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# Bioactive compounds and antioxidant activity of black and green tea available in Bangladesh

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## Abstract

People in Bangladesh are traditionally used to consume mainly black tea. However, some tea manufacturing companies are now producing green tea, though in a small scale. To create new knowledge as well as awareness about the consumption of green tea, the present study was carried out to compare the black and green tea available in Bangladesh based on their bioactive compounds and antioxidant activity. A total of eight brands of black tea and two brands of green tea were bought from the supermarkets. Total phenolics, total tannin, total flavonoids, and caffeine content were measured as bioactive compounds, and antioxidant activity was evaluated by using two different methods such as DPPH (1,1-diphenyl-2-picrylhydrazyl) radical-scavenging activity and ABTS+ radical scavenging activity of methanol extracts of black and green tea. Every bioactive compound in black and green tea was found to be significantly different ($P < 0.05$). The total phenolic content, on average, was measured at 242.46 mg GAE/g dry extract and 763.41 mg GAE/g dry extract in black and green tea, respectively. Black tea contained 6.47 mg TAE/g dry extract tannin, whereas green tea had much more tannin content, 14.51 mg TAE/g dry extract, which is more than double in amount. On the other hand, the total flavonoid content was almost double in black tea (61.82 mg QE/g dry extract) compared to green tea (31.85 mg QE/g dry extract). Antioxidant activities were determined at different concentrations of tea samples. At every concentration, green tea presented higher ABTS+ and DPPH radical scavenging activity than black tea. The highest percentage of inhibition was observed at 20 ppm both in black and green tea, finding 98.50 % and 99.07 % inhibition, respectively. Overall, significantly ($P < 0.05$) higher amount of phenolic compounds as well as antioxidant activity were observed in green tea.

## 1. Introduction

Tea, the most widely consumed beverage in the world, is remarkable dietary sources of phenolic antioxidants (Shannon *et al.*, 2017). Tea is made basically in three forms. In green tea preparation, the oxidation of fresh leaf polyphenols is precluded. Black tea is produced in such a way as to allow the oxidation almost of all these substances, whereas Oolong tea undergoes partial oxidation. However, teas are quite different from herbal teas, also known as tisanes made from spices, herbs, or other plant materials (Alexieva *et al.*, 2020). In tea leaves, three basic polyphenol groups can be distinguished: catechin, theaflavins and thearubigenes (Yanishlieva-Maslarova and Heinonen, 2001). As green tea skips the oxidation process, the same biologically active components as in fresh leaves remain intact in green tea, though - in various concentrations, except for theaflavins and thearubigens that are mainly abundant in black tea (Rahardiyan, 2019). Tea polyphenols, particularly catechins in green tea, are not only essential in plant physiology but also tremendously beneficial to human health, acting as antioxidants, as well as anti-inflammatory, anti-allergic, antihypertensive, anticancer, and antimicrobial agents

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(Lorenzo and Munekata, 2016; Rahardiyan, 2019). It is evident that different types of teas work synergistically to exhibit benefits to human health, also enhancing consumer acceptance. Likewise, combining teas and tisanes or medicinal drugs were found useful (Malongane *et al.*, 2017).

No other beverage is so popular as tea in Bangladesh. The geographical location of Bangladesh favors the tea climate. Traditionally people in Bangladesh are used to drink black tea. Despite much research on phenolic compounds of tea worldwide, little is known about black and green tea available in Bangladesh, necessitating the study on bioactive compounds and antioxidant properties of both teas to compare them, and eventually, that can act as strong scientific evidence in making awareness about green tea consumption in the country.

## 2. Materials and methods

### *2.1 Sample collection*

Eight brands of black tea such as Ispahani, Taaza, Seylon, Finlays, Kazi and Kazi, Fresh, National, and Tetley and two brands of green tea such as Finlays and Kazi and Kazi were collected from the local supermarket at Sylhet.

### *2.2 Extraction of tea sample*

Tea samples were extracted with methanol/water (80:20, v/v) in a ratio of 3:1. The mixture of tea and methanol was shaken for 48 h using an orbital shaker and then filtered, and the procedure was repeated three times for the maximum extraction. The methanol from the filtrate was removed by a rotary evaporator (R-205, Buchi, Switzerland). Finally, the filtrate was freeze dried to obtain the solid extract for further analysis.

### *2.3 Determination of total flavonoids, tannins and phenolics content*

The aluminium chloride colorimetric method described by (Chang *et al.*, 2002) was used to estimate the total flavonoid content in the tea extracts.

The tannins were determined using the Folin-Ciocalteu Phenol reagent, as reported by (Amorim *et al.*, 2008).

The total phenolics content (TPC) was determined using the modified Folin-Ciocalteu method (Wootton-Beard *et al.*, 2011).

### *2.4 Estimation of caffeine*

Tea caffeine was determined using the method described by Maidon *et al.* (2012).

### *2.5 DPPH and ABTS$^+$ Radical s Scavenging Activity Assay*

The stable DPPH (1,1-diphenyl-2-picrylhydrazyl) radical-scavenging activity was measured using the modified method described by Shimada *et al.* (1992).

ABTS$^+$ radical scavenging activity of tea extract was determined according to the method described by Baltrušaitytė *et al.* (2007) with slight modification.

### *2.6 Statistical Analysis*

IBM SPSS Statistics software, version 20, was used to analyze the data by one-way ANOVA, setting a significant level at $p<0.05$. The mean values were adapted by Duncan's Multiple Range Test (DMRT) (Duncan, 1955).

## 3. Results and discussion

Total Flavonoids, total Phenolic, total Tannin, and caffeine content were analyzed to evaluate bioactive compounds and two complementary methods *viz*. DPPH (1,1-diphenyl-2-picrylhydrazyl) and ABTS$^+$ radicals scavenging activity assays were used to determine antioxidant activity.

### *3.1 Bioactive compounds*

Tea is well known for possessing a wide range of bioactive compounds. In this research work, several bioactive compounds were analyzed to compare the black and green tea, which were summarized in Table 1 as well as in Table 2.

#### *3.1.1 Total phenolic content*

The range of total phenolic content of black tea was found from 224.69 mg GAE/g dry extract to 254.62 mg GAE/g dry extract. The phenolic content of green tea was found 725.52 mg GAE/g dry extract, for Kazi and Kazi and 801.31 mg GAE/g dry extract for Finlays. The highest amount of phenolic content was determined in Finlays green tea, whereas the lowest amount of phenolic content was found in Taaza black tea. Phenolic content in black and green tea varied significantly, and on average, black and green tea contained 242.46 mg GAE/ g dry extract and 763.42 mg GAE/g dry extract, respectively. (Craig, 1999) studied the health-promoting properties of common herbs. He analyzed tea, including others, and found an average total phenolic compound of 248 mg GAE/g dry extract in black tea and 776 mg GAE/g dry extract in green tea. The main active phenolics in green tea are catechins found in fresh

 

Table 1. Bioactive compounds of black and green tea

| Types of tea | Brand of tea | Total phenolic content (mg GAE/g dry extract) | Total tannin content (mg TAE/g dry extract) | Total flavonoid content (mg QE/g dry extract) | Caffeine content (ppm) |
|---|---|---|---|---|---|
| Black Tea | Ispahani | 254.55±2.14$^{f}$ | 6.40±0.15$^{a}$ | 63.60±1.23$^{h}$ | 33.41±1.16$^{f}$ |
| | Taaza | 224.69±2.16$^{a}$ | 6.40±0.20$^{a}$ | 68.53±2.05$^{j}$ | 34.15±1.51$^{gh}$ |
| | Seylon | 253.60±3.18$^{e}$ | 6.68±0.18$^{a}$ | 65.70±2.29$^{e}$ | 30.74±2.08$^{c}$ |
| | Finlays | 254.62±4.08$^{f}$ | 6.41±1.05$^{a}$ | 64.44±2.10$^{i}$ | 31.47±1.11$^{d}$ |
| | Kazi & kazi | 232.41±5.27$^{c}$ | 6.37±1.13$^{a}$ | 61.3±0.13$^{g}$ | 32.60±2.11$^{e}$ |
| | Fresh | 230.67±3.28$^{b}$ | 6.43±1.35$^{a}$ | 59.70±1.12$^{d}$ | 33.83±2.24$^{g}$ |
| | National | 254.57±4.15$^{f}$ | 6.50±1.14$^{a}$ | 60.77±1.19$^{f}$ | 30.65±2.18$^{c}$ |
| | Tetley | 234.55±3.18$^{d}$ | 6.60±1.16$^{a}$ | 60.41±2.15$^{e}$ | 34.41±1.16$^{h}$ |
| Green Tea | Kazi & kazi | 725.52±4.34$^{g}$ | 13.64±1.33$^{b}$ | 33.27±1.17$^{b}$ | 21.30±2.16$^{a}$ |
| | Finlays | 801.31±6.12$^{h}$ | 15.38±1.28$^{c}$ | 30.42±1.42$^{a}$ | 27.30±1.05$^{b}$ |

Values are expressed as mean±SD (n=3), different letters in the same column denote significantly different at P<0.05

Table 2. Comparison of bioactive compound of black and green tea

| Types of Tea | Total phenolic content (mg GAE/g dry extract) | Total tannin content (mg TAE/g dry extract) | Total flavonoid content (mg QE/g dry extract) | Caffeine content (ppm) |
|---|---|---|---|---|
| Black | 242.46±12.42$^{a}$ | 6.47±1.19$^{a}$ | 61.81±3.62$^{b}$ | 32.63±1.47$^{b}$ |
| Green | 763.42± 10.51$^{b}$ | 14.51±1.99$^{b}$ | 31.87±1.58$^{a}$ | 24.36±3.28$^{a}$ |

Values are expressed as mean±SD (n=3), different letters in the same column denote significantly different at P<0.05

leaves, such as epicatechin (EC), epicatechin gallate (ECG), epigallocatechin (EGC) and epigallocatechin gallate (EGCG), which partly undergo oxidation process in black tea to form condensation products like theaflavins, lowering the overall phenolic contents in black tea (Peluso and Serafini, 2017).

*3.1.2 Total tannin content*

The total tannin content of black tea ranged from 6.37 mg TAE/g dry extract to 6.68 mg TAE/g dry extract, whereas green tea contained 13.64 mg TAE/g dry extract (Kazi and Kazi) and 15.39 mg TAE/g dry extract (Finlays). In a study on the antioxidative action of tea polyphenols, the average total tannin content was found 7.59 mg TAE/g dry extract in black tea and 15.16 mg TAE/g dry extract in green tea (Hara, 2001). According to another study (Hodgson and Croft, 2010), total tannin content in black tea ranged from 5.5 to 7.50 mg TAE/g dry extract, and in green tea, it varied between 12 to 15 mg TAE/g dry extract.

*3.1.3 Flavonoid content*

Total flavonoid content was plentiful in black tea, ranged from 59.70 mg QE/g dry extract to 68.53 QE/g dry extract, whilst a lesser amount was observed in green tea: 33.27 QE/g dry extract for Kazi and Kazi and 30.43 QE/g dry extract for Finlays. Thus, a significant difference was observed in the mean flavonoid content of black and green tea, with 61.82 mg QE/g dry extract and 31.85 mg QE/g dry extract in black and green tea, respectively.

*3.1.4 Caffeine content*

The caffeine content of black tea ranged from 30.65 ppm to 34.41 ppm, whereas green tea contained 21.30 ppm (Kazi and Kazi) and 27.30 ppm (Finlays). A significantly higher amount (P<0.05) of caffeine was observed in black tea (32.66 ppm) than that in green tea (24.30 ppm). Chu and Juneja (1997) observed 20 to 35 ppm caffeine on a dry weight basis in green tea. In another study, it was reported that black tea containd 30 to 50 ppm caffeine (Juliano and Griffiths, 2004).

*3.2 Antioxidant activity*

A compound is considered as an antioxidant only if it can delay or inhibit the oxidation of a substrate by forming a stable complex compound or in any other way at a low concentration and antioxidant free radicals generated after the neutralization should be stable (Ahmed *et al.*, 2020). Phenolic compounds having radical scavenging capacity in tea mainly show antioxidant activity. Free radicals play a deadly role in the biological system, and the importance of antioxidants is that they scavenge free radicals by donating electrons or transferring hydrogen atom. There are several methods of determination of antioxidant activity. Of those, two methods like DPPH· and ABTS· radicals scavenging assay were applied in the present study.

*3.2.1 $ABTS^{+}$ radical scavenging activity*

In both assays (ABTS and DPPH free radicals), the reduction of these radicals is estimated to determine the scavenging capacity of a sample. Green tea was observed to exert remarkably superior scavenging activity in both

 

assays. Figure 1 represents that both black and green tea showed more scavenging activity than the standard, and green tea had more antioxidant activity than that of black tea. At different concentrations, the percentage of inhibition by green tea was significantly higher (P>0.05) than that by black tea. The highest mean value of percent inhibition for black tea was found at 98.50% at 20 ppm; likewise, the highest inhibition (99.07%) in the case of green tea was also determined at 20 ppm. The higher TPC in green tea is the main reason to exhibit higher scavenging activity because TPC and scavenging activity is closely correlated (Erkan *et al.*, 2008; Ahmed *et al.*, 2021). The present result is also comparable to other findings (Oh *et al.*, 2013), where green tea scavenged

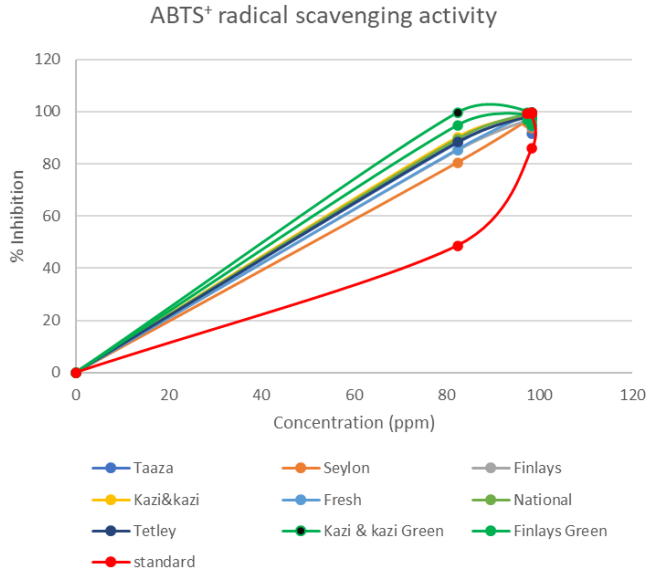


Figure 1. ABTS+ radical scavenging activity of black and green tea with standard

more ABTS· radicals (187.36 mg AAE/g tea) than that (118.53 mg AAE/g tea) of black tea.

### *3.2.2 DPPH radical scavenging activity*

The two assays (DPPH and ABTS) respond differently to antioxidants. DPPH is exclusively soluble in organic media (providing limitation to determine hydrophilic antioxidants); on the contrary, ABTS is soluble in both organic and inorganic media, being capable of estimating both hydrophilic and hydrophobic antioxidants (Awika *et al.*, 2003). Figure 2 illustrates that green tea considerably performed more scavenging activity, but black tea had lesser activity than that of standard as well as green tea. At every concentration, the variance of inhibition of black and green tea was found significant. The highest inhibition for black tea was found 86.70% at 80 ppm, while green tea inhibited most (90.58%) at 100 ppm. The variation in composition, such as phenolic compounds, of plant materials makes the difference in scavenging free radicals. Green tea was reported to have more scavenging effect than black tea in

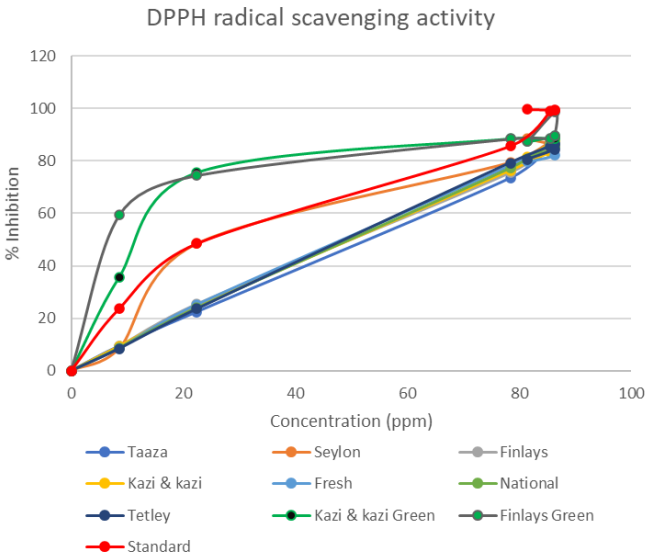


Figure 2. DPPH radical scavenging activity of black and green tea with standard

several previous investigations (Oh *et al.*, 2013; Peluso and Serafini, 2017; Shannon *et al.*, 2017).

## 4. Conclusion

Black and green tea of different brands were compared with respect to bioactive compounds and antioxidant activity. Green teas were found to have more amounts of bioactive compounds except for flavonoid and caffeine and subsequently showed much more antioxidant activity than black teas. In addition, green tea demonstrated significantly higher antioxidant activity than standard ascorbic acid both in ABTS and DPPH free radical scavenging assays. As the green tea exhibited better antioxidant activity than the black tea, this study will help to promote the consumption of green tea in Bangladesh. It is also recommended for further investigation on the feasibility survey on large scale production of green tea in Bangladesh.

## Conflict of interest

The authors declare no conflict of interest.

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