Phenolic Compounds in Green Tea and Green Tea Kombucha

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## Introduction

Green tea, white tea, and black tea originate from the *Camellia sinensis* plant which is native to the tropical and temperate regions of Asia, Africa, and South America (Gopal et al. 2016). Small leaves and leaf buds are unfermented and are used to make green tea, while white tea is composed of semi-fermented buds, and black tea is composed of fully fermented old leaves (NCSU 2022). Tea is a globally consumed beverage second only to water and the drink is praised for its numerous health benefits. Briefly, tea has anti-carcinogenic, anti-angiogenic, anti-mutagenic, anti-inflammatory, anti-bacterial, hypocholesterolemic, anti-diabetic, and shows protection against Parkinson’s and Alzheimer’s disease (Chacko et al. 2010, Gopal et al. 2016). These and other health benefits are attributed to green tea polyphenols, less so to flavonols and gallic acid derivatives, vitamins, minerals, enzymes, and others (Gopal et al. 2016). Another health drink rising in popularity is Kombucha - which is a fermented beverage resulting from a symbiotic culture of bacteria and yeast (SCOBY) in a sweetened tea solution for about two weeks. The flavor profile shifts from sweet to tart to sour with increased fermentation time, temperature, and SCOBY microbe composition. The total phenolic content of kombucha made from green or black tea is over three times greater than regular green or black tea (Zhou et al. 2022). Tea is popular for its touted health benefits and fermentation with a SCOBY into kombucha enhances these benefits. It is suggested by Jakubczyk et al. (2020) a diet including kombucha can help support the body’s antioxidative response, especially for those exposed to mental and physical stress.

## Polyphenols of Green Tea and Green Tea Kombucha

The components of green tea (GT) that gives the drink its health benefits come from polyphenols, flavanols, and gallic acid derivatives (Gopal et al. 2016). Important compounds among the polyphenols includes alkaloids, carbohydrates, catechins, enzymes, free amino acids, minerals and vitamins. The health benefits are most particularly associated with the catechins, which make up 25% - 35% of GT dry weight. The two most effective antioxidant compounds are epigallocatechingallate (EGCG), epicatechingallate (ECG). EGCG is also the most active and abundant - one cup of GT could have 100-200 mg EGCG. Gopal et al. (2016) found that EGCG can help prevent dental cavities, can help inhibit multiple drug resistant Staphylococcus aureus infections, and inhibition of HIV infection.

Fermentation is the metabolic breakdown of carbohydrates that produces chemical changes in the organic substances via the action of enzymes associated with the SCOBY (Redzepi and Zilber 2018). Fermentation with a SCOBY - symbiotic culture of bacteria and yeast - can increase the total polyphenolic concentration over five-fold when compared to GT without SCOBY fermentation (Zhou et al. 2022). Likewise, antioxidant ability increased over three-fold in green tea kombucha (GTK) with tea residue vs GTK or GT. The health benefits associated with GT are related to catechin consumption and absorption but is limited by absorption. Fermentation increases the concentration of phenolic compounds and increases the antioxidant potential. Cardoso et al. (2020) found 127 phenolic compounds associated with GTK with 70% of compounds in the flavonoid class (i.e., catechins), and the last 30% split between phenolic acids and other polyphenols. Among these compounds are also vitamins B, E, and K, fluoride, potassium, and manganese ions, amino acids, and a variety of organic acids - acetic, citric, gluconic, glucuronic, lactic, malic, malonic, and succinic (Jakubczyk et al. 2020). Tea type has a significant effect on antioxidant potential, pH, acidity, alcohol content, and sugar content, with greater effects in GTK vs black, white, or red tea derived kombuchas. It is likely that tea quality also influences these properties, potentially increasing antioxidant activity and concentration even further. However, further research is needed to determine the phenolic profiles of various tea combinations for kombucha fermentation.

## Health Benefits of Green Tea and Green Tea Kombucha

Green tea (GT) has antioxidative, anti-carcinogenic, anti-diabetic, anti-hypertensive, anti-inflammatory, anti-mutagenic, anti-proliferative, anti-thrombogenic, among other properties (Velayutham et al. 2008, Chacko et al. 2010, Gopal et al. 2016) giving the drink its status as a health drink. Heinrich et al. (2011) have shown that a flavanoid-rich diet can help prevent certain cancers and cardiovascular diseases, specifically related to UV-induced damage following sun exposure in women. Polyphenols in GT were shown to protect against many damaging effects of UV radiation, such as sunburn response, immunosuppression, and photo-aging. Flavonoids – EGCG, EGC, and epicatechin (EC) - are easily absorbable and bioavailable upon ingestion with the latter two primarily absorbed in the small intestine and 20-50% recovered in urine. They show that consumption of GT with ~1400 mg of catechins per serving can reduce UV-induced erythema by 16% at 6 weeks and 25% at 12 weeks. Prolonged consumption of GT polyphenols and carotenoids at 6 months decreases overall solar damage and at 12 months reduces UV-induced erythema telangiectasis. This research shows that the body readily absorbs polyphenolic compounds and can use and recycle them before excretion. Future research endeavors should examine the amount of catechins absorbed before excretion and how differently they are absorbed following fermentation.

Green tea extract (GTE), a concentrated form of GT with high concentration of antioxidants, influences working memory modulation by increasing neural connectivity (Schmidt et al. 2014). EGCG increases connectivity from the right superior parietal lobule to the middle frontal gyrus and connectivity is positively correlated with task performance. Protection of cognitive function by EGCG is conducted through antioxidation, iron-chelation, and modulation of cell signalling and cell survival pathways. These actions were shown to reduce oxidative stress induced by neurotoxicity, promote neural plasticity in mice, decrease beta-amyloid levels and plaques in Alzheimer’s mice, and help Calcium-dependent glutamate release in rats. Mice and rats are biologically analogous to humans and the effects shown have the potential to transfer to humans, however there is a need for more research in human subjects. These effects shown in GTE by Schmidt et al. (2014) show effects on neurodegenerative diseases such as Alzheimer’s and Parkinson’s in humans, but more research is needed to decide the validity. Both neurodegenerative diseases have foundations of researchers associated with them, effects of GT and GTK as a health drink and their posed health benefits would be a worthwhile avenue of research.

Obesity is a global problem, and a catechin-rich diet has been shown to decrease intra-abdominal fat in an overweight Asian population (Wang et al. 2010). They show that regular consumption of catechin-rich GT for greater than 90 days led to significant responses in body weight, waist circumference, and the most consistent effect in intra-abdominal fat. Catechin-rich diets have been shown to increase lipolysis during moderate-intensity exercise and decrease intra-abdominal fat (Venables et al. 2008, Wang et al. 2010). GTE was shown to inhibit catechol 0-methyltransferase (5,6) and increase fat oxidation by 17% through lipolysis instead of lipid storage. Venables et al. (2008) show that GTE effects are not limited to fat oxidation. In men, GTE consumption can increase insulin sensitivity by 13% and improve glycemic control, thus reducing the risk of type II diabetes. Another foundation to study the effects of GT and GTK in human models. Alzheimer’s, Parkinson’s, and diabetes disease foundations are researching multiple avenues to help ease these diseases and should devote more research efforts into the effects from GT and GTK.

Several studies have found a positive correlation between GT consumption and cardiovascular health. In a review by Velayutham et al. (2008), they found evidence that GT catechins improve blood lipid profiles, regulate vascular tone, prevent vascular inflammation, inhibit vascular smooth muscle proliferation, and inhibit thrombogenesis. In this review they found that plasma catechin levels were 0.2-2%, showing that bioavailability of catechins is lower than amount absorbed. Despite this limitation in GT, the authors confirm the health benefits in literature with emphasis on cardiovascular health. They show that catechins positively affect plasma lipid profile and vascular function and inhibitory effects on oxidation, vascular inflammation, atherogenesis, and thrombogenesis. GT catechin antioxidant activity scavenges free radicals, chelates transition-metal ions, inhibit pro-oxidant enzymes in favor of antioxidant enzymes. GT clearly has many health benefits and their ability in GTK are likely to still be active, perhaps even be enhanced.

Fermentation of tea with a SCOBY into kombucha provides the drink the health benefits associated with GT and increases antioxidant activity and concentration. This potentially supplies yet unstudied health benefits. Kombucha is a popular beverage for its role as a functional food that supplies vitamins, minerals, organic acids, polyphenols, and enzymes. A commonly touted health benefit of kombucha comes from the microflora of the SCOBY that supply the drink its probiotic effect. Among the *in vitro* and *in vivo* studies reviewed by Kapp and Sumner (2019), they do confirm kombuchas potential to have antimicrobial effects, liver responses, gastrointestinal responses, stimulate the immune system, detoxify the liver, antioxidant activity, anti-tumor activity, and could aid the body against diabetes, cardiovascular disease, and neurodegenerative diseases. Further research about the health benefits of kombucha are needed, especially in human subjects.

## Kombucha Considerations

While fermentation is an effective method to supply flavors and vital nutrients not often obtained in one’s diet (Hsieh et al. 2021), kombucha is influenced by tea type, fermentation time and temperature, and SCOBY microfloral composition. Mentioned previously and clear in the studies discussed here, tea type influences the phenolic profile and antioxidant potential of kombucha. Depending on which quantifying method is used - FRAP assay, DPPH assay, total phenolic content (TPC), or total flavonoid content (TFC) - kombucha tend to follow a trend of greatest values in green tea or pu’er tea, followed by red, white, and then black teas (Jakubczyk et al. 2020, Hsieh et al. 2021). Fermentation time also has a significant effect on TPC and TFC, as fermentation time increases these values were increased (Hsieh et al. 2021). This effect lasts around ten days before the increase plateaus and catechin degradation begins (Jayabalan et al. 2007), however it is less in GTK versus kombucha derived from red, black, or white teas. Hsieh et al. (2021) noticed a decrease in antioxidant potential following the eighth day of fermentation in all tested kombucha. These changes associated with fermentation time are dependent on temperature during fermentation. Lower temperatures decrease mircofloral activity slowing the fermentation process, while higher temperatures increase microfloral activity, hastening the fermentation process, and invite the opportunity of infection from harmful bacteria (Crum and LaGory 2016). Among the microflora that associate with the kombucha SCOBY, each species is active at a different period(s) of fermentation. Yeasts species - *Saccharomyces*, *Brettanomyces*, or *Zygosaccharomyces* - can act in both the aerobic and anaerobic environments associated with kombucha fermentation. SCOBY bacteria can function in either aerobic conditions, *Acetobactor* or *Lactobacillus* species, or in anerobic conditions, such as *Pediococcus* and *Gluconacetobacter* species. A combination of these yeasts and bacteria composes the zoogleal SCOBY film produced with each batch and phenolic composition changes depending on the combination of these microflora [Kurt.2001; Teoh et al. (2004)]. Future research should take these factors into account when designing a beneficial health beverage that can have beneficial effects on neurodegenerative diseases, diabetes, cardiovascular diseases, and the benefits associated with GT.

## Future Research

Future research into the efficacy of kombucha as a health beverage in human experiments is needed. Research should focus on GTK with tea residues fermented for no longer than two weeks at 68-86 degrees F (Crum and LaGory 2016). The effect of tea residue on kombucha was shown by Zhou et al. (2022) to be slightly higher than kombucha fermented without residue. This is due to the SCOBY microfloral enzymes acting on tea residue, thus further extracting the phenolic compounds from the tea. A widespread practice in kombucha brewing is to use a combination of teas to increase the possible health benefits and alter the flavor profile. According to Zhou et al. (2022), kombucha has greater antioxidant ability and concentration than unfermented teas and based on the articles presented here, a combination of Green and Pu’er or Black teas could show promise compared to kombucha derived from one tea type. Another widespread practice when brewing kombucha, is a secondary fermentation in a sealed container to add flavors and carbonation. As noted by Heinrich et al. (2011), GT catechins have a range of effects similar to cocoa polyphenols, an addition of cocoa to kombucha during the secondary fermentation could cause a synergistic effect with GT polyphenols. Other compounds with health benefits could also be used during the secondary fermentation and reactions caused by the SCOBY could have beneficial and/or synergistic effects on phenolic profile. There is, however, a paucity in the literature regarding kombucha health benefits in humans, effect of tea profile, and secondary fermentation changes. It is therefore vital for the food science community to devote more time into researching these effects in humans. Green tea, the second most consumed beverage globally, is well studied and its efficacy as a health drink has concrete evidence in the literature. As a rising globally consumed health beverage, kombucha should be further studied to decide its efficacy as a health drink.

## Conclusion

Green tea is a globally consumed health beverage, second only to water, and is touted for its numerous health benefits, from neurodegenerative diseases to protection from UV-induced skin damage in women. GT has been shown to be anti-carcinogenic, anti-angiogenic, anti-mutagenic, anti-inflammatory, anti-bacterial, hypocholesterolemic, anti-diabetic, and shows protection against Parkinson’s and Alzheimer’s disease (Chacko et al. 2010, Gopal et al. 2016). These health benefits are likely present in kombucha, but human studies are lacking in this regard. As shown by Zhou et al. (2022), fermentation of tea with a SCOBY increases in antioxidant activity x3.25 and polyphenol concentration x5.68 as compared to tea. Rich chemical compounds found in GTK includes organic acids, vitamins, minerals, amino acids and polyphenols, many of which are not often found in food (Crum and LaGory 2016). It is therefore imperative that more research be conducted on the efficacy of GTK as a health drink. Kombucha has the potential to have the same health benefits as GT, perhaps even some health benefits not yet elucidated from GT consumption. The phenolic profiles of GT and GTK indicate elevated levels of catechins, particularly ECG and EGCG, with EGCG being the most active. GT and GTK are highly suggested additions to one’s diet, especially for anyone experiencing elevated levels of stress or those who desire to lose weight.

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