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Effect of Steaming and Moisture Reduction Process on Phytochemical Content, Physicochemical and Microbiological Qualities of White (Sang Mon) Bamboo (*Dendrocalamus sericeus*) Leaf Tea

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Abstract

Bamboo leaves are a rich source of phytochemicals and antioxidants, traditionally used to support immune health and alleviate chronic conditions. This study aimed to develop high-phytochemical-content bamboo leaf tea by optimizing the tea processing steps, specifically examining steaming times and moisture reduction methods, roasting and hot air drying. Results revealed that steaming white bamboo (*Dendrocalamus sericeus*) leaves for 15 minutes produced the highest total phenolic content (TPC) at 14.97 ± 0.05 mg GAE/g. In terms of moisture reduction, roasting for 30 minutes resulted in the highest TPC, total flavonoid content (TFC), and catechin levels (11.73 ± 0.07 mg GAE/g, 9.46 ± 0.19 mg QE/g, and 19.10 ± 0.12 mg/100 g, respectively), though isovitexin was undetectable. Conversely, hot air drying for 30 minutes preserved higher levels of orientin (71.41 ± 0.01 ppm), isoorientin (36.73 ± 0.01 ppm), and isovitexin (166.61 ± 0.00 ppm). Vitexin was not detected in either method. Both methods effectively reduced moisture to below 10%, aligning with Thai Ministry of Public Health standards for tea infusions. Microbiological assessments confirmed that the processed tea met Thai community standards for dried herbs. Importantly, brewed bamboo leaf tea contained no detectable caffeine, making it suitable for consumers seeking stimulant-free alternatives. The optimal production process was identified as steaming for 15 minutes followed by hot air drying at 60°C for 30 minutes. This approach not only enhances phytochemical retention but also offers a sustainable strategy for utilizing bamboo foliage, often discarded during culm processing, as a valuable resource in the herbal tea industry. Overall, this study underscores the importance of process optimization in preserving functional compounds and supports community-based herbal tea production as a viable and health-promoting enterprise.

Keywords**:** *bamboo leaf; herbal teas; Camellia sinensis, isovitexin; orientin; Sang Mon; vitexin; ornamental medicine*

1. Introduction

Herbal teas (herbal infusions) are among the most widely consumed non-alcoholic beverages globally, valued for their high-quality flavor and health benefits. Most are region-specific and are typically brewed from the leaves, flowers, seeds, fruits, stems, or roots of various plants, excluding the traditional tea leaves of *Camellia sinensis* (Onaolapo, & Onaolapo, 2019). The global herbal products market is projected to grow by 110% over the next decade. In Thailand, the number of businesses processing herbs into food products (such as herbal tea and ready-to-drink herbal beverages) and medicines has steadily increased. In the post-COVID era, the domestic herbal market has grown at an average annual rate of 13%, with sales having

doubled. As of 2024, there are 1,778 active operators in this sector, representing investments totaling at least 476 million USD (Business Data Processing and Analysis Section, 2024). These trends underscore the growing importance of herbal teas as a health-oriented beverage both locally and globally.

In this context, it was reported that bamboo leaf was used as an ornamental medicine for cough, fever, and leprosy, and its extract exhibited multiple effects such as antioxidant, free radical scavenging, antimicrobial, anti-inflammatory, anti-aging, and cardioprotective properties (Das, 2019; Singhal, 2024; Tundis et al., 2023; Kasemsukphaisan et al., 2023). Its excellent antioxidant capacity was attributed to the presence of several phenolics and flavonoids (Gong et al., 2014). The main antioxidants included phenolics (protocatechuic acid, p-hydroxybenzoic acid, catechin, syringic acid, p-coumaric acid, chlorogenic acid, caffeic acid, ferulic acid), flavonoids (orientin, isoorientin, vitexin, homovitexin, tricin), and vitamins C and E (Nirmala et al., 2018). Accumulating evidence indicated that four C-glycosyl flavonoids (orientin, isoorientin, vitexin, and isovitexin) were the main pharmacologically active components (Cheng et al., 2023). It was also suggested that bamboo leaf and its infusion contained higher levels of vitamins C and E and phytosterols than green tea (Indira et al., 2022). In China, it was reported that bamboo wine, rich in water-soluble B vitamins, essential and non-essential amino acids, and various antioxidants, was made from bamboo leaves (Sangija, & Wu, 2022). In addition, the antioxidant extract from bamboo leaves was approved by the Food Additive Standardization Committee of the People's Republic of China on December 28, 2003. It can be used in edible oils, meat products, aquatic products, and other foods, and has been listed in the state standard GB-2760 (Hygienic Standards for Food Additives in Use) since April 2004 (Singhal et al., 2011). In recent years, bamboo leaf tea has also gained popularity in numerous Western countries (Horn, & Häser, 2016). Thus, processing bamboo leaf into herbal tea is a tangible approach to provide consumers with access to the benefits of these health-promoting compounds.

Bamboo is classified as a grass species, with a total of 1,662 species classified into 121 genera. It is a traditionally used plant with a broad range of benefits, as almost all parts can be converted into food and non-food products, and it is considered an alternative source of non-wood materials (Canavan et al., 2017; Sungkaew, & Teerawatananon, 2018; Ahmad et al., 2021; Khantayanuwong et al., 2022). A diverse range of bamboo species has been identified in Thailand, comprising up to 15 genera and 80 species thus far (Tangphadungrat et al., 2023). Among these, white bamboo (Dendrocalamus sericeus Munro.), known locally as Phai Sang Mon or Phai Nuan Rachini, is an economic crop native to the northern region, widely utilized in various industries (Second Regional Office of Agricultural Economics (2nd Regional OAE), 2023).

White bamboo forestry (cultivation) has been increasingly implemented to sufficiently meet the growing demand from industry (Phuangchik et al., 2021). The Thai government has also issued a policy promoting the cultivation of white bamboo as an alternative crop with market potential (2nd Regional OAE, 2023). The production of herbal tea from white bamboo leaves is therefore feasible in terms of raw material availability, and research should be conducted to evaluate the effect of the tea-making process on the content of health-promoting compounds in white bamboo leaves.

Bamboo leaf tea is an unfermented type of tea and can be processed in a similar way to green tea. The processing steps include plucking, steaming/roasting, rolling, drying, firing, sorting, and packing (Bokuchava et al., 1980). The steaming/roasting step is essential for inactivating browning enzymes, while the drying step helps inhibit bacterial growth, ensures product quality, and prevents the oxidation of chemical constituents. However, these processing steps can significantly affect the degradation of phenolic compounds and flavonoids (Benjamin et al., 2022).

Considering the impact of the manufacturing process on the quality of bamboo leaf tea, this research was conducted to evaluate the optimal steaming duration for white bamboo leaves and to determine the appropriate moisture reduction method by assessing the effects of two different drying techniques, roasting and hot air drying, on the phytochemical content of the dried leaves.

Previously, the tea-making process of five bamboo species in the Dendrocalamus genus (excluding D. sericeus) was reported by Indira et al., (2022). Fresh leaves (3–4 weeks old from 3–4-month-old culms) were washed, and hard portions were removed. The leaves were then dried at 40°C for 24 hours in a hot air oven and subsequently ground. The tea was brewed by steeping in hot water (95–100°C) for 3–5 minutes at a ratio of 1:100 (g/mL). The total phenolic content (TPC) and total flavonoid content

(TFC) in the tea ranged from 732–1051 mg/100 g and 69–120 mg QE/g, respectively. The retention of these compounds in the brewed tea ranged from 57–79% for TPC and 14–16% for TFC. The tea-making process of Beijing bamboo (*Dendrocalamus* sp.) planted in Thailand was also reported by Sansawat et al., (2024). One-centimeter-wide pieces of cleaned fresh leaves were dried in a tray dryer until their moisture content dropped below 8%. The dried leaves were then ground and passed through a 35-mesh sieve. The optimum drying temperature was found to be 65°C compared to 60°C and 70°C, as this condition yielded the highest levels of TPC (123.98 mg GAE/L), TFC (273.26 mg QE/L), and antioxidant activity.

Producing white bamboo leaf tea will not only create a healthy food product but also contribute to the sustainability of the bamboo processing industry by reusing industrial waste. This is because the culms of white bamboo are highly valuable for construction purposes and furniture making (Sungkaew, & Teerawatananon, 2018; Karawak et al., 2022). Moreover, they are a key alternative biodegradable material that has been extensively used to produce environmentally friendly food-related products. These include disposable cutlery (e.g., chopsticks, forks, food and drink picks), kitchen utensils (e.g., skewers, spatulas, turners, ladles, spoonulas, steamer baskets, decorative picks), and food containers (e.g., various forms of wickerwork, bamboo tubes) (Ekhuemelo et al., 2018; Boro et al., 2020; Zhang et al., 2024).

Thus, it is reasonable to expect a considerable amount of bamboo leaves left over from culm processing. However, in Thailand, bamboo foliage is largely underutilized and used primarily as animal feed, resulting in a significant amount of waste. Successfully processing leftover white bamboo leaves into a functional food would be a compelling option for achieving sustainability goals. This aligns with Tundis et al., (2023), who reported increasing interest in utilizing healthy constituents from industrial waste for the development of functional foods.

Additionally, herbal tea processing is not complex and does not require advanced technologies or machinery, making it feasible for community enterprises. A group of local individuals, including farmers, can easily establish and operate such a business. This concurs with reports indicating that over 97% of herb-related businesses in Thailand are small enterprises (Business Data Processing and Analysis Section, 2024). Hence, the results of this research can benefit most community enterprises in Thailand.

Despite the health-promoting properties of bamboo leaf tea and the growing industrial interest in bamboo products, limited research has been conducted on how steaming and drying processes affect the phytochemical and physical qualities of white bamboo (*D. sericeus*) leaf tea, particularly within the Thai context. This study aims to address that gap.

2. Objectives

This research aimed to compare the effects of steaming duration and moisture reduction methods (either by roasting or hot air drying) on the phytochemical content, including total phenolics, total flavonoids, catechin, orientin, isoorientin, vitexin, and isovitexin, as well as key physicochemical properties (instrumental color and moisture content) of white bamboo leaves. Additionally, the study aimed to optimize the tea processing steps (steaming and moisture reduction) to produce white bamboo leaf tea with high phytochemical content that meets relevant product standards.

3. Materials and methods

3.1 Bamboo Leaves

Foliage leaves of white bamboo were harvested from cultivated plants located in a bamboo plantation in Kanchanaburi Province, central Thailand. Leaves were specifically selected from mature plants (at least 6 months old) and were carefully collected to avoid any physical damage. Harvesting was done in the morning, and the leaves were initially hand-washed with tap water. After washing, they were laid out for outdoor airing during the day to eliminate any residual moisture. Once dry, the leaves were packed in plastic bags, vacuum sealed, and stored in a chilled, light-omitted environment at 4°C.

3.2 Chemicals and reagents

Standard chemicals including gallic acid, quercetin, catechin, ferulic acid, orientin, isoorientin, vitexin, isovitexin, and caffeine as well as Folin-Ciocalteu's phenol reagent were purchased from Sigma-Aldrich Co. Ltd. (Burlington, Massachusetts, USA). HPLC grade solvents were purchased from Thermo Fisher Scientific Inc. (Waltham, Massachusetts, USA). All other chemicals were analytical reagent (AR) grade.

3.3 Bamboo leaf tea processing

3.3.1 Steaming experiment

Each bamboo leaf sample, weighing 40 g, was manually cut into small pieces approximately 5 cm in width and length, then placed in a batch steamer. Six samples were steamed over boiling water (100°C) for different durations: 5, 10, 15, 20, 25, and 30 minutes. To prevent overheating and moisture loss, the leaves were arranged with minimal overlap. The internal temperature of the steamer was monitored throughout the process, and the steamer was equipped with a tight-fitting lid to prevent the escape of water vapor. After steaming, the samples were air-dried on perforated trays using a tray dryer (FNB Machinery & Solutions Co. Ltd., Bangkok, Thailand) with the heater turned off, exposing the samples only to ambient air at 30°C. An unsteamed sample was included as the control. All seven samples were then analyzed for phytochemical content, including total phenolics, total flavonoids, catechin, ferulic acid, orientin, isoorientin, vitexin, and isovitexin. The color of each sample was also measured. Based on these results, the optimal steaming time was selected for use in the subsequent experiment.

3.3.2 Moisture reduction experiment

Another set of bamboo leaf samples was prepared following the method described in Section 3.3.1, using the optimal steaming time previously determined. These steamed samples were then subjected to two different moisture reduction methods, roasting and hot air drying, to reduce the moisture content to below 10% (w/w), in accordance with the Thai Ministry of Public Health standard for tea infusion (Thai Ministry of Public Health, 2021).

Three samples were roasted for different durations (10, 20, and 30 minutes) in a brass round-bottomed pan placed on a gas stove. The pan had a diameter of 35 cm and a depth of 13 cm. The samples were manually stirred periodically with a spatula to ensure heat distribution. The temperature of each sample was monitored using a digital thermocouple and maintained at approximately 60°C by continuously adjusting the stove's temperature knob.

Another three samples were dried in a tray dryer at 60°C for different durations (10, 20, and 30 minutes). The leaves were spread evenly on perforated stainless steel trays with minimal overlapping. The average air velocity above the samples was 6 m/s. A control sample, which was not subjected to either roasting or hot air drying, was also included.

All seven samples were evaluated for phytochemical content (including total phenolics, total flavonoids, catechin, ferulic acid, orientin, isoorientin, vitexin, and isovitexin) and instrumental color, as described in Section 3.3.1. Moisture content was also measured. The appropriate moisture reduction method and drying time were then selected based on these evaluation results.

3.3.3 Grinding, packaging and quality checking

Bamboo leaf tea was prepared using the optimized processing conditions determined from previous experiments. The steamed and dried leaves were coarsely ground using an herb grinder and packed into tea bags (sachets), each containing 1.5 g of ground leaves. The tea bags were then stored in aluminum foil zip-lock bags to protect them from light and air and placed in an ambient temperature storage room.

Microbiological quality assessments of the bamboo leaf tea were conducted, including aerobic plate count (APC), yeast and mold count, and detection of *Escherichia coli*.

3.4 Bamboo leaf tea brewing

Brewed bamboo leaf tea samples were prepared using a standard steeping process. Each tea bag was steeped for 10 minutes in 150 mL of freshly boiled bottled water (brought to a boil at 100°C and allowed to sit for 1 minute until all bubbles disappeared). The remaining boiled water was discarded before preparing the next sample. After steeping, the tea bag was removed, and the brewed tea was collected in a glass bottle. The chemical and physical properties of the brewed tea were then analyzed, including caffeine content, pH, total soluble solids (TSS), and instrumental color.

3.5 Chemical analysis

3.5.1 Extraction of phytochemicals

To determine the phytochemical content, each sample was converted into powder following the method of Sutharut, & Sudarat (2012), with slight modifications (Baluchamy, & Subramanian, 2023). The sample was pulverized into a fine powder using liquid nitrogen. Then, 3 mL of 60% ethanol was added to 1 g of the dried sample and mixed for 30 seconds using a vortex mixer (Vortex-Genie 2 G650E, Scientific Industries Inc., Bohemia, New York, USA). The mixture was subsequently heated at 60°C for 20 minutes in a water bath and centrifuged at 10,000 rpm for 10 minutes using a centrifuge (Jouan CR3i,

Thermo Fisher Scientific Inc., Waltham, Massachusetts, USA). The supernatant was transferred to a 10 mL volumetric flask. The residue was re-extracted three additional times using the same procedure, each time with 3 mL of 60% ethanol. All collected supernatants were combined and adjusted to a final volume of 10 mL.

The extracted solution was stored in an opaque bottle at 4°C until further analysis (Ji-u, & Apisittiwong, 2022).

3.5.2 Total phenolics content (TPC)

TPC was determined using the Folin-Ciocalteu method as described by Macwan et al., (2010), with slight modifications. A volume of 100 µL of the extracted sample was mixed with 500 µL of 10% (v/v) Folin-Ciocalteu reagent using a vortex mixer for 30 seconds and kept in the dark for 1 minute. Subsequently, 1.5 mL of 7.5% Na_2CO_3 and 1.5 mL of distilled water were added to the mixture and vortexed again. The resulting solution was left at room temperature for 30 minutes. Absorbance was measured at 760 nm using a spectrophotometer (Genesys 20, Thermo Fisher Scientific Inc., Waltham, Massachusetts, USA). TPC, expressed as milligrams of gallic acid equivalent per gram of dried sample (mg GAE/g), was calculated from a calibration curve prepared with various concentrations of standard gallic acid solution (Orsuwan et al., 2024).

3.5.3 Total flavonoids content (TFC)

TFC was determined based on the formation of aluminum–flavonoid complexes as described by Pękal, & Pyrzynska (2014), with slight modifications. A volume of 3 mL of the extracted sample was mixed with 150 µL of 5% $NaNO_2$ using a vortex mixer for 20 seconds. After 6 minutes, 150 µL of 10% $AlCl_3$ was added and the mixture was vortexed again. Six minutes later, the sample was neutralized with 1 mL of 1 M NaOH and left at room temperature for 10 minutes. Absorbance was measured at 510 nm using a spectrophotometer. TFC was calculated using a calibration curve prepared with various concentrations of standard quercetin and expressed as milligrams of quercetin equivalent per gram of dried sample (mg QE/g).

3.5.4 Catechin and ferulic acid

The contents of catechin and ferulic acid were determined following the method of Burin et al., (2011) using a high-performance liquid chromatography (HPLC) system (1260 Infinity III, Agilent Technologies Inc., Santa Clara, California, USA) equipped with a photodiode array (PDA) detector operated at 280 nm. Chromatographic separation was conducted on a C18 reversed-phase column (4.6 mm × 250 mm, 5 µm particle size; Inertsil ODS-3, GL Sciences Inc., Torrance, California, USA). The column temperature was maintained at 20°C, and the injection volume was 20 µL. The mobile phase, in which its flow rate was set at 1.2 mL/min, consisted of acetic acid in purified water (Milli-Q apparatus, MilliporeSigma, Massachusetts, USA) adjusted to pH 2.6 as solvent A and 20% of solution A in 80% acetonitrile as solvent B. The gradient elution condition based on proportions of solvent B was used as follows: 0-30% B (0-35 min), 30-50% B (35-40 min), and 50-100% B (40-45 min). Standard curves for catechin and ferulic acid were obtained at concentrations of 10-80 ppm.

3.5.5 Orientin, isoorientin, vitexin, and isovitexin

Quantification of the four C-glycosyl flavonoids, orientin, isoorientin, vitexin, and isovitexin, was performed following the method described by Yang et al., (2014) using an HPLC system (Nexera LC-40 series, Shimadzu Corp., Kyoto, Japan) equipped with a photodiode array (PDA) detector (SPD-M40) set to 330 nm. Chromatographic separation was achieved using a C18 reversed-phase column (4.6 mm × 250 mm, 5 µm particle size; C18(2), Phenomenex Ltd., Torrance, California, USA). The mobile phase consisted of solvent A (0.3:99.7 v/v phosphoric acid-water) and solvent B (acetonitrile). The flow rate was maintained at 1.0 mL/min. Gradient elution was programmed from 15% to 25% solvent B over 0–35 minutes. Calibration curves for orientin, isoorientin, vitexin, and isovitexin were prepared using standard solutions with concentrations ranging from 6.25 to 200 ppm.

3.5.6 Caffeine

Caffeine content was determined using a modified method from Sharif et al., (2014). A 50 mL sample of brewed tea was extracted with 25 mL of dichloromethane in a separatory funnel. The aqueous phase was then re-extracted twice, each time with an additional 25 mL of dichloromethane. All dichloromethane extracts were combined in a 100 mL volumetric flask, and the volume was adjusted to 100 mL with dichloromethane. The absorbance of the final extract was measured using a UV-VIS spectrophotometer (Shimadzu Corp., Kyoto, Japan) at 275 nm. Caffeine concentration was determined by

comparing the absorbance to a calibration curve prepared with standard caffeine solutions in dichloromethane, ranging from 20 to 100 ppm.

3.6 Physicochemical and microbiological analysis

Moisture content of the samples was measured using a moisture analyzer (MA 30, Sartorius AG, Göttingen, Germany). Instrumental color of both solid and liquid samples was assessed using a colorimeter (HunterLab ColorQuest XE, Hunter Associates Laboratory Inc., Reston, Virginia, USA) and reported in CIE Lab color space values (L*, a*, and b*). The pH and total soluble solids (TSS) of the brewed tea were measured using a pH meter (ST300, OHAUS Corp., Parsippany, New Jersey, USA) and a Brix refractometer (MA871, Milwaukee Instruments Inc., Rocky Mount, North Carolina, USA), respectively. Microbiological quality was analyzed following standard AOAC methods (AOAC, 2000). Aerobic plate count (APC) and yeast and mold counts were expressed in colony-forming units per gram (CFU/g). *Escherichia coli* was enumerated using Petrifilm™ EC (E. coli and Coliform count) plates (Neogen Corp., Lansing, Michigan, USA) and also reported as CFU/g.

3.7 Statistical analysis

Each experiment was conducted in triplicate, and the data were expressed as mean ± standard deviation (SD). Statistical comparisons were performed using one-way analysis of variance (ANOVA) based on a completely randomized design (CRD), followed by Duncan's Multiple Range Test (DMRT). A significance level of $p < 0.05$ was used to determine statistical differences.

4. Results and discussion

4.1 Bamboo leaf tea processing experiments

4.1.1 Steaming step

The phytochemical contents of steamed bamboo leaf samples were shown in Table 1 and 2, while their appearance and color ~~of~~ were shown in Figure 1 and Table 3 respectively.

Thermal processing can disrupt the food matrix, enhancing the availability of phytochemicals, an effect that may outweigh thermal degradation (Paciulli et al., 2018). As shown in Table 1, the total phenolic content (TPC) of bamboo leaves significantly increased with steaming time up to 15 minutes, after which it gradually declined. This increase may result from the heat-induced conversion of insoluble phenolics into soluble forms (Liang et al., 2018). Additionally, steaming may cause substantial cellular damage, promoting the release of bound phenolic compounds (Xiong et al., 2019). A similar trend was observed in tea leaves (*Camellia sinensis* var. *assamica*), where TPC and antioxidant activity rose significantly after steaming at 100°C for 1 hour (Chupeerach et al., 2021).

Table 1 Content of phenolics, flavonoids, catechin and ferulic acid in bamboo leaves after steaming under different times.

Steaming time (min)	Total phenolics (mg GAE/g)	Total flavonoids (mg QE/g)	Catechin (mg/100 g)	Ferulic acid (mg/100 g)
0	10.74 ± 0.12^{e}	7.20 ± 0.06^{e}	4.41 ±0.02 ^{f}	1.68 ±0.02 ^{g}
5	12.96 ± 0.10^{b}	10.98 ± 0.11^{a}	6.67 ±0.09 ^{d}	2.67 ±0.00 ^{e}
10	11.83 ± 0.05^{c}	9.34 ± 0.03^{c}	5.77 ±0.02 ^{e}	2.18 ±0.01 ^{f}
15	14.97 ± 0.05^{a}	10.40 ± 0.03^{b}	8.21 ±0.05 ^{b}	3.11 ±0.01 ^{d}
20	11.15 ± 0.15^{d}	8.31 ± 0.12^{d}	8.32 ±0.14 ^{b}	4.78 ±0.05 ^{b}
25	11.26 ± 0.12^{d}	858. ± 0.09^{d}	15.01 ±0.13 ^{a}	8.51 ±0.09 ^{a}
30	11.67 ± 0.14^{c}	8.24 ± 0.08^{d}	7.05 ±0.01 ^{c}	4.27 ±0.03 ^{c}

Values are expressed as mean ± standard deviation.
Different letters in the same column indicate significantly different value ($p \leq 0.05$).

(a) (b) (c) (d) (e) (f) (g)

Figure 1 Appearances of bamboo leaves after steaming under different times.
(a) = 0 min, (b) = 5 min, (c) = 10 min, (d) = 15 min, (e) = 20 min, (f) = 25 min, and (g) = 30 min.

Table 2 Content of orientin, isoorientin, vitexin and isovitexin in bamboo leaves after steaming under different times.

Steaming time (min)	Orientin (ppm)	Isoorientin (ppm)	Vitexin (ppm)	Isovitexin (ppm)
0	7.50 ±0.00 ^{g}	ND	3.32 ±0.00	ND
5	24.20 ±0.01 ^{f}	80.65 ±0.02 ^{a}	ND	509.31 ±0.01 ^{a}
10	29.05 ±0.01 ^{e}	50.53 ±0.02 ^{c}	ND	299.45 ±0.02 ^{c}
15	46.72 ±0.01 ^{b}	63.86 ±0.01 ^{b}	ND	395.40 ±0.02 ^{b}
20	45.67 ±0.01 ^{c}	24.67 ±0.01 ^{e}	ND	100.28 ±0.00 ^{e}
25	38.31 ±0.01 ^{d}	22.37 ±0.01 ^{f}	ND	87.54 ±0.02 ^{f}
30	90.59 ±0.01 ^{a}	35.11 ±0.00 ^{d}	ND	188.96 ±0.00 ^{d}

Values are expressed as mean ± standard deviation, ND = not detected.
Different letters in the same column indicate significantly different value ($p \leq 0.05$).

The trends in total flavonoid content (TFC) and catechin levels mirrored the changes in TPC during steaming. The highest TFC was found at 5 minutes, while catechin peaked at 25 minutes. These outcomes may stem from the dual role of heat in disrupting plant cells and inactivating oxidative enzymes that degrade flavonoids, as well as interfering with plant metabolism affecting flavonoid biosynthesis (Hollman, 2004; Ahmed, & Eun, 2018).

Ferulic acid, a key component in plant cell walls responsible for cross-linking polysaccharides and other polymers (Paiva et al., 2013), also increased with steaming up to 25 minutes. This likely resulted from the release of bound phenolics from the cell wall and degradation of conjugated polyphenolics, such as tannins, which break down into simpler phenolics at high temperatures (Wu et al., 2013). While TPC peaked at 15 minutes, the highest catechin and ferulic acid levels were observed at 25 minutes, consistent with the fact that the predominant phenolic acids in bamboo leaves are cryptochlorogenic, chlorogenic, and neochlorogenic acids (Ma et al., 2020).

Regarding the four C-glycosyl flavonoids (Table 2), vitexin was undetectable post-steaming, possibly due to thermal structural degradation. Conversely, isoorientin and isovitexin peaked after 5 minutes of steaming, followed by a significant decline. This pattern aligns with the theory that initial heat exposure enhances phytochemical availability through cell disruption, which is then offset by thermal degradation (Paciulli et al., 2018). Based on the highest TPC observed at 15 minutes of steaming, this condition was selected for subsequent experiments.

Steaming is a crucial step in inhibiting the enzymatic browning of tea leaves prior to drying, and variations in steaming duration can lead to different quality outcomes. As shown in Table 3, the lightness (L*) of steamed bamboo leaf samples did not significantly differ from that of the unprocessed control. However, greenness (a*) decreased when steaming exceeded 5 minutes, which corresponded with a decline in yellowness (b*) after more than 10 minutes of steaming.

This change in color is consistent with findings in thermally processed green vegetables, where heat induces chlorophyll degradation and the formation of pheophytins or their derivatives, resulting in a color shift from bright green to olive brown (Nisha et al., 2004). In this case, longer steaming times intensified chlorophyll degradation. Interestingly, a slight increase in green color intensity was observed after 5 minutes of steaming, which may be attributed to the conversion of non-colored or less intensely colored chlorophyll precursors into greener compounds (Turkmen et al., 2006).

Despite these changes, the 15-minute steaming condition, selected for its optimal TPC, caused only a slight alteration in color compared to the control. All color parameters (L*, a*, and b*) remained statistically similar to the unprocessed sample, indicating minimal visual impact at the selected steaming duration.

4.1.2 Moisture reduction step

The moisture reduction step significantly affected the quality of bamboo leaves, primarily due to its impact on various heat-sensitive components. The phytochemical content of bamboo leaf samples subjected to different drying durations and methods is presented in Tables 4 and 5, while other characteristics, including instrumental color and moisture content, are shown in Table 6.

All samples underwent a 15-minute steaming process prior to moisture reduction, following the optimal condition identified in the previous experiment. Subsequent moisture reduction was performed using either roasting or hot air drying, with variations in drying time to evaluate their respective effects on the final quality attributes of the bamboo leaf tea.

Table 3 Instrumental color of bamboo leaves after steaming under different times.

Steaming time (min)	Lightness (*L**)	Greenness (*a**)	Yellowness (*b**)
0 (control)	53.93 ± 2.43^{ab}	-5.30 ± 0.70^{ab}	12.38 ± 0.85^{bc}
5	51.93 ± 0.28^{ab}	-6.48 ± 0.90^{a}	14.13 ± 0.59^{ab}
10	54.98 ± 5.25^{a}	-4.75 ± 2.16^{bc}	16.08 ± 3.03^{a}
15	55.70 ± 2.69^{a}	-4.43 ± 0.39^{bc}	14.15 ± 3.16^{ab}
20	50.25 ± 0.62^{b}	-3.88 ± 0.32^{bc}	10.43 ± 0.77^{c}
25	51.83 ± 2.67^{ab}	-3.65 ± 0.61^{c}	10.60 ± 0.95^{c}
30	54.38 ± 0.28^{ab}	-3.28 ± 0.25^{c}	10.88 ± 1.04^{c}

Values are expressed as mean ± standard deviation.
Different letters in the same column indicate significantly different value ($p \leq 0.05$).

Table 4 Content of phenolics, flavonoids, catechin and ferulic acid in bamboo leaves after steaming for 15 min followed by roasting or hot air drying under different times.

Drying method	Time (min)	Total phenolics (mg GAE/g)	Total flavonoids (mg QE/g)	Catechin (mg/100 g)	Ferulic acid (mg/100 g)
Control	0	14.97 ± 0.05^{a}	10.40 ± 0.03^{a}	8.21 ±0.05 ^{e}	3.11 ±0.01 ^{d}
Roasting	10	11.26± 0.12^{d}	7.70± 0.02^{e}	15.58 ± 0.14^{c}	2.18 ± 0.01^{f}
	20	11.48± 0.03^{c}	7.79± 0.03^{e}	15.77 ± 0.06^{b}	1.62 ± 0.00^{g}
	30	11.73± 0.07^{b}	7.87± 0.12^{e}	19.10 ± 0.12^{a}	2.33 ± 0.01^{e}
Hot air drying	10	10.56± 0.05^{g}	8.68± 0.27^{d}	13.77 ± 0.12^{d}	7.06 ± 0.09^{c}
	20	10.82± 0.03^{f}	9.09± 0.15^{c}	13.78 ± 0.13^{d}	7.89 ± 0.08^{b}
	30	10.98± 0.11^{e}	9.46± 0.19^{b}	13.96 ± 0.10^{d}	8.12 ± 0.02^{a}

Values are expressed as mean ± standard deviation.
Different letters in the same column indicate significantly different value ($p \leq 0.05$).

Table 5 Content of phytochemicals in bamboo leaves after steaming for 15 min followed by roasting or hot air drying under different times.

Drying method	Time (min)	Orientin (ppm)	Isoorientin (ppm)	Vitexin (ppm)	Isovitexin (ppm)
Control	0	46.72 ±0.01 ^{f}	63.86 ±0.01 ^{a}	ND	395.40 ±0.02 ^{a}
Roasting	10	49.42 ± 0.02^{e}	ND	ND	ND
	20	29.11 ± 0.01^{g}	ND	ND	ND
	30	61.41 ± 0.01^{d}	5.07 ± 0.00^{e}	ND	ND
Hot air drying	10	67.11 ± 0.01^{b}	35.10 ± 0.00^{c}	ND	163.87 ± 0.02^{c}
	20	62.68 ± 0.00^{c}	31.07 ± 0.02^{d}	ND	134.71 ± 0.00^{d}
	30	71.41 ± 0.01^{a}	36.73 ± 0.01^{b}	ND	166.61 ± 0.00^{b}

Values are expressed as mean ± standard deviation, ND = not detected.
Different letters in the same column indicate significantly different value ($p \leq 0.05$).

As shown in Table 4, both total phenolic content (TPC) and total flavonoid content (TFC) increased with longer processing times (10 to 30 minutes) for both moisture reduction methods, roasting and hot air drying. This increase may be attributed to thermal disruption of plant cell structures, which facilitates the release of bound phenolics and flavonoids (Ahmed, & Eun, 2018), thereby enhancing TPC and TFC in the dried bamboo leaf samples.

Notably, the increase in phytochemical content was more pronounced in the roasted samples. This can be attributed to the additional mechanical force applied through continuous spatula pressing during roasting in a brass open-pan, which likely intensified cell rupture and compound release. The impact of heating was further highlighted by comparing the TPC of bamboo leaves dried by hot air at 60°C in this study (ranging from 10.56 to 10.98 mg GAE/g) with that of mature leaves from six bamboo species dried at 50°C for 24 hours in an oven, which showed lower TPC values ranging from 4.29 to 7.19 mg GAE/g, as reported by Benjamin et al., (2022).

Based on these results, a 30-minute moisture reduction using either roasting or hot air drying proved to be the most effective option, as these conditions yielded significantly higher levels of TPC and TFC.

However, when evaluating other key phytochemicals (Tables 4 and 5), hot air drying emerged as the more suitable moisture reduction method. This approach preserves four important bioactive compounds: ferulic acid, orientin, isoorientin, and isovitexin. The highest concentrations of these compounds, 8.12 mg/L for ferulic acid, 71.41 ppm for orientin, 36.73 ppm for isoorientin, and 166.61 ppm for isovitexin, were achieved when samples were hot air dried for 30 minutes.

Interestingly, isovitexin was detected only in the hot air dried samples. This absence in roasted samples may result from thermal degradation, as roasting involved frequent direct contact with the hot surface of the brass pan, which reached temperatures of 180–200°C. As reported by Lv et al., (2016), isovitexin exhibits notable anti-inflammatory and antioxidant effects, particularly in cases of acute lung injury.

Vitexin, on the other hand, was not detected in any of the samples, consistent with its low initial presence in unprocessed leaves. Previous studies have shown that the composition and concentration of phenolics and flavonoids vary among bamboo species. For example, vitexin was not detected in *Bambusa multiplex* cv. *Silverstripe* and *Phyllostachys acuta* (Wang et al., 2012). Similarly, Karawak et al., (2020) found the highest concentrations of orientin, isoorientin, and isovitexin in the bamboo variety Pai Pak King at 6.68, 2.18, and 2.05 ppm, respectively, levels that were notably lower than those observed in this study, further confirming the enhancing effect of heat on C-glycoside content in bamboo leaves.

Therefore, based on the retention and enhancement of TPC, TFC, and key C-glycosides, hot air drying for 30 minutes is recommended as the optimal moisture reduction method in the production of bamboo leaf tea.

For the instrumental color measurements of roasted and hot air dried samples (Table 6), the highest lightness value (L* = 52.93) was observed in the sample roasted for 10 minutes, indicating a greater retention of brightness in the bamboo leaves. In contrast, samples subjected to longer roasting durations (20 and 30 minutes) and all hot air drying durations (10, 20, and 30 minutes) exhibited L* values below 50, suggesting darker or more opaque leaf appearances as a result of thermal exposure.

Regarding greenness (a*), more negative values indicate a stronger green hue. The greenest sample was obtained with 30 minutes of hot air drying, showing an a* value of -5.28. As for yellowness (b*), the highest value (12.05) was recorded in the sample hot air dried for 10 minutes. However, the differences in b* values among the three hot air drying durations were not statistically significant.

The differences in color may also be influenced by the final moisture content of the samples. In general, lower moisture content can slightly diminish the visual brightness of the leaves, leading to reduced lightness. This suggests that moisture content is a contributing factor to the color changes observed during the moisture reduction process.

As shown in Table 6, moisture content decreased consistently with increasing processing time. The method of moisture reduction had a notable impact on the final moisture content of the bamboo leaves, even though the color differences among treatments were relatively minor.

In the roasting process, the average temperature of the bamboo leaves remained around 60°C, while the brass pan surface, directly exposed to gas flame, reached temperatures between 180–200°C. Due to the continuous stirring and uniform heat distribution, roasted samples exhibited significantly lower moisture content than those subjected to hot air drying. Qi et al., (2021) noted that during tea roasting at 200°C, dehydration occurred rapidly, with more than 80% of water content lost within 500 seconds.

Table 6 Instrumental color and moisture content of bamboo leaves after steaming for 15 min followed by roasting or hot air drying under different times.

Drying method	Time (min)	Lightness (*L**)	Greenness (*a**)	Yellowness (*b**)	Moisture content (%wb)
Control	0	55.70 ± 2.69^{a}	-4.43 ± 0.39^{bc}	14.15 ± 3.16^{a}	-
Roasting	10	47.00 ± 1.74^{b}	-3.35 ± 0.42^{a}	11.90 ± 0.84^{a}	5.38 ± 0.07^{d}
	20	46.13 ± 1.04^{b}	-4.30 ± 0.71^{abc}	10.85 ± 0.83^{a}	5.22 ± 0.18^{d}
	30	46.60 ± 1.70^{b}	-4.08 ± 0.79^{ab}	11.15 ± 1.59^{a}	4.79 ± 0.24^{d}
Hot air drying	10	52.93 ± 1.86^{a}	-4.98 ± 0.68^{bc}	12.05 ± 1.38^{a}	9.70 ± 0.86^{a}
	20	47.60 ± 2.54^{b}	-4.98 ± 0.33^{bc}	10.78 ± 1.70^{a}	8.56 ± 0.18^{b}
	30	44.15 ± 1.35^{b}	-5.28 ± 0.50^{c}	8.85 ± 1.18^{a}	7.81 ± 0.19^{c}

Values are expressed as mean ± standard deviation.

Different letters in the same column indicate significantly different value ($p < 0.05$).

Table 7 Microbiological analysis of bamboo leaf tea powder

Total bacteria (CFU/g)	Yeasts and molds (CFU/g)	*E. coli* (CFU/g)
1×10^2	ND	ND

ND = not detected

Table 8 Total soluble solid (TSS), pH, color values, and caffeine content of bamboo leaf infusion tea

pH	TSS (°brix)	Color values			Caffeine content (ppm)
		L^*	a^*	b^*	
8.60 ± 0.01	0	82.97 ± 0.06	-10.67 ± 0.25	1.87 ± 0.06	ND

ND = not detected

Despite these differences, both roasting and hot air drying successfully reduced the moisture content to below 10% (w/w), in compliance with the Thai Ministry of Public Health standard for tea infusion (Thai Ministry of Health, 2021).

Considering the combined factors of high total phenolic content (TPC), total flavonoid content (TFC), orientin, isoorientin, isovitexin levels, and the enhanced greenness of the leaves, hot air drying at 60°C for 30 minutes was selected as the optimal moisture reduction method for bamboo leaf tea processing.

4.2 Qualities of ground and brewed bamboo leaf tea

The microbiological quality of ground bamboo leaf tea, processed by steaming for 15 minutes and then hot air drying at 60°C for 30 minutes, is presented in Table 7. According to the Thai Community Product Standard for dried herbs (TCPS 480) (Thai Industrial Standard Institute, 2004), the acceptable limits are a total viable count not exceeding 5×10^5 CFU/g and a yeast and mold count below 100 CFU/g.

The analysis results confirmed that the ground bamboo leaf tea met all specified criteria, indicating its microbiological safety and compliance with national quality standards for herbal tea products.

To evaluate the quality of brewed bamboo leaf tea, parameters including pH, total soluble solids (TSS), instrumental color, and caffeine content were measured, as shown in Table 8. The results clearly indicated that the bamboo leaf infusion contained no detectable caffeine, which can be considered a health advantage for certain consumer groups.

While caffeine is known for benefits such as increased alertness, it also poses neurophysiological risks when consumed in excessive amounts. These include anxiety, panic attacks, and hallucinations (Persad, 2011). Furthermore, even low doses of caffeine have been shown to influence behavior in children and may promote repeated consumption (Riddell, & Keast, 2007).

Thus, the absence of caffeine in bamboo leaf tea enhances its appeal as a naturally caffeine-free beverage option, suitable for individuals sensitive to caffeine or seeking to reduce their intake.

5. Conclusion

This study successfully identified optimal processing conditions for producing white bamboo (*Dendrocalamus sericeus*) leaf tea with enhanced phytochemical content and acceptable physicochemical and microbiological qualities. Steaming the bamboo leaves for 15 minutes significantly increased total phenolic content (TPC) and total flavonoid content (TFC), establishing this duration as the most effective pretreatment step. Among the moisture reduction techniques, hot air drying at 60 °C for 30 minutes proved superior to roasting, as it preserved higher levels of key bioactive compounds, particularly orientin, isoorientin, and isovitexin, while effectively reducing moisture content to below 10%, in compliance with Thai Ministry of Public Health standards. Additionally, the final tea product was confirmed to be caffeine-free, enhancing its attractiveness to consumers seeking stimulant-free herbal infusions. Microbiological analysis verified that the tea met the safety requirements of the Thai Community Product Standard for dried herbs.

Overall, this research highlights the importance of optimizing processing conditions to preserve valuable phytochemicals during herbal tea production. The findings support the development of a functional, health-promoting beverage and offer a sustainable solution for utilizing underused bamboo foliage. This contributes to waste reduction and value-added processing in the bamboo industry. The study provides practical guidance for community enterprises and small-scale producers in Thailand

aiming to create high-quality herbal tea products from locally available resources.

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