve.^{5,6,8} By the results of this measurement, it was found that the increase in antioxidant activity was proportional to the fermentation time and based on the type of tea that was used in several studies, the results were as shown in table 1:

Table 1. Antioxidant Activity in 0 – 21 Days of Fermentation

Author (unit)	Type of tea	Day 0	Day 7	Day 14	Day 21
Jakubczyk <i>et al</i> (mg/L) ⁵	Black tea kombucha	183.1 ± 2.3	219.5 ± 2.1	206.0 ± 1.2	-
Tanticharakunsiri <i>et al</i> (mg/L) ⁸	Oolong tea kombucha	455.2 ± 2.3	473.27 ± 2.7	485.2 ± 1.7	480.6 ± 5.0
Jakubczyk <i>et al</i> (mg/L) ⁵	Green tea kombucha	269.0 ± 0.9	299.6 ± 3.1	320.1 ± 3.5	-
Watawana <i>et al</i> (mg/mL) ¹⁰	Black tea kombucha	31.5 ± 1.9	39.7 ± 1.5	No Measurement taken	-
Li <i>et al</i> (µm/mL) ⁶	Green tea kombucha	-	-	362.9*	-
Tan <i>et al</i> (µm/mL) ¹¹	Black tea kombucha		42.0 ± 0.1	38.0 ± 0.1	42.2 ± 0.1

*measured on day 15

From Table 1, it can be concluded that the polyphenol content in Kombucha tea increased with the length of fermentation time. However, in a study conducted by Wanlapa et al, on fermentation day 21 there was a decrease in the number of polyphenols in kombucha tea, proving that the optimal fermentation time for kombucha was between 7-14 days.¹⁰

Total polyphenol content in kombucha also affects the color of the tea, and the color of the tea gets more concentrated as the number of polyphenols increases. The antioxidant activity was reduced by 25% after the time mentioned before.⁸ Total polyphenol content in kombucha tea is greater than the unfermented tea.¹² The release of enzymes from bacteria and fungus

in the SCOBY produces the polyphenols in kombucha tea.¹¹ Phytase, an enzyme found in kombucha, can break down complex polyphenols into smaller ones.^{8,12} As a result, Kombucha tea contains higher phenol than unfermented tea.^{5,6,8,11,13}

2. Polyphenol Effect on lipid profile and Histological Structure on Animals

The antioxidant effect depends on the total polyphenol content in the kombucha. The antioxidant effect is greater when the concentration is higher. The polyphenols in kombucha can lower LDL levels by reducing LDL oxidation. Similarly, kombucha's free radical scavenging action can raise HDL and lower total cholesterol, triglycerides, VLDL-

C, and LDL-C.¹³ The lipid profile of mice given kombucha is described in Table 2.

Table 2. lipid profile of mice and phenolic content given kombucha

Author	Experimental Material	Length of Fermentation (days)	TPC	TC	TG	LDL	VLDL
Bellassoued et al ¹³	kombucha	7	955 ± 0.75	↓ (26%)	↓ (27%)	↓ (36%)	↓ (28. %)
Lobo et al ¹⁴	kombucha	7	-	↓	↓	↓	↓
Bhattacharya et al ¹²	kombucha	14	-	↓	-	↓	-
Cardoso et al ⁷	kombucha	7	-	-	↓	-	-

Notes: Total polyphenol content (TPC), total cholesterol (TC), triglycerides (TG). Low Density Lipoprotein (LDL), Very Low-Density Lipoprotein (VLDL), in experiment number 2 and 3 it is not explained the percentage of the decrease is obtained.

In the experiment conducted by Khaled et al, experimental rats given a high cholesterol diet were divided into two groups, the first group was treated with kombucha, and the other was given a drink with *Camellia sinensis* as the main ingredient in these two drinks contained polyphenol.¹³ This study proved that kombucha has a higher concentration of polyphenols and as result it has more significant antioxidant activity. The findings in this study show that HDL levels in the blood were increased, causing total cholesterol, triglycerides, VLDL, and LDL levels to decrease. Reema et al and Semantee et al discovered the same outcome in their experiments.^{12,14} Reema et al researched with 24 male rats separated into four groups: the control, the treatment control group which was induced by Isoproterenol (ISO), the third group which was induced by ISO dan tea, and the last group induced with ISO and Bio-tea pre-treated (kombucha) for 30 days followed by an analysis of lipid profile. The results indicated that rats receiving kombucha had lower total cholesterol, lower LDL and VLDL cholesterol, and lower triglycerides.¹⁴ The rats in the Sementee experiment were divided into six groups: control, normal rats given kombucha tea at a dose of 150mg/kg BW, alloxan induced diabetic rats, alloxan induced diabetic rats given kombucha tea at

a dose of 150mg/kg BW for 14 days, alloxan induced diabetic rats given black tea at a dose 150 mg/ kg BW, and alloxan induced diabetic rats given glibenclamide at a dose of 1 mg/kg BW. This treatment was given for 14 days and then a cardiac puncture was performed to analyze the lipid profile of the mice. The results of the analysis revealed a decrease in serum levels of total cholesterol and triglycerides and an increase in HDL.¹² In study conducted by Cardoso et al wistar rats with metabolic changes by given high fat high fructose (HFHF) diet for 8 weeks were divided into 4 groups AIN-93M control group, HFHF control group, green tea kombucha + HFHF diet, and black tea kombucha + HFHF diet, this treatment were observed for 10 weeks and were tested for blood triglyceride, both kombuchas reduced the blood triglyceride levels.⁷

This antioxidant effect of kombucha can also be analyzed on histology of the aorta in rabbits. In this experiment, the kombucha had been fermented for 7 days, then given to rabbits which were treated with a high-cholesterol diet. After 40 days of kombucha treatment, the rabbit aorta was examined histologically under a microscope and the results were normal coronary arteries and no intima lesions, whereas rabbits receiving only a high-cholesterol diet without kombucha had intimal lesions.¹⁵

The antioxidant activity of polyphenols in kombucha was responsible for the reduction in lipid profile levels. Catechins are one of the polyphenol compounds that have been evaluated for their ability to lower cholesterol levels in the blood, as indicated by studies, including Liu et al and Mika et al.^{16,17} Liu separated their study's experimental mice into four groups of four for the purpose of the feeding tests for 12 weeks: 0.3% catechins, 0.05% caffeine, 0.3% catechins + 0.05% caffeine, and control. After the feeding period each aorta shown that mice treated with both catechins and caffeine had smaller lipid staining than the other group.¹⁶ On the other hand, studies conducted by Mika et al use mice as experimental animals and they were divided

into 5 groups: the first received standard diet, the other had high fat diet, high fat diet with native catechins, high fat diet with modified catechins, and the last group had high fat diet with thermally modified catechins for 10 weeks, in this studies shown groups who received catechins had lower total plasma cholesterol level and lower triglyceride plasma level with a simultaneous higher HDL level than groups with high fat diet. Mika et al studies also shows that catechins can reduced the area of atherosclerotic lesions up to 45% compared to the group with high fat diet.¹⁷ It is concluded that the antioxidant activity obtained from polyphenols can reduce total cholesterol levels and have antiatherogenic effects on aorta.^{16,17}

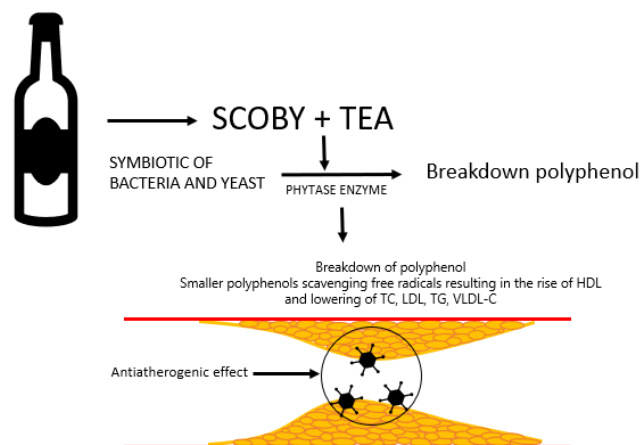


Figure 2 antioxidant effect from kombucha^{13,14}

The mechanism of antioxidant effect in kombucha is described in 2, it is produced by the phytase enzyme generated by SCOBY, which breaks down polyphenols into smaller components allowing them to have an atherogenic effect by inhibiting LDL oxidation, lowering TC, TG, VLDL-C, and LDL-C, and increasing HDL levels in the blood. Furthermore, this smaller complex will scavenge free radicals in the circulation, preventing lesions from forming in blood vessels.

Studies included in this review have shown a similar result that increasing fermentation time will increase total polyphenols in kombucha tea, with 7-14 days as an optimal fermentation time. Regarding atherosclerosis, kombucha tea has a

potential role in preventing atherosclerosis because SCOBY produced phytase enzyme that breaks down polyphenols into smaller components that scavenges free radicals.

According to research by Herald et al, some individuals such as those with low antioxidant levels may benefit from antioxidant supplementation only under certain circumstances. However, the administration of antioxidants does not always have a positive impact. Exogenous antioxidants whether used on their own or in combination with lipid lowering medications, may inhibit the activity of enzymes that prevents the activation of signals associated with endothelial inflammation. The consequence of this is worse.¹⁸

Further studies needed to elaborate more about which content in kombucha tea have the greatest role in preventing atherosclerosis and which molecular pathway involved in the mechanism. The limitation of this study is the limited number of original articles focusing on the effect of kombucha tea in relation to its fermentation time and effect on atherosclerosis prevention.

CONCLUSION

Coronary heart disease has become the leading cause of premature death in Indonesia, and one of the most common factors is atherosclerosis as the cause of fat accumulation in the blood vessels. This is caused by the oxidation of LDL that blocks the blood flow. This can be exacerbated when the patient adopts an unhealthy lifestyle. However, this can be prevented by changing the unhealthy diet to a diet rich in antioxidants such as kombucha.

Kombucha tea is a functional drink that contains high levels of antioxidants. Polyphenols in kombucha tea have antioxidant properties that will elevate proportional to the fermentation time. SCOBY and tea break down polyphenols to scavenge free radicals and reduce LDL oxidation and aggregation. This activity resulted in an improvement in lipid profile. It can also inhibit fat accumulation in the aorta so that it can reduce the risk of coronary heart disease. In conclusion, kombucha tea can be considered a way to prevent atherosclerosis by improving lipid profile through the mechanism of scavenging free radicals by breaking down polyphenols.

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CONFLICT OF INTEREST AND FUNDING RESOURCES

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