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EFFECT OF OVEN AND MICROWAVE DRYING ON POLYPHENOLS CONTENT AND ANTIOXIDANT CAPACITY OF HERBAL TEA FROM STROBILANTHES CRISPUS LEAVES

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ABSTRACT

Objective: This study aimed to evaluate the effect of oven and microwave drying on total phenolic content (TPC), total flavonoid content (TFC), and antioxidant capacity of unfermented and fermented tea developed from *Strobilanthes crispus* leaves.

Methods: TPC and TFC were estimated using a spectrophotometric method, while antioxidant capacity was determined using ferric reducing antioxidant power assay and 2,2-diphenyl-1-picrylhydrazyl radical scavenging assay. *Camellia sinensis* (tea plant), that is, used for the production of all varieties of commercial tea and fresh *S. crispus* leaves were served as controls in this study.

Results: The highest antioxidant activity and TPC were observed in *S. crispus* tea developed from microwave-dried leaves, while the highest TFC was observed in oven-dried tea. Unfermented *S. crispus* tea showed significantly higher values (p<0.05) for antioxidant activity, TPC, and TFC as compared to fermented *S. crispus* tea. A strong and moderate correlation was observed between antioxidant activity and TPC as well as TFC values.

Conclusion: The present work clearly showed that *S. crispus* tea developed from microwave-dried leaves able to preserve the polyphenols and hence contribute to excellent antioxidant capacity. Incorporation of unfermented *S. crispus* tea in the diet can be a good source of natural antioxidant.

Keywords: Green tea, Black tea, Total phenolic, Total flavonoid, 2,2-diphenyl-1-picrylhydrazyl, Ferric reducing antioxidant power assay.

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INTRODUCTION

Tea is one of the most widely consumed beverages in the world since ancient time. It was manufactured from the young leaves of the true tea plant, *Camellia sinensis* [1]. Tea can be found in different forms, depending on the fermentation and preparation methods, for example, black (fermented), green (unfermented), and oolong (semi-fermented) tea [2]. Numerous researches have shown the effectiveness of tea as the preventive agent for toxic chemicals and carcinogens [3,4]. Tea polyphenols are mostly recognized for their antioxidant activities, arising from their ability to scavenge reactive oxygen species [5,6]. The polyphenols bind to metal ions, preventing them from participating in peroxidative reactions [3]. Therefore, tea polyphenols appeared to be associated with reduced risk of cardiovascular diseases (CVD) and cancer as proven by *in vitro* and *in vivo* studies [4]. Recently, many researchers have focused their investigation on developing herbal teas from potential plants other than *C. sinensis* [7].

Strobilanthes crispus (L.) Bremek is known locally as "*pecah kaca*" or "*jin batu*" in Malaysia or "*daun picah beling*" in Jakarta. It is local to nations from Madagascar to Indonesia and Malaysia [8]. *S. crispus* contain different phytochemical groups, including polyphenols, alkaloids, vitamins, and minerals [9]. The mixture of these compounds may exert a positive impact on the chronic diseases such as hypertension, CVD, and cancer [9,10]. Numerous researches have been conducted on the bioactivity of ethanolic extract of *S. crispus* herbal tea, and it has been shown that it possesses anticancer, antidiabetic [11], and antibacterial properties [12].

Tea manufacturing processes involved withering, steaming, blending, and drying [7,13]. These processes can exert a strong impact on the

stability of the bioactive compounds, especially the polyphenols. Drying, specifically act as a preservation method, where the function is to lower the water activity to inhibit the growth of microorganisms [14]. However, it is also involved in the enzymatic and non-enzymatic processes that may lead to significant changes in the composition of phytochemicals in the plants [15].

Drying can be performed by traditional sun drying or conventional oven drying [16]. Various researches have reported that these drying methods can bring adverse effects to the antioxidant activity of the plant products [17-20]. Meanwhile, microwave technology has been considered as the best drying method for food products due to uniform drying and better quality of the products [21]. Microwave drying may exert little or no significant losses or enhancement in antioxidant properties compared to the oven drying method [7]. These two methods (microwave and oven drying) can be easily performed to produce herbal tea in the domestic setting. However, to the best of the author's knowledge, there is limited information available on the effect of microwave drying on the polyphenols content of unfermented and fermented tea. Therefore, this study was conducted to evaluate the effect of microwave and convection oven drying on total phenolic content (TPC), total flavonoid content (TFC), and antioxidant capacity of unfermented and fermented S. crispus tea.

METHODS

Materials

All reagents and chemical used were analytical grade: Absolute ethanol (Merk, Darmstadt, Germany), Folin–Ciocalteu reagent (Merk, Darmstadt, Germany), sodium bicarbonate (Merk, Darmstadt, Germany), aluminum chloride (Merk, Darmstadt, Germany), potassium acetate (Merk, Darmstadt, Germany), gallic acid (Sigma Chemical Co., St Louis, MO, USA), quercetin (Sigma Chemical Co., St Louis, MO, USA), butylated hydroxytoluene (BHT) (Sigma Chemical Co., St Louis, MO, USA), and 2,2-diphenyl-1-picryl-hydrazyl (DPPH) (Sigma Chemical Co., St Louis, MO, USA).

Sample preparation

The fresh leaves of *S. crispus* were collected from Sg. Ramal, Selangor, Malaysia. The young leaves were selected from the plants (the apex of the 5^{th} leaf). Then, the leaves were washed thoroughly with tap water and rinsed with distilled water. The commercial green tea (*C. sinensis*, Premium) was purchased from the market and used as a control. All the samples were stored at - 20°C until extraction and analysis.

Preparation of unfermented S. crispus tea

The preparation of unfermented *S. crispus* tea was based on the preparation of *C. sinensis* green tea as described by Rasmussen and Rhinehart [13]. The method consists of four steps which are withering, steaming, blending, and drying. In the drying process, two methods were used: Convection oven and microwave drying. In the convection oven drying, the sample was dried for 10 min at 95°C–100°C or until dry. While for microwave drying, the sample was spread on the tray in a standard domestic microwave oven (Samsung 900 W). The leaves were dried at 900 W for 2 min [22].

Preparation of fermented S. crispus tea

The preparation of fermented *S. crispus* tea was based on the preparation of *Camellia theifera* black tea as described by Adisewojo [23]. The method consists of four steps which include withering, rolling/blending, fermentation, and drying. In the drying process, convection oven and microwave drying methods were used as previously mentioned in the preparation of unfermented *S. crispus* tea.

Sample extraction

The extraction of all types of tea was based on the method by Gadow *et al.* [24]. Two grams of the sample was extracted by pouring boiling distilled water (40 ml) into the beaker containing the tea. The solution was mixed by stirring with a wire shaker for 30 min and then centrifuged for 10 min at 40 rpm. The mixture was allowed to cool and subsequently filtered. The filtrate was used for determination of TPC, TFC, and antioxidant capacity. Three replications were done for each analysis (n=3).

Determination of TPC

The Folin–Ciocalteau assay was used to determine the TPC as described by Velioglu *et al.* [25]. The Folin–Ciocalteau reagent was diluted 10-fold with distilled water. Then, 2.25 ml of Folin–Ciocalteau reagent solution was mixed with 300 μ l of extract. The solution was mixed using vortex and then allowed to stand for 5 min at room temperature (24°C), followed by the addition of 2.25 ml of sodium bicarbonate (60 g/l) solution to the mixture. The mixture was incubated for 90 min at room temperature (24°C). Triplicate measurements were carried out and the absorbance was measured at 725 nm using a spectrophotometer. TPC was quantified using a calibration curve obtained by measuring the absorbance of the known concentrations of gallic acid solutions. The results were calculated as gallic acid equivalent (GAE)/g of dry extract.

Determination of TFC

TFC was determined according to the aluminum chloride colorimetric method as described by Ling and Tang [26]. About 0.1 ml of 10% aluminum chloride and 0.1 ml of 1 mol/l potassium acetate was mixed with the tea extract (0.5 ml), ethanol (1.5 ml), and distilled water (2.8 ml). After 40 min of incubation at room temperature (24° C), the absorbance was measured using a spectrophotometer at 415 nm. Quercetin was used as a standard (the concentration range: 0.015–0.9 mg/ml) and the flavonoid content was expressed as milligram QE/g of dry extract. Analysis of sample extract and standard were conducted in triplicate (n=3).

Determination of antioxidant capacity

DPPH free radical scavenging assay

DPPH free radical scavenging assay was done according to the method by Nurul and Asmah [27]. An aliquot of 400 μ l of samples or control (distilled water) or BHT (50 mg/l and 200 mg/l) was mixed with 1600 μ l of tris-HCI buffer. The mixture was then added to 2 ml of 500 μ mol DPPH in absolute ethanol. The mixture was shaken vigorously and allowed to stand at room temperature (24°C) for 20 min in the dark. The mixture was measured using a spectrophotometer at 517 nm using ethanol (100%) and DPPH as blank. The analysis was performed in triplicate for sample extract and standard. The free radical scavenging activity was calculated using the formula below:

Scavenging effect $(\%) = 1 - \frac{absorbance of sample}{absorbance of control} \times 100$

Ferric reducing antioxidant power (FRAP) assay

FRAP assay was determined using the method from Benzie and Strain [28]. The working FRAP reagent was prepared by mixing 300 mM acetate buffer (pH 3.6), 10 mM 2, 4, 6 –trispyridyl-s-triazine solution, and 20 mM FeCl₃. $6H_2O$ in a 10:1:1 ratio before use. Then, the FRAP reagent was heated at 37°C in water bath for 10 min. After that, 3 ml of FRAP reagent was added into the cuvette and blank reading was taken at 593 nm using a spectrophotometer. A total of 100 µl sample extract and 300 µl distilled water was added to the test tubes. The mixture was incubated for 4 min and the second absorbance reading was performed at 593 nm. The FRAP values were determined by the change in the absorbance value after 4 min from the initial blank reading. Standards of known Fe sulfate (ferrous sulfate heptahydrate) concentrations were prepared using several concentrations from 1 to 10 mM. FRAP value was expressed as µmol of ferrous equivalent/g of dry extract.

Statistical analysis

All analyses were performed using MINITAB version 16. The experimental results were reported as a mean \pm standard deviation. Analysis of variance (ANOVA) with a *post hoc* test (Fisher) was used to determine the significance of differences for multiple comparisons at p<0.05. The Pearson's correlation coefficient (*r*) was used to determine the correlation between total phenolic and TFC and antioxidant activity DPPH free radical scavenging assay and FRAP assay). Differences at p<0.05 were considered statistically significant.

RESULTS AND DISCUSSION

We investigated the antioxidant components (TPC and TFC) and antioxidant capacity of *S. crispus* herbal tea. In this work, two types of tea (unfermented and fermented) were developed from *S. crispus* leaves that were dried using microwave and oven drying method. As previously discussed, the procedures chosen to dry the sample may result in the retention of most of its biofunctional components or otherwise, the procedures may cause a significant loss of the bioactive compounds [27]. The *S. crispus* leaves were dried using the selected methods and extracted using hot boiling water extraction to mimic the household preparation and brewing conditions of herbal tea [29].

TPC and TFC

The TPC and TFC of the samples were presented in Table 1. TPC was expressed as mg GAE/g of dry extract, while TFC were expressed as mg QE/g of dry extract. *S. crispus* tea was compared to controls and results showed that *S. crispus* tea had significantly lower TPC than the green tea (*C. sinensis*, Premium). Microwave-dried unfermented tea had the highest TPC (41.94 mg GAE/g) followed by oven-dried unfermented tea (24.68 mg GAE/g); microwave-dried fermented tea (15.96 mg GAE/g) and oven-dried fermented *S. crispus* tea (6.53 mg GAE/g) (Table 1).

Similarly, the previous study had reported the same trend of TPC in *S. crispus*, but with lower values [11]. The difference results obtained from the previous study may be attributed to the variation of TPC with

the different climate, season, horticulture practices [30], or the storage condition and the time gap before the samples being analyzed [27]. The largest component in phenolic compounds is flavonoid. In the present study, the estimation of TFC was based on the reaction between aluminum trichloride and the hydroxyl group of flavones and flavonols as well as ortho-dihydroxyl groups of flavonoids [31]. The reactions lead to color development that can be measured using a spectrophotometer.

Table 1 showed the highest TFC was observed in the commercial green tea (C. sinensis, Premium) (86.51±11.80 mg QE/g followed by unfermented S. crispus tea, fermented S. crispus tea, and fresh S. crispus leaves. Analysis from one-way ANOVA showed that the TPC and TFC of unfermented S. crispus tea were significantly higher (p<0.05) than fermented S. crispus tea (Table 1). This result may be explained by the fact that the oxidation of polyphenols during the fermentation stage contribute to lower quality of the food product [32]. Hence, fermented tea had lower polyphenol content than that of unfermented tea. Besides, heat treatment (steaming) was applied during the production of unfermented tea, causing deactivation of the enzyme that catalyzes their oxidative polymerization. Thus, it prevented the tea polyphenols from being damaged during the process [33]. On the other hand, the fermented tea had undergone a fermentation step which caused the disruption of the leaf cells and promotes enzymatic oxidation of the flavanols to produce polymeric flavonoids [34].

Microwave-dried unfermented S. cripus tea had significantly higher TPC (p<0.05) as compared to oven-dried S. cripus tea (Table 1). These results confirm the findings of Valadez-Carmona et al. [35] who found higher retention of phenolic compounds in microwave-dried Cocos nucifera as compared to oven-dried sample. During microwave drying, the heat generated from the microwave has high energy and able to inactivate the degradative enzymes at much faster rate than the conventional techniques [36]. The plant tissues were also destroyed after microwave drying causes the release of bound phenolic compounds [14]. Besides, the moisture content in the sample plays a significant role in antioxidant and bioactive components analysis as its influence the penetration of solvent and chemicals into the food matrix. Ismail et al. [9] found that the moisture content of fresh S. crispus leaves was 69.3±0.1%, while microwave and oven drying decreased the moisture content in the range of 8.5-4.4%, respectively [35]. As shown in Table 1, fresh S. crispus leaves and fermented S. crispus tea prepared using oven-dried leaves had the lowest TPC (6.53±2.53 and 6.58±1.80 respectively). Similarly, Chan et al. [37] reported a significant decrease in the TPC of ginger leaves after sun- and oven-drying. During microwave drying, the moisture evaporated at higher rate which could be due to the increase in internal temperature and greater vapor pressure gradient [38]. These conditions assisted the released of bioactive components from the samples. Thus, microwave drying improves the final quality of S. cripus tea at a shorter drying period.

On the other hand, statistical analysis showed there was no significant difference in TFC between microwave-dried and oven-dried for both types of *S. crispus* tea (p>0.05) (Table 1). It is important to note that the chemical changes that occur after drying are due to the complex mechanism [39]. Therefore, it is always challenging to forecast the changes even though both phenolic and flavonoid compounds are affected by food processing [40]. The type of polyphenols and their location in the plant cells may also influence the increase or decrease of TPC and TFC after the drying process [39]. For instance, the major polyphenolic compounds in *C. sinensis* are the flavan-3-ols called catechin [41] while the major flavonoid compound in *S. crispus* is quercetin [9].

Antioxidant capacity

DPPH free-radical scavenging assay

The antioxidant activity of the samples was assessed using DPPH free radical scavenging activity and the values were compared to synthetic antioxidant, BHT, and commercial green tea (*C. sinensis*, Premium). The value of free radical scavenging activity (%) was corresponded to

antioxidant activity of the sample. As can be seen from Table 2, 200 mg/l BHT had the highest radical scavenging activity (74.42 \pm 2.50%) followed by the commercial green tea (65.43 \pm 0.31%), unfermented *S. crispus* tea (61.17 \pm 0.74% and 58.27 \pm 0.48% for microwave-dried and oven-dried leaves, respectively), fresh *S. crispus* leaves and fermented *S. crispus* tea (p<0.05) (Table 2). Microwave drying of the leaves displayed higher radical scavenging activity than oven drying method (p<0.05). Fresh *S. crispus* leaves showed similar radical scavenging activity with BHT at 50 mg/l (51.73 \pm 3.36 and 48.39 \pm 0.62 respectively) (p>0.05) (Table 2).

The data from the present study were consistent with those of Mohd-Fadzelly *et al.* [11] who found that the unfermented tea had higher antioxidant activity compared to fermented tea. Another study by Muslim *et al.* [42] who investigated the antioxidant activity of methanolic and aqueous extracts of *S. crispus* leaves using DPPH free radical scavenging assay showed moderate antioxidant properties compared to controls which were evidenced by the quenching of the DPPH radicals. Gallic acid, quercetin, BHA, and ascorbic acid displayed potent DPPH free radical scavenging activity which produced EC_{50} values of 12.6, 15.3, 21.9, and 25.5 µg/ml, respectively. Similarly, Suhaila *et al.* [43] revealed that the aqueous extracts of *S. crispus* leaves had higher antioxidant activities of the extracts were considered to be lower than that of gallic acid. The difference values obtained from the previous study may be attributed to different sample preparation and extraction method.

FRAP assay

For FRAP assay, a similar trend was observed, whereby the commercial green tea and unfermented *S. crispus* tea prepared using microwave-

Table 1: TPC and TFC of unfermented and fermented *S. crispus* tea

Sample	TPC (mg GAE/g)	TFC (mg QE/g)
Unfermented S. crispus tea		
Microwave-dried	41.94±16.96 ^b	34.68±1.08 ^b
Oven-dried	24.68±5.24 ^c	36.00±2.31 ^b
Fermented S. crispus tea		
Microwave-dried	15.96±4.32°	22.43±1.09°
Oven-dried	6.53±2.53 ^d	21.52±1.78°
Green tea	228.48±4.86ª	86.51±11.80 ^a
(C. sinensis, Premium)		
Fresh S. crispus leaves	6.58±1.80 ^d	1.14±0.08 ^d

Results are presented as mean±SD (n=3). For each column, values followed by the different letters in superscripts (*d) are statistically different at *P*<0.05. SD: Standard deviation, TPC: Total phenolic content, TFC: Total flavonoid content, *S. crispus: Strobilanthes crispus, C. sinensis: Camellia sinensis*, GAE: Gallic acid equivalent

Table 2: DPPH free radical scavenging activity of unfermented and fermented S. crispus tea

Sample	DPPH free radical scavenging activity (%)
Unfermented S. crispus tea	
Microwave-dried	61.17±0.74 ^b
Oven-dried	58.27±0.48 ^c
Fermented S. crispus tea	
Microwave-dried	30.50±4.52 ^e
Oven-dried	15.44±0.39 ^f
Green tea (C. sinensis, Premium)	65.43±0.31 ^b
Fresh S. crispus leaves	51.73±3.36 ^d
BHT 50 mg/l	48.39±0.62 ^d
BHT 200 mg/l	74.42±2.50 ^a

DPPH: 2,2-diphenyl-1-picrylhydrazyl free radical scavenging assay. Results are presented as mean±SD (n=3). For each column, values followed by different letters in superscripts (^{a-f}) are statistically different at *P*<0.05. *S. crispus: Strobilanthes crispus, C. sinensis: Camellia sinensis,* BHT: Butylated hydroxytoluene

dried leaves had the highest antioxidant activity (Table 3). Then, it was followed by oven-dried unfermented *S. crispus* tea and fermented *S. crispus* tea, while fresh *S. crispus* leaves showed the lowest antioxidant activity (p<0.05) (Table 3). ANOVA showed that microwave-dried leaves had significantly higher (p<0.05) FRAP values compared to oven-dried leaves of unfermented *S. crispus* tea (p>0.05) (Table 3).

In this study, the effect of drying methods on unfermented and fermented tea was investigated. Analysis showed that there were significant differences on antioxidant activity (p<0.05) assayed by DPPH and FRAP between microwave-dried and oven-dried leaves for unfermented but not in fermented S. crispus tea. These results are in agreement with Chan et al. [44] who reported that microwave-dried green tea showed outstanding DPPH (IC_{ro} 0.015 mg/ml; AEAC 26,213 mg AA/100g) and higher FRAP values (123.0 mg GAE/g) compared to other commercial green tea. This result indicated that there are increases in antioxidant activity of tea using microwave drying compared to the conventional oven drying. This may be attributed to higher retention of phenolic compounds. As previously discussed, the intense heat was generated in microwave drying from the volumetric heating where the microwave energy was absorbed directly and internally by the plants and then converted into heat [45,46]. This heat resulted in disruption of the plant cell wall polymers due to the production of high vapor pressure and temperature inside the plant tissues. Therefore, more phenolic could be released [47]. Meanwhile, the heat from hot air oven drying resulted in the vaporization of volatile compound that causes the loss of compound in the water vapor [48]. Besides, inefficient denaturation of enzyme makes the initial enzyme degradation of antioxidant compound, thus significantly decrease in antioxidant activity of oven drying [49].

Previously, Yen and Chen [50] found that the antioxidant activity in the tea extract decreased in the order of semi-fermented tea > unfermented tea > fermented tea. Fermentation causes oxidation or heat exposure toward the phytochemicals such as flavonol glycosides, caffeine, saponin, and ascorbic acid [51]. This might lead to reduction of antioxidant activity during tea fermentation [52]. It may be inferred that increased in scavenging activity of fresh *S. crispus* leaves despite of lower phenolic compounds compared to fermented tea could be the result of the preservation of antioxidants vitamins such as ascorbic acid.

Correlation between TPC, TFC, and antioxidant capacity of samples using DPPH radical scavenging and FRAP assays

The correlations between the assays were analyzed using Pearson correlation coefficient and r-values were presented in Table 4. The antioxidant activity for both assays, TPC and TFC were significantly correlated to each other. The r-values were as follows: DPPH/FRAP r=0.878; TPC/DPPH r=0.551; TPC/FRAP r=0.809; TFC/DPPH r=0.640; TFC/FRAP r=0.861; and TFC/TPC r=0.960 (Table 4). This could be attributed to the similar mechanistic basis for DPPH and FRAP assays which is the transfer of electrons from the antioxidant to reduce the oxidant [53].

The results of the present study indicated that high antioxidant activity is associated with high phenolic and flavonoid content. These results are consistent with those of Sudha et al. [54] who reported that both phenolics and flavonoid are categorized as a potential antioxidant. The antioxidant activity of phenolic compound could be explained by its unique structure and a high tendency for metal chelation as well as their redox properties [55]. Phenolic compounds contain at least one aromatic ring (C6) that bears one or more hydroxyl groups. These allow them to act as reducing agents, hydrogen donators, and singlet oxygen quenchers [56,57]. Flavonoid compounds are considered to be the largest group of naturally occurring phenol and their antioxidant capacity are depending on the position of OH group [58]. Therefore, the antioxidant activity of plant extracts also comes from the presence of other antioxidant secondary metabolites, such as flavonoids, proanthocyanidins, anthocyanins, and not limited to phenolics content [59,60].

 Table 3: Antioxidant activity of unfermented and fermented
 S. crispus tea

Sample	FRAP value (μ mol FE/g of dry extract)
Unfermented	
S. crispus tea	
Microwave-dried	7320.05±326.28 ^b
Oven-dried	6010.85±101.13°
Fermented S. crispus	
tea	
Microwave-dried	2434.82±189.63 ^d
Oven-dried	2332.43±231.28 ^d
Green tea	9993.65±130.26ª
(C. sinensis, Premium)	
Fresh S. crispus leaves	1059±95.0°

FRAP: Ferric reducing antioxidant power assay. Results are presented as mean±SD (n=3). For each column, values followed by different letters in superscripts (^a·e) are statistically different at *P*<0.05. FE: Ferrous equivalent, SD: Standard deviation, *S. crispus: Strobilanthes crispus, C. sinensis: Camellia sinensis*

Table 4: Pearson's correlation coefficient of TPC and TFC of *S. crispus* tea with their antioxidant activities

Analysis	DPPH assay	FRAP assay	ТРС	TFC
DPPH assay	1			
FRAP assay	0.878**	1		
TPC	0.551*	0.809**	1	
TFC	0.640**	0.861**	0.960**	1

**Correlation is significant at P<0.01, *Correlation is significant at P<0.05. DPPH: 2,2-diphenyl-1-picrylhydrazyl free radical scavenging assay, FRAP: Ferric reducing antioxidant power assay, TPC: Total phenolic content, TFC: Total flavonoid content, *S. crispus: Strobilanthes crispus*

CONCLUSION

In conclusion, unfermented *S. crispus* tea exhibits excellent antioxidant activity and is a potential source of phenolic compounds. Microwave drying of *S. crispus* leaves produced the highest antioxidant activity and TPC compared to the oven-drying method. This suggested that microwave drying can be used as an alternative drying method due to successful extraction of phenolic compound and energy and time efficient as well as affordable for household use compared to the oven-drying method. However, the variation in drying conditions and moisture content may induce stresses inside food components and subsequently affect the biofunctional compounds in the food products. Other factors such as the economy and consumer preference and acceptability should also be considered. Therefore, further study to determine the optimum drying conditions and sensory quality parameters of the tea such as color, flavor, and fragrance should be carried out.

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AUTHORS CONTRIBUTION

Nur Fatimah Lasano: Designed and conducted the experiments, prepared the manuscript. Asmah Rahmat: Designed the experiments and revised the manuscript. Nurul Shazini Ramli: Analyzed the data and prepared the manuscript. Mohd Fadzelly Abu Bakar: Designed the experiments and revised the manuscript. All authors have read and approved the final manuscript.

CONFLICT OF INTERESTS

All authors have none to declare.

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