



The effect of brewing time on the antioxidant properties and consumer's preference of green tea and jasmine tea

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KEYWORDS

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ABSTRACT

Jasmine tea is a popular tea type obtained from scented tea (*Camellia sinensis* (L.) Kuntze) leaves with jasmine (*Jasminum sambac* (L.) Aiton) flowers. This study aimed to evaluate the antioxidant properties and sensory aspects of jasmine and green tea brewed at different times. The commercial jasmine and green tea were brewed in boiling water at a ratio of 1:100 for 0.5, 1, 2.5, 5, 10, 30, and 60 min. The obtained tea was evaluated for colour, pH, total flavonoid content (TFC), total phenolic content (TPC), 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activity, and ferric-reducing antioxidant power (FRAP) by standard methods. Thirty-three untrained consumers evaluated the preference for tea from different brewing times. Jasmine tea exerted darker colour, lower pH, higher TFC and TPC, and higher FRAP than green tea. The values of those parameters increased with longer brewing time, with the optimum time being 30 min. However, the longer brewing generated tea liked less by consumers. The most favoured tea was jasmine green tea brewed for 5 min with aroma as the most preferred attribute. Our study suggested that brewing tea for a longer time benefited with better antioxidant properties but disadvantaged in sensory aspects.

Introduction

Tea (*Camellia sinensis* (L.) Kuntze, Theaceae) is widely consumed worldwide for its flavour, health benefits, and safety profile. Among other forms of tea, green tea is popularly associated with antioxidant properties. It does not undergo excessive withering and oxidation processes, resulting in a richer profile of antioxidants (Hu et al., 2018). Green tea and its main constituents have shown promising effects in various oxidative reaction-related health problems, i.e., breast cancer, cognition and brain function, hyperlipidemia, inflammation, photoaging, and etc. (Mancin et al., 2017; Ohishi et al., 2016; Prasanth et al., 2019; Rofida and Hartanti, 2021; Schoeneck and Igman, 2021).

Scented tea is tea leaves mixed with other plant materials during processing to naturally absorb the plant's fragrance. Jasmine tea is prepared from tea leaves and jasmine (*Jasminum*

sambac (L.) Aiton, Oleaceae) flowers. Jasmine-scented tea is commonly prepared from various forms of tea, i.e., green tea, black tea, white tea, and oolong tea. Among those, jasmine green tea is the most popular in China (Li et al., 2018). In Indonesia, jasmine green tea is commercially available and branded as jasmine tea (*teh melati* in Indonesian). Consumers less favoured the plain and somewhat unpleasant aroma of green tea, and the scented process with jasmine flowers greatly improved it (BPS Statistic Indonesia, 2020).

Tea brewing conditions significantly affected the obtained tea's content and bioactivity. Total phenolic content (TPC), epigallocatechin gallate content, and radical scavenging activity of fresh leaves, oolong tea, and green tea varied widely according to the brewing temperature used. In contrast, the brewing time and temperature were significantly affected the caffeine, catechin, free amino acid, and theanine contents of white tea

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(Kowalska et al., 2021; Zhang et al., 2017). However, such information on green tea is still limited. This study evaluated the effects of brewing time on the colour, pH, TPC, total flavonoid content (TFC), antioxidant activities, and sensory aspects of jasmine and green tea. Also, the correlation between antioxidant contents and antioxidant activities of both tea types was analyzed in this current study.

Research Methods

This study was conducted at the Laboratory of Food Bioprocess, Faculty of Engineering and Science, Universitas Muhammadiyah Purwokerto, Indonesia, on January – May 2022.

Materials

Jasmine and green tea samples were purchased from a local supermarket in Purwokerto, Central Java, Indonesia. Jasmine tea comprises 20% jasmine flower and 80% green tea, while green tea is the dried unfermented tea leaves. Both tea types were from the same brand produced by a tea processing plant in Tegal, Central Java. Ethanol, Folin-Ciocalteu reagent, Gallic acid, Quercetin, Trolox, AlCl_3 , CH_3COONa , DPPH (2,2-diphenyl-1-picrylhydrazyl), FeCl_3 , HCl, TPTZ (2,4,6-Tris(2-pyridyl)-s-triazine), and NaOH, all were bought from Sigma-Aldrich (USA), were also used in this study.

Preparation of tea samples

One tea bag (2 g) of each tea type was brewed in 200 mL of freshly boiled water. The teas were allowed to stand for 0.5, 1, 2.5, 5, 10, 30, and 60 minutes before the bag was removed. The brewed teas were allowed to reach room temperature and were freshly analyzed.

Determination of pH

The pH of the teas was measured by a pH meter (Ohaus, USA).

Determination of TPC

TPC of the teas was analyzed according to the standard method in the Indonesian Herbal Pharmacopeia (IHP) with a slight modification (Indonesian MoH, 2017). A reaction mixture of 1000 μL of tea sample or Gallic acid solution and 5000 μL of 7.5% Folin-Ciocalteu reagent was stood for 8 min. The mixture was added with 4000 μL of 1% NaOH and was stood for 60 minutes. The absorbance of the mixture was read at a UV-Vis Spectrophotometer (Shimadzu, Japan) at 730 nm.

The standard curve was prepared from Gallic acid at 0-100 $\mu\text{g}/\text{ml}$, and the TPC was presented as mg Gallic acid equivalent (GAE)/g dry weight (DW) tea.

Determination of TFC

The TFC of the teas was analyzed according to the standard method in the Indonesian Herbal Pharmacopeia (IHP) with a slight modification (Indonesian MoH, 2017). A reaction mixture of 500 μL of tea sample or Quercetin solution and ethanol (1500 μL), 10% AlCl_3 (100 μL), 1M CH_3COONa (100 μL), and water (2800 μL). After 30 minutes, the absorbance was read at 370 nm. The standard curve was prepared from Quercetin at 0-250 $\mu\text{g}/\text{ml}$, and the TFC was presented as mg Quercetin equivalent (QE)/g DW tea.

Determination of DPPH scavenging activity

DPPH scavenging activity of the teas was determined according to the standard method with a slight modification (Thaipong et al., 2006). A reaction mixture of 500 μL of tea sample or Trolox solution and 5000 μL of 25 $\mu\text{g}/\text{ml}$ DPPH was allowed to stand in the dark for 30 minutes. The absorbance of the mixture was read at 517 nm. The standard curve was prepared from Trolox at 0-400 μM , and the DPPH scavenging activity was presented as μM Trolox equivalent (TE)/g DW tea.

Determination of FRAP

FRAP of the teas was determined according to the standard method with a slight modification (Thaipong et al., 2006). Briefly, the freshly mixed 10 parts of 300 mM CH_3COONa buffer, a part of 10 mM TPTZ in HCl, and a part of 20 mM FeCl_3 at the final pH of 3,6 were used as the FRAP reagent. A reaction mixture of 210 μL of tea sample or Trolox solution and 3990 μL of FRAP reagent stood for 30 minutes. The absorbance of the mixture was read at 594 nm. The standard curve was prepared from Trolox at 0-225 μM , and the FRAP was presented as μM TE/g DW tea.

Sensorial analysis

Based on the result of the antioxidant evaluations, both jasmine and green tea brewed for 5, 10, and 30 minutes were subjected to sensory analysis. Tea samples were prepared the same way as those used for the chemical evaluations and antioxidant assays. Thirty-three untrained participants (9 men and 24 women, 19–58 years) who signed informed consent evaluated their preference for tea samples prepared as in the antioxidant properties evaluation. In the monadic presentation, tea was blind-coded

with 4-digit random numbers, and was served to participants. Participants were asked to evaluate aroma, colour, taste, aftertaste, and overall liking. A 4-point hedonic scale, in which one was for extremely disliking and four was for being extremely like, was used to measure those attributes. The questionnaire also contained open-ended questions about the most liked tea sample and its reason.

Data analysis

The data in triplicate were reported as the mean value \pm standard deviation (SD). The effects of tea type and brewing time on TPC, TFC, DPPH radical scavenging activity, FRAP, and each attribute of sensory aspects of teas were analyzed by two-way ANOVA. The mean separation of those parameters between the group was evaluated by one-way ANOVA and the post-hoc Duncan's test. The correlation between TPC and TFC with the antioxidant activities of each tea sample was analyzed by the Pearson correlation test. All statistical analysis was performed in the IBM SPSS Statistics ver 13.0 (IBM, USA) at $p \leq 0.05$.

Results and Discussion

Jasmine tea visually showed a darker colour than the green one. The colour of green tea was pale yellow to pale brown. On the other hand, the colour of jasmine tea was yellow and gradually turned to dark brown with the increasing brewing time (Figure 1). The greenish-yellow to pale yellowish shade of green tea was attributable to the flavonol and flavone pigments (Sakamoto, 1971). Those compounds were soluble in hot water, and the amount extracted increased with prolonged brewing time. Adding jasmine flowers to tea leaves during processing likely contributed to the

development of the reddish-brown colour. Like other flowers, jasmine contains betalain, carotenoid, chlorophyll, and flavonoid pigments (Narbona et al., 2021). Carotenoids consisted of orange carotene and yellow xanthophyll sub-groups and were likely responsible for a darker colour observed in jasmine tea (Fu et al., 2022).

Both types of tea ($p = 0.000$) and brewing time ($p = 0.000$) affected the pH of the tea. Green tea exerted a higher pH than jasmine tea. The longer brewing time resulted in tea with a lower pH. The highest and lowest pH of both teas were observed at 0.5 and 60 min of brewing, with the values of 5.27 ± 0.04 and 5.23 ± 0.01 for jasmine tea and 5.88 ± 0.13 and 5.30 ± 0.06 for green tea (Figure 2). The organic acids, mainly citric, malic, and oxalic, contributed to tea's low pH. The longer brewing time extracted more organic acids and eventually lowered the pH of the tea samples. The teas' pH varied according to the tea type, temperature, and brewing time (Panpan et al., 2013). Our result suggested that the jasmine flower addition significantly lowered the pH of tea. Unfortunately, the data on the acidity or the organic acid content of the jasmine flower was unavailable.

The flavonoids extracted from the tea were affected by tea type ($p = 0.000$) and brewing time ($p = 0.000$). Jasmine tea contained a significantly higher flavonoid content than the green one. The low level of flavonoid content was extracted from jasmine tea in 0.5 min brewing (6.68 ± 0.24 mg QE/g DW), which was increased with the brewing time and reached a maximum after 30 min (34.94 ± 0.47 mg QE/g DW). A similar pattern was observed in green tea, with TFC at min 0.5 and 30 min was 3.98 ± 0.18 and 26.30 ± 1.27 mg QE/g DW, respectively. Interestingly, the TFC of both teas decreased after 60 min brewing (Figure 3).

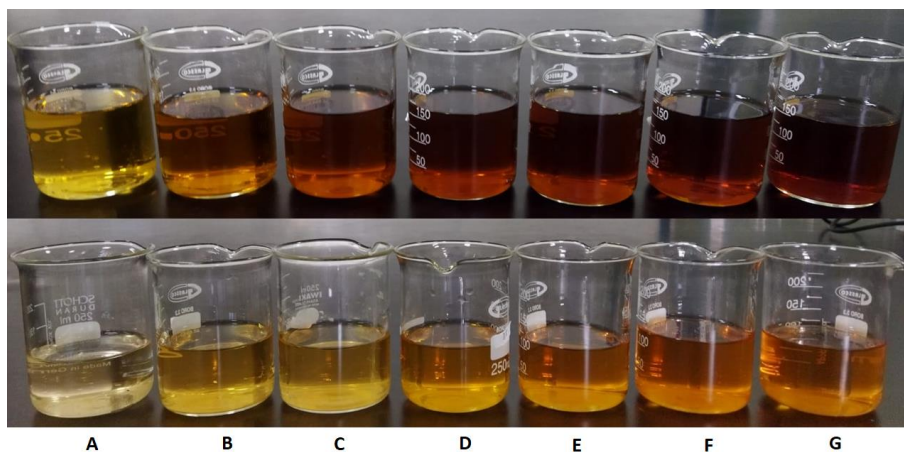


Figure 1. Jasmine (upper panel) and green tea (lower panel) were obtained from different brewing times, i.e., 0.5 min (A), 1 min (B), 2.5 min (C), 5 min (D), 10 min (E), 30 min (F), and 60 min (G)

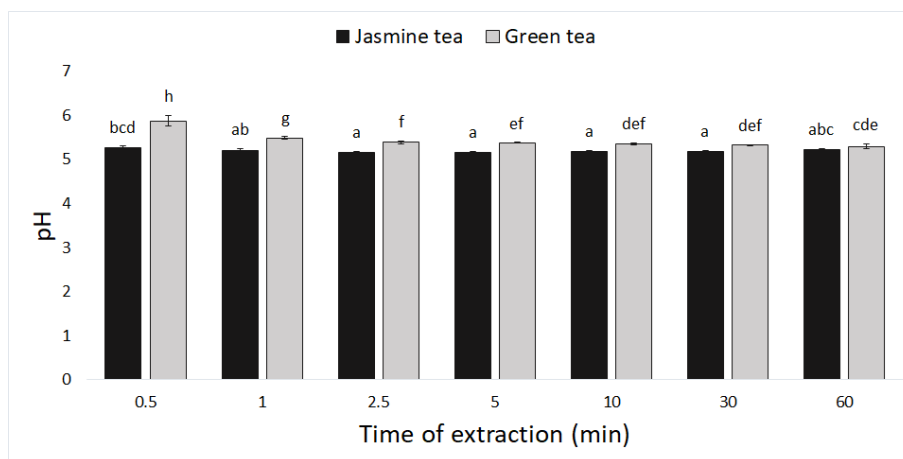


Figure 2. The pH profile of jasmine and green tea was obtained from different brewing times. The different alphabet on each bar represented the statistically different value, tested at a p-value of <0.05

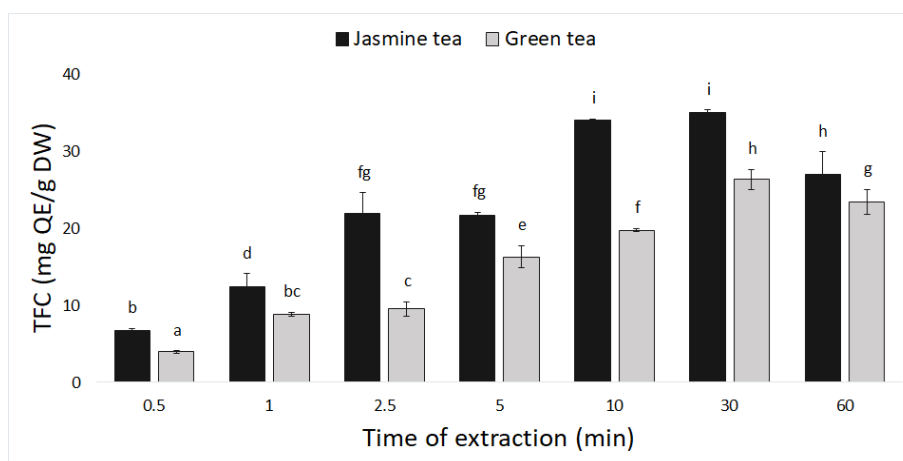


Figure 3. The profile of the TFC of jasmine and green tea is brewed at different times. The different alphabet on each bar represented the statistically different value, tested at a p-value of <0.05

There are three subclasses of flavonoids in tea, i.e., catechins, oligomeric flavonoids, and flavonols. The presence of catechins is dominant in green tea, while oligomeric flavonoids are vastly found in black tea (Dwyer and Peterson, 2013). Catechins and flavonols are soluble in hot water, but the solubility is decreased with the decreasing temperature (Cuevas-Valenzuela et al., 2014). After 30 minutes of brewing, the water temperature dropped, and the flavonoid extraction rate slowed down while the already extracted flavonoids became insoluble. Hence, the TFC of teas in 60 minutes of extraction was lower than those brewed for 30 minutes. The TFC of green tea is mainly affected by the drying method and the maturity of the leaves used. The TFC of green tea collected in Lahijan City (Iran) ranged from 14.3-38.18 mg QE/g DW, according to the different drying methods used (Roshanak et al., 2016). Similarly, Azores (Portugal)-originated green teas prepared

from other parts of tea leaves and twigs exerted TFC of 23.84-72.02 Rutin equivalent (RE)/ g DW (Paiva et al., 2020). Our data suggested that the jasmine flower significantly contributed to the TFC of jasmine tea. Jasmine flowers contain flavonoids, among other phytochemicals (Kunhachan et al., 2012). The methanol extract of jasmine flower from Foshan (China) had a TFC of 4.70 mg RE/g DW, while that of Kuliypitiya (Sri Lanka) was 6.6 mg RE/g DW (Chen et al., 2018; Janarny et al., 2021).

Jasmine and green tea exerted an equal content of phenolic compounds ($p = 0.792$), while brewing time affected their TPC ($p = 0.000$). The TPC of both teas showed a similar pattern to that of TFC. Brewing for 30 min likely generated tea with the optimum TPC, i.e., 84.05 ± 7.93 and 88.66 ± 4.81 mg GAE/g DW for both jasmine and green tea, respectively (Figure 4).

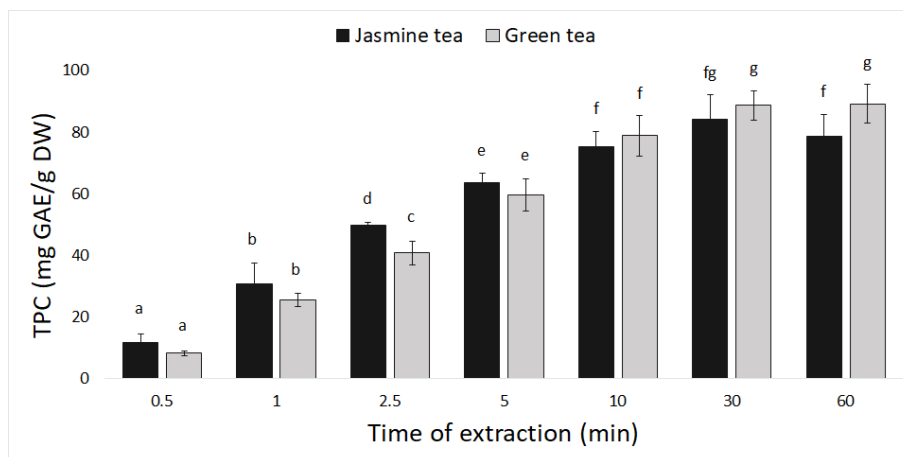


Figure 4. The profile of TPC of jasmine and green tea was obtained from different brewing times. The different alphabet on each bar represented the statistically different value, tested at a p-value of <0.05

The phenolic compounds in green tea were flavanols, flavonols, and phenolic acids (Lorenzo and Munekata, 2016). The solubility of phenolic acids in water is better than those of catechins and flavonols, which are soluble in water at room temperature (Cuevas-Valenzuela et al., 2014; Furia et al., 2021). However, the phenolic acid content in tea is much lower than in catechins, so the TPC of tea brewed for 30 and 60 minutes was statistically equal (Lorenzo and Munekata, 2016). As in TFC, the TPC of green tea vary according to numerous factors. Chinese green tea of various origins showed a TPC of 148.16-252.65 mg GAE/g DW, while those extracted in different parameters were 152-243 mg GAE/g DW (Luo et al., 2020; Zhao et al., 2019). Also, Japanese matcha green tea showed a TPC of 1345.41 ± 238.69 - 1577.01 ± 43.22 , depending on the extraction temperature used (Jakubczyk et al., 2020). On the other hand, the TPC of jasmine green tea was affected by the ratio of tea-to-water used, with values ranging from 1061.0 ± 127 - 3021.6 ± 52.2 mg GAE/l (Koutelidakis et al., 2016). The TPC of jasmine flowers is also widely varied. For example, the flowers collected from Changhua County (Taiwan) showed a TPC of 20.65 ± 0.82 - 182.36 ± 9.23 mg GAE/g DW according to the solvents and extraction methods utilized (Wu et al., 2021). Also, one obtained from China showed a TPC of 9.37 ± 0.38 mg GAE/g DW (Chen et al., 2018). However, the jasmine flower likely did not contribute to the TPC of the jasmine tea in this study. Further studies are needed to understand this phenomenon.

As in the TPC determination, the type of tea did not affect the DPPH scavenging activity ($p = 0.088$), while the brewing time defined the movement ($p = 0.000$). Brewing for 30 minutes

exerted tea with the best radical scavenging activity. DPPH scavenging activity of both teas gradually increased with the brewing time and remained static toward the end of extraction time. The highest scavenging potential was shown by both jasmine (4246.32 ± 394.27 mM TE/g DW) and green tea (5061.91 ± 59.80 mM TE/g DW) extracted for 30 min (Figure 5).

DPPH represents free radicals in the body that, upon release, can disrupt the cell membrane and cause damage to the organelles and the whole cells. The antioxidants can prevent the harmful effects of free radicals. An antioxidant compound donates an electron to the radical and stops the damaging reaction. DPPH assay also evaluates an antioxidant compound's ability to contribute Hydrogen atoms (Craft et al., 2012; Pisoschi et al., 2016; Santos-Sánchez et al., 2019). Green tea typically showed high DPPH scavenging activity. For example, according to the subjected extraction temperature, Japanese matcha green tea showed DPPH inhibition of 12.08 ± 0.58 - $23.48 \pm 5.54\%$ (Jakubczyk et al., 2020). Also, the IC_{50} value of a commercial United Kingdom green tea was 315.41 ± 24.18 μ g/ml (Sirichaiwetchakoon et al., 2020). Individually, green tea leaves exerted a much higher DPPH scavenging activity than jasmine flowers, with quenching values of 94.5 ± 0.15 and $13.0 \pm 1.07\%$, respectively (Tsai et al., 2008). The relatively low DPPH scavenging activity of jasmine flowers did not likely contribute to the activity of jasmine tea. Hence, both tea types showed a similar DPPH scavenging activity in this study.

The type ($p = 0.000$) and brewing time ($p = 0.000$) affected the FRAP of the teas. Jasmine tea exerted a significantly higher reduction capacity than its green counterpart. The activity of both teas

increased with the brewing time. The most suitable brewing time for jasmine tea was 10 min, with a FRAP value of 11470.24 ± 447.17 mM TE/g DW, with the prolonged brewing time did not affect the FRAP. On the other hand, the optimum brewing time for green tea with the highest FRAP was 30 min, with a value of 9178.57 ± 789.11 mM TE/g DW. As in the jasmine tea, a prolonged brewing time of 60 minutes did not favourable affect on green tea's FRAP (Figure 6).

As in the DPPH scavenging assay, the FRAP of green tea varied according to numerous factors. The different brewing temperatures of Japanese matcha green tea exerted FRAP of 5863.03 ± 156.51

- 6129.53 ± 68.40 $\mu\text{M Fe(II)/l}$ (Jakubczyk et al., 2020). Similarly, teas of different origins in China showed a varied FRAP, with the values ranged 2824.80 ± 107.63 - 4099.47 ± 105.10 $\mu\text{M Fe}^{2+}/\text{g DW}$, while those from Korea were 1555.06 ± 20.57 - 1697.81 ± 6.37 mM TE/g DW (Lee et al., 2014; Zhao et al., 2019). On the other hand, the profile of FRAP of jasmine flowers is still limited. The only report available mentioned that it was pretty low and considered insignificant compared to hesperidin (Widowati et al., 2018). Hence, further studies are needed to understand the contribution of jasmine flowers' addition to green tea toward jasmine green tea's FRAP.

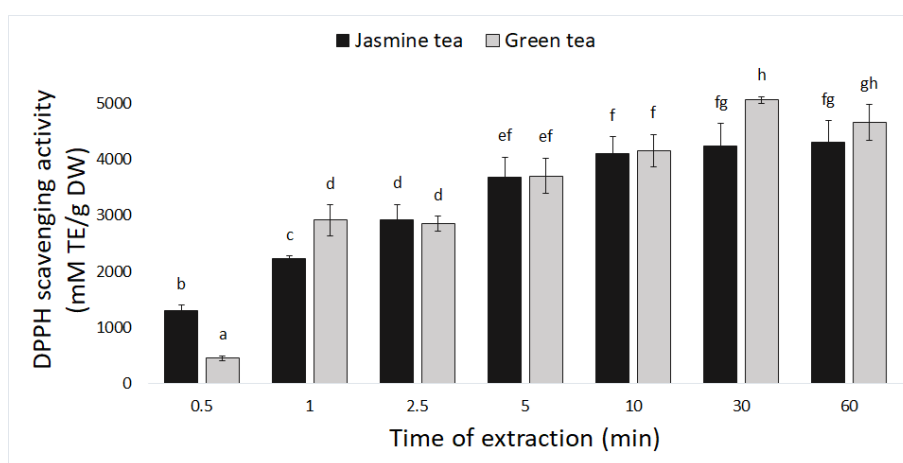


Figure 5. The profile of the DPPH scavenging activity of jasmine and green tea was obtained from different brewing times. The different alphabet on each bar represented the statistically different value, tested at a p-value of <0.05

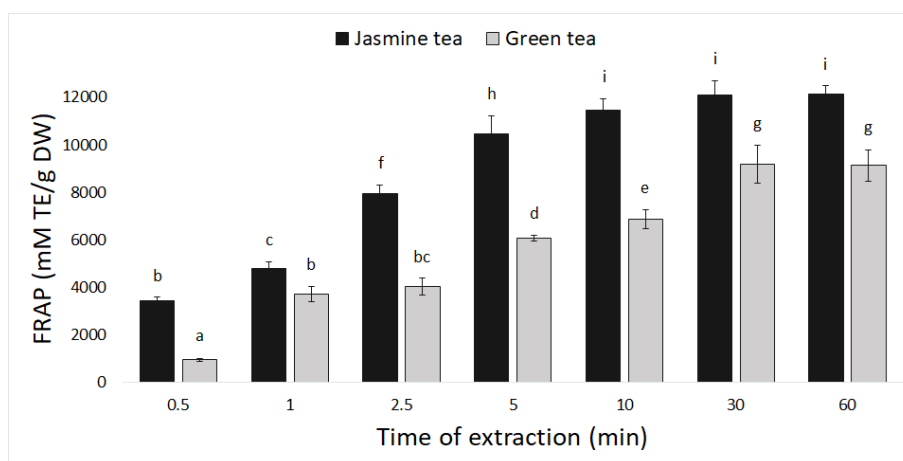


Figure 6. The profile of FRAP of jasmine and green tea was obtained from different brewing times. The different alphabet on each bar represented the statistically different value, tested at a p-value of <0.05

Table 1. Correlation between bioactive content and the antioxidant activity in tea brewed for 39 min

Antioxidant content	R-value			
	Jasmine tea		Green tea	
	DPPH SA	FRAP	DPPH SA	FRAP
TFC	0.964	0.871	0.955	0.899
TPC	0.908	0.939	1.000	0.988

Note: R = Pearson's correlation coefficient, DPPH SA = DPPH scavenging activity

Table 2. The preference scores of jasmine and green tea samples (N = 33)

Tea type	Brewing time (min)	Flavour liking	Colour liking	Aroma liking	Aftertaste liking	Overall liking
Jasmine tea	5	1.88±1.29 ^b	2.82±1.04 ^c	2.67±0.99 ^b	1.97±1.15 ^c	2.30±1.13 ^c
	10	1.64±1.27 ^{ab}	2.64±1.08 ^{bc}	2.70±1.07 ^b	1.81±1.22 ^c	1.88±0.99 ^{bc}
	30	1.88±1.24 ^b	2.52±1.18 ^{bc}	2.73±1.23 ^b	1.72±1.20 ^{bc}	1.94±1.25 ^{bc}
Green tea	5	1.21±1.29 ^{ab}	1.91±1.00 ^a	1.76±1.28 ^a	1.13±1.04 ^{ab}	1.55±1.03 ^{ab}
	10	0.94±1.11 ^a	2.09±1.00 ^{ab}	1.76±1.20 ^a	1.50±1.22 ^{abc}	1.39±1.14 ^{ab}
	30	1.00±1.35 ^a	2.06±1.29 ^{ab}	1.76±1.32 ^a	1.03±1.06 ^a	1.76±3.78 ^a

Note: Different alphabets in each column represented a statistically different value of each sensorial attribute at $p < 0.050$

The correlation evaluation between antioxidant content and the antioxidant activity was subjected to tea extracted for 30 minutes as they showed optimum value for most parameters. TFC was strongly correlated to both DPPH scavenging activity and FRAP. TPC showed the same trend in both antioxidant mechanisms (Table 1). Hence, flavonoids and phenolic compounds were likely responsible for DPPH scavenging activity and FRAP of both jasmine and green tea. Thus, the phenolic compounds were likely the main contributor to the green and jasmine tea's free radical scavenging and ferric-reducing capacities. The antioxidant action of phenolic compounds and flavonoids is mediated by the transfer of hydrogen atoms from the hydroxyl group, which has been well understood as one of the antioxidant mechanisms performed in the DPPH assay. In addition, the double bonds of phenolic compounds might supply a single electron for transferring during the redox reaction, which was the mechanism of antioxidant tested in the FRAP assay (Craft et al., 2012; Pisoschi et al., 2016; Santos-Sánchez et al., 2019). A strong correlation between TPC and FRAP was reported in Sri Lankan commercial green tea products, while that of TPC and DPPH scavenging capacity was said in Romanian commercial ones (Dobrinas et al., 2021; Molan et al., 2009). On the other hand, a strong positive correlation between TFC and DPPH scavenging activity and TFC and FRAP was

reported in Lahijan (Iran)-originated green tea and in Malaysian commercial green tea (Abdullah and Mazlan, 2020; Samadi and Fard, 2020).

A consumer preference test was performed to understand how the different types of tea and brewing time tea affected the consumers liking the tea samples. The type of tea significantly affected the panelists' liking toward flavor ($p = 0.000$), color ($p = 0.000$), aroma ($p = 0.000$), aftertaste ($p = 0.000$), and overall ($p = 0.000$) of the tea. However, the brewing time did not define those attributes, with a p -value of 0.501, 0.905, 0.989, 0.390, and 0.124, respectively. Overall, the panelists' liked the jasmine tea better than the green one. Tea with the highest preference score for flavour, colour, aroma, aftertaste, and overall liking was jasmine tea brewed for 5 min (Table 2). Similarly, the answers to open-ended questions showed that jasmine tea brewed for 5 min was the favourite tea sample of 45.45% of panelists. A total of 46.67% of those who liked jasmine tea brewed for 5 min cited that the attribute they liked most was the refreshing aroma, while 40.00% wanted it for its lovely colour and subtle flavour.

Our result suggested that scenting jasmine flowers to green tea contributed strongly to consumer liking toward aroma. It might be attributable to the pleasant fragrance of jasmine flowers, mainly linalool and benzyl acetate (Chen et al., 2021). Correlating this preference evaluation to our TPC and TFC results, the panelists likely

liked teas with lower flavonoids and phenolic compounds over those with higher contents. Catechins, tea's main flavonoids, and phenolic compounds were previously associated with negative consumer preference due to their bitterness and astringency (Kim et al., 2016).

Conclusion

The influence of the brewing time of jasmine and green tea on their TFC, TPC, antioxidant activity, and consumer preference were investigated in this study. Overall, jasmine tea brewed for 30 minutes generated the highest TFC and TPC as well as the best DPPH scavenging activity and FRAP. However, the consumers like jasmine tea brewed for 5 min most for its refreshing aroma, nice colour, and subtle flavour.

Declarations

Conflict of interests The authors declare no competing interests.

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