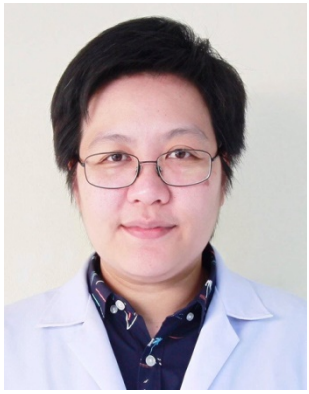
ajfand SCHOLARLY, PEER REVIEWED
AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT
Volume 19 No. 4
November 2019
PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST
ISSN 1684 5374

Afr. J. Food Agric. Nutr. Dev. 2019; 19(4): 14950-14970 DOI: 10.18697/ajfand.87.18120

PROCESS OPTIMIZATION, ANTIOXIDANT ACTIVITY AND SENSORY CHARACTERISTICS OF GREEN TEA MADE FROM YOUNG FRAGRANT RICE LEAVES CULTIVAR DAMGATONDAM

Uthai N[1*], [1]Thamakorn P[1], Kerdpiboon S[1] and R Nokkoul[2]



Narissara Uthai

*Corresponding author *e*mail :uthai.narissara@gmail.com

[1]Faculty of Agro-Industry, King Mongkut's Institute of Technology Ladkrabang, Bangkok 10520, Thailand

[2]Faculty of Agriculture, King Mongkut's Institute of Technology Ladkrabang Prince of Chumphon Campus, Chumphon 86160, Thailand

ajfand SCHOLARLY, PEER REVIEWED
AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT
Volume 19 No. 4
November 2019
PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST
ISSN 1684 5374

ABSTRACT

Green tea was made from one-month old leaves of the Thai upland black waxy rice cultivar Damgatondam (DGTD), which has a pleasant smell of jasmine. The objective was to determine the optimum process required to produce green tea that is acceptable to consumers and has health benefits. The processing conditions were steaming at 98 ± 2°C for 30, 45, 60 75, 90 or 105 sec then pan roasting at 70 ± 2°C for 5, 10, 15, 20, 25 or 30 min and finally tray drying by using electrical cabinet tray dryer at 60 ± 2°C for 30, 45, 60 or 90 min. Brewed samples were compared to brewed samples of seven commercially available *Camellia sinensis* tea products by descriptive analysis with 10 trained panelists. The highest quality of DGTD tea was: steaming at 98 ± 2°C for 60 sec, which completely inactivated peroxidase enzymes and preserved the green color, followed by pan roasting at 70 ± 2°C for 30 min and tray drying at 60 ± 2°C for 90 min in order to achieve a moisture content of less than 5%. The proximate analysis showed the DGTD tea prepared by the above method had: 3.77% moisture content, 7.94% ash, 22.59% crude fiber, 2.58% fat and 17.73% protein. Moreover, Ca, Mg, K, Fe, Mn and Na were 3400, 3200, 20400, 66.4, 139.1 and 113.8 mg/kg, respectively. It was found that protein, fat, crude fiber, ash, Mg, K and Na contents in DGTD tea, were higher than levels in tea from *Camellia sinensis*. Total phenolic content of DGTD tea was 9.53 mg GAE/g and antioxidant activity was 357.52 mM TE/g by Ferric reducing antioxidant power (FRAP), l34.08 mM TE/g by 2,2 diphenyl-1-picrylhydrazyl assay (DPPH) and 108.89 mM TE/g by 2,2'-azino-bis (3-ethylbenzothiazo line-6-sulphonic acid) (ABTS). The descriptive analysis of trained panels identified 11 particular attributes of DGTD tea, consisting of yellowness, clearness, tea odor, hay odor, green odor, seaweed odor, jasmine rice odor, tea flavor, seaweed flavor, bitter taste and astringent taste. The descriptive analysis showed that the DGTD tea was less bitter and astringent than the *Camellia sinensis* tea and its jasmine odor was positively accepted.

Key words: Damgatondam, rice grass, green tea, processing, bioactive compounds, sensory attributes

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

INTRODUCTION

Most tea is made from the leaves of *Camellia sinensis* (L.) Kuntze, which are fermented to produce black tea or not fermented to produce green tea [1]. Green tea is produced by inactivating peroxidase enzymes in the fresh leaves by applying either heat or steam before drying. There are many other plants used to produce tea including white mulberry leaves, pandanus, safflower, and chamomile flower [2, 3]. These teas are mainly produced by the same process as that used for *C. sinensis* tea.

Thailand is a major world producer of rice both for local consumption, where it forms the major staple, and for export. Most rice is irrigated but some is produced, mainly by smallholder farmers, without irrigation called upland or hill rice [4]. Having a by-product from the young plants (rice grass) could enhance the income of many hill rice farmers. Hill rice is grown from seed either by direct seeding or by transplanting. In either case, young seedlings are removed and many are discarded. These young seedlings could be utilized to provide an additional income for farmers.

New pure-line selections of local upland fragrant rice cultivars (*Oryza sativa* L.) in Thailand have resulted in many new varieties with particular characteristics, including Damgatondam (DGTD), which has a distinct jasmine aroma in both the seeds and leaves [5]. Green tea has previously been reported to be successfully produced from 14 - 21 day old leaves from 3 rice cultivars (Khao Dawk Mali 105, Pathum Thani 1 and Sakon Nakhon) that had a mean proximate analysis of 5.50 - 6.05% moisture, Ca 365.40 - 422.30 mg/100g, Na 20.4 - 31.60 mg/100g, K 1180.60 - 1925.20 mg/100g, Mg 194.20 - 421.40 mg/100g and Fe 6.06 - 11.0 mg/100g [6]. Yun [7] successfully made freeze dried rice grass juice from samples of 4 week old DGTD leaves and showed that it had abundant bioactive compounds and high antioxidant activity. The objective was, therefore, to determine the optimum processes required to produce green tea from young DGTD rice grass leaves and evaluate their sensory characteristics and composition in terms of total phenolic compounds, antioxidant activity by Ferric Reducing Antioxidant Power (FRAP, 2,2 - Diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) (ABTS) and key minerals.

MATERIALS AND METHODS

Raw material preparation

The upland black waxy rice cultivar Damgatondam (DGTD) was organically grown (September to October 2016) in a greenhouse at Prince of Chumphon Campus in Chumphon province in Southern of Thailand. The rice grass was harvested, about 1 month after planting, by cutting it about 1 cm above soil level. The rice grass was washed with tap water, sealed in polyethylene bags and wrapped in newspaper and stored in an insulated box containing ice over night for transport from Chumphon to the laboratory in Ladkrabang, Bangkok which took about 7 h. The external temperature during transport was about 30°C, but the temperature in the box was maintained at 3 - 5°C. On arrival the rice grass was washed again and cut into 1 cm long piece.

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

Chemicals

2,2 diphenyl-1-picrylhydrazyl (DPPH), 2,2-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), 6-hydroxy-2, 5, 7, 8-tetramethylchroman-2-carboxylic acid (trolox), 2,4,6-tripyridyl-s-triazine (TPTZ), iron (III) chloride hexahydrate, gallic acid monohydrate, quaiacol and Folin - Ciocalteu reagents were purchased from Sigma Aldrich Corp., St. Louis, MO, USA and sodium acetate trihydrate, sodium carbonate, potassium persulfate, 96% ethanol and 95.5% methanol were purchased from Merck KGAA, Darmstadt, Germany. Hydrochloric acid and acetic acid were procured from RCI Labscan Co., Ltd., 1 Rama Road, Bangkok. All other reagents were of analytical grade.

Inhibition of peroxidase enzyme activity by steaming

Determination the optimum amount of rice grass leaves to steam

The entire process used for making DGTD tea is shown in Figure 1. Varied amounts of rice grass leaves 100, 200, 300 or 400 g were placed in a stainless steamer (45 cm diameter) with a steaming time of 60, 90 or 120 sec at 98 ± 2°C. The rice grass leaves were evenly spread over the steamer racks. Clean cheese cloth was placed on the bottom of the rack before spreading the rice grass leaves. The effects of using a single or double steamer rack with rice grass leaves on each rack were tested. The accepted DGTD rice grass leaves should be dark green without damage and was judged by visual examination.

Determination of the optimum steaming time to inhibit peroxidase enzyme activity

The rice grass leaves samples (200 g) were steamed at 98 ± 2°C for 30, 45, 60, 75, 90 or 105 sec, and then their peroxidase activities were measured following the method of Alvarez *et al*. [8] where each sample was chopped and homogenized with 30 mL of distilled water in a homogenizer (Wise Tis HG-15A, Germany) at high speed for 3 min at 4°C. The slurry was filtered through two layers of cheese cloth and centrifuged for 15 min at 10,000 rpm also at 4°C. This extract was used for the enzyme source experiment.

Peroxidase activity was determined with a UV-Visible spectrometer (Shimadzu UV-1800, Japan) at 470 nm and 25ºC. In brief, the substrate mixture contained 10 mL of 0.01 mL/mL quaiacol solution, 10 mL of 3 mg/mL hydrogen peroxide solution and 100 mL of 0.05 mol/L sodium phosphate buffer (pH 6.5). The reaction cuvette contained 2.9 mL substrate mixture and 0.1 mL crude extract in a total volume of 3 mL. The blank sample (control) contained only 3 mL of substrate mixture. The determination was done in triplicate.

Effect of pan roasting on moisture content and antioxidant activity

After steaming, the samples of rice grass leaves were withered for 5 min by evenly spreading them on an aluminum tray at room temperature (about 25ºC) and then pan roasting them at 70 ± 2 °C for 5, 10, 15, 20, 25 or 30 min whilst their moisture contents were observed. Total phenolic content (TPC) and antioxidant activity by FRAP, DPPH and ABTS assays of roasted sample were measured. The determinations were done in triplicate.

ajfand SCHOLARLY, PEER REVIEWED
AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT
Volume 19 No. 4
November 2019
PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST
ISSN 1684 5374

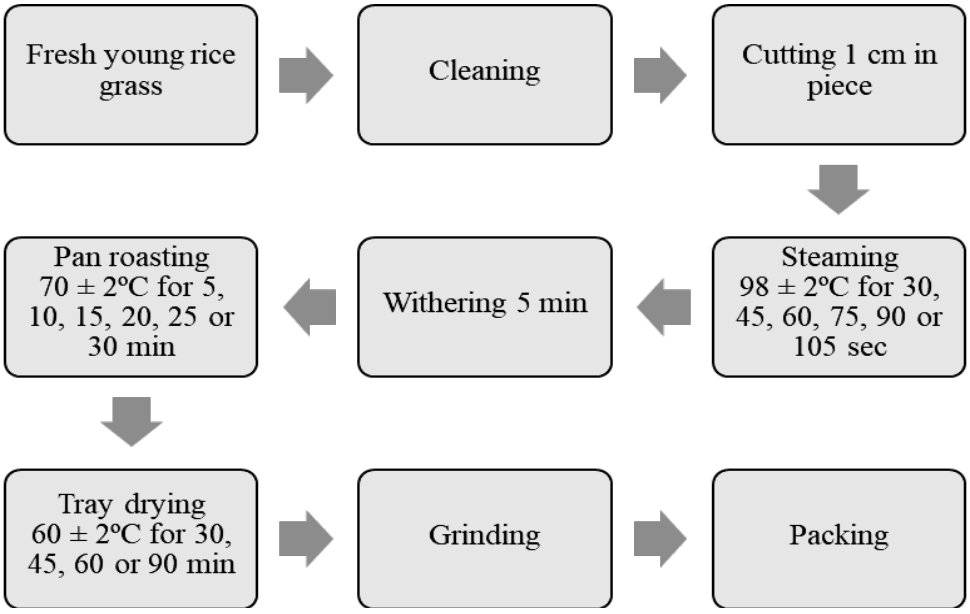


Figure 1: Process of DGTD tea production

Dehydration steps of DGTD tea processing

After roasting, the rice grass leaves were dried in a tray dryer at 60 ± 2ºC for 30, 45, 60 or 90 min. The target moisture content of tea products should be less than 5% [9]. Moisture content, TPC, and antioxidant activity by FRAP, DPPH and ABTS assays were determined in the same way as in the previous step.

Antioxidant activity determination

Damgatondam rice grass leaves (2 g) were extracted with 50 mL of methanol and thoroughly mixed for 1 min, centrifuged at 4500 rpm for 10 min and kept at -20ºC until used in the following analyses:

Total phenolic content

The procedure followed that of Yun *et al*. [10] with some modifications. In brief, 0.5 mL sample was reacted with 5 mL of 1:10 Folin - Ciocalteu: distilled water reagent. After 5 min the solution was neutralized with 4 mL of 1 M sodium carbonate solution. After 10 min the absorbance of the extract at 765 nm was measured in a UV visible spectrophotometer (UV-1800 Shimadzu, Japan). The linear standard curve of gallic acid was plotted in µg gallic acid of 0, 25, 50, 75, 100 and 125 and the results were calculated as gallic acid equivalent per 1 g of sample (mg GE/g).

Antioxidant capacity using Ferric reducing antioxidant power

The FRAP assay used the method of Wu *et al*. [11] with some modifications. The antioxidant in the FRAP assay consisted of acetate buffer (pH 3.6), ferric chloride solution (20 mM) and TPTZ solution (10 mM TPTZ in 40 mM HCl) in a proportion of 10:1:1, respectively and was freshly made for the analysis. To obtain a FRAP value for the antioxidant activity 2850 µL of FRAP solution (warmed to 37°C) was added to 0.15 mL of appropriately diluted sample, which was in a tube. The tubes were vortexed and

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

incubated at 37°C in the dark for exactly 30 min and the absorbance of each sample at 593 nm was measured in a UV visible spectrophotometer. A Trolox standard curve was prepared from a 1 mM Trolox methanolic stock solution and used to calculate the antioxidant capacity of the sample, which was expressed in mM Trolox equivalent per g sample (mM TE/g).

Antioxidant capacity using 2,2 diphenyl-1-picrylhydrazyl assay

The DPPH analysis was by the method adapted from Wu *et al*. [11]. The DPPH working solution was prepared by dissolving 0.003 g of DPPH in 50 mL ethanol. 1.5 mL of the diluted sample (the same amount of antioxidant standard) was reacted with 1.5 mL DPPH, vortexed and incubated for 30 min in the dark. Its absorbance at 517 nm was measured with a UV visible spectrophotometer (UV-1800 Shimadzu, Japan). A trolox standard curve was prepared from 1 mM trolox methanolic stock solution in order to calculate the antioxidant capacity, which was expressed in mM trolox equivalent per g sample (mM TE/g).

Antioxidant capacity using 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid)

The ABTS assay used the method described by Wu *et al*. [11] method with some modifications. Stock solutions of 7.4 mM ABTS and 2.6 mM potassium persulphate were freshly prepared on the day of analysis. The ABTS working solution was made by mixing the stock ABTS solution with potassium persulphate in a 1:1 ratio and leaving them to react for 12 h. Then, the mixture was diluted with 95.5% methanol such that an absorbance of 1.1 ± 0.02 at 734 nm was obtained. The filtrate from the 95.5% methanol extraction (0.15 mL) was allowed to react with the ABTS working solution (2.85 mL) in a test tube, vortexed, incubated in a dark place for 2 h, and its absorbance at 734 nm was measured with a UV visible spectrophotometer (UV-1800 Shimadzu, Japan). 1 mM Trolox methanolic stock solution was prepared and used for calculating the antioxidant capacity of the sample, which was expressed in mM Trolox equivalent per g sample (mM TE/g).

Determination of chemical composition and key minerals

Sample of DGTD tea product from the optimum process condition was analyzed based on AOAC [12] for water activity, moisture content, ash, protein, fat, crude fiber and several key minerals (Ca, Na, Mg, K, Fe, and Mn) using sub components 978.18, 925.10, 945.38, 981.10, 922.06, 978.10, 984.27, 984.27, 984.27, 984.27, 999.10 and 984.27, respectively.

Sensory descriptive analysis

Panel Selection

Ten panelists, aged between 25 and 32 years, were selected from 20 Food Science post-graduate students in the Faculty of Agro-Industry of King Mongkut's Institute of Technology Ladkrabang, who had previous experience in sensory evaluation and who were available to participate throughout the program. Before being selected to take part, each panelist was tested for ability to identify taste (sweet, salty, sour and bitter) and odor (banana, orange, strawberry, tea, coffee and vanilla) and to describe odor correctly and be able to correctly rank the intensity of taste (sweet, salty, sour and

bitter). As proposed by Meilgaard *et al.* [13], only candidates that were able to describe 80% of the stimuli and describe the characteristics of the stimuli were selected.

Training

Sensory descriptive analysis was carried out by panels using the technique described by Meilgaard *et al*. [11], Stone and Sidel [12] and Vittayaporn and Chompreeda [13] where panelists scored attributes on a 0 to 15 cm scale with one significant digit. The panels were then trained in 3 sessions for 3 months (180 h). The first session was to describe taste, odor and flavor characteristics of tea samples. Each panelist received a spit cup, napkins, drinking water, and green tea samples and then was asked to generate product attributes, define these attributes by consensus and identified reference standards for rating these attributes. In the second session, panelists reviewed the terms used and rated the intensity scores to create a reference standard for each attribute and produce a consensus for scores for each reference standard. In the third session, the panelists reviewed the reference standards and evaluated all of tea samples by the referable limit of reference standards that they had agreed to and then adjusted their scales until the standard deviation (SD) of scores was less than 1.0. On each day of training, panelists were adjusted with warm-up samples by rating the sample using 0 to 15 numerical scale ballots.

Sample preparation

Two grams of tea samples (seven brands of commercial green tea (*C. sinensis*)) from China, Japan, Poland, Sri Lanka or Thailand, which are available in the Thai market both as tea bags and tea leaves were packed in tea filter paper bag and brewed with 200 mL of hot boiled water (80°C) in a ceramic cup for 3 - 5 min (following the instructions on the tea packages). The brewed tea was kept in vacuum flask for just a few minutes and then served to panelists when the temperature was about 60ºC. For DGTD tea, the sample was used for the panelists tests and was prepared by brewing 2 g with 150 mL of hot boiled water (98°C) in a ceramic cup for 3 min then kept in vacuum flask for just a few minutes, then served to panelists when the temperature was about 60ºC.

Sample evaluation

After completing the training program, the panelists were given 50 mL of the brewed green tea samples in a ceramic cup and asked to evaluate the sample in triplicate in terms of appearance, aroma, flavor and taste and described the attributes of each sample. The samples were presented at 60°C and coded with three - digit random numbers. The panelists were informed that they would be served only one sample each time, and they were given a break for 5 min between each tea and were given a 1 h break after evaluating 4 samples. A spit cup for expectoration, paper napkins and drinking water were provided to each panelist for using during their evaluations.

Statistical Analysis

Data obtained were analyzed by analysis of variance (ANOVA) using Statistical Package for Social Sciences (SPSS) version 24 for windows. Duncan's Multiple Range Test (DMRT) was used to compare the different of mean $p \leq 0.05$ was considered as significant.

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT
Volume 19 No. 4 November 2019
PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST
ISSN 1684 5374

RESULTS AND DISCUSSIONS

Inhibition of peroxidase enzyme activity by steaming

Determination the optimum amount of rice grass leaves to steam

Inhibition enzyme activity was the most crucial step in the process in order to retard the loss of green color in the rice grass leaves. Attempts to use a double rack stemmer was not successful because when the double rack steamer was used, there was a possibility of insufficient heat to enzyme inactivation in the upper rack, while damage could occur simultaneously to the sample on the lower lack. Using a single rack steamer resulted in more consistent quality of the rice grass leaves due to a better temperature distribution. Among the sample sizes and steaming times tested, the optimum was the combination of 200 g for 60 sec (Table 1) using a single rack steamer, 60 sec was the minimum steaming time that inhibited peroxidase activity (Figure 2). Steaming samples of 300 g and 400 g resulted in irregular color even when steaming for only 60 sec and samples of 100 g gave similar results to 200 g. All sample sizes resulted in some damage when steamed for 90 or 120 sec.

Test for optimum steaming time to inhibit peroxidase enzyme activity

A sharp decrease in peroxidase activity levels with increasing steaming times over the range of 30 and 45 sec was observed. Peroxidase activity was completely inhibited after steaming for 60 sec (Figure 2). Precise steaming time is essential in producing an acceptable product since browning can occur, as observed in the experiments, due to the tissue and the chlorophyll being damaged by over exposure to heat or by enzyme action on the polyphenols because the temperature was insufficient to deactivate them. Therefore, on balance, it was concluded that the most suitable steaming time for DGTD tea was 60 sec at 98 ± 2°C, which is consistent with the results described by Wang *et al*. [16] for *C. sinensis* tea.

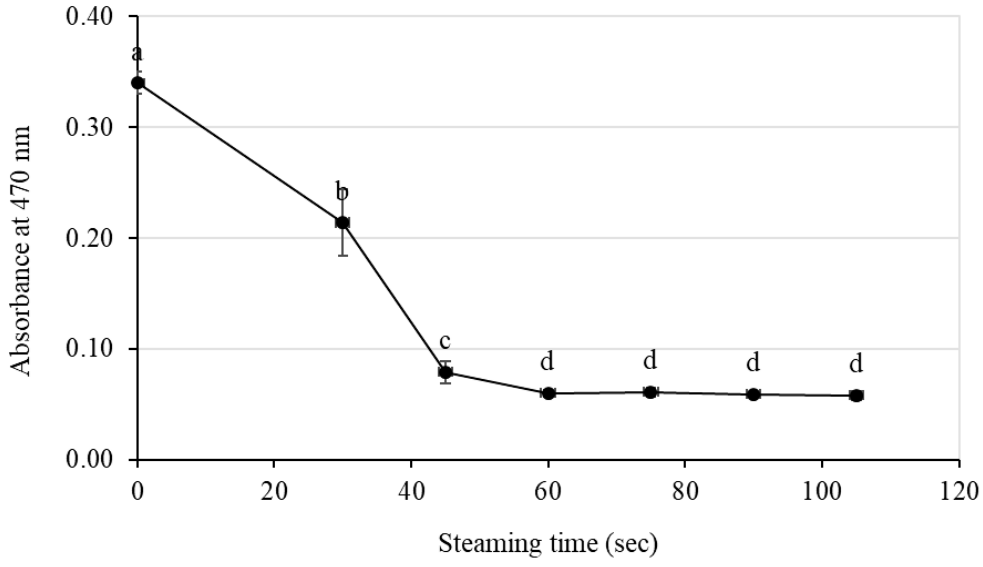


Figure 2: Effect of steaming time on peroxidase enzyme activity

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

Points with different letters were significantly different ($p \leq 0.05$)

Effect of pan roasting on moisture content and antioxidant activity

After 5 min of withering the moisture content of rice grass leaves was approximately 70%, which then continuously and sharply decreased during pan roasting for 5, 10, and 15 min giving moisture contents of 54.0, 44.3 and 37.1%, respectively. Subsequently, the moisture content continued to decrease then leveled off (Figure 3). During pan roasting for up to 25 min the leaves still had a fresh green color but this was not consistent after drying. After roasting for 35 min, the moisture content was about 36%, which was not significantly different ($p>0.05$) from 30 min but gave the leaves a green, dry, crisp appearance although some parts were burnt. It was considered that pan roasting at 70 ± 2°C for 30 min was the most effective since it gave the leaves a curled appearance and a mild roasted smell of tea. As Uhl [17] described, pan frying is traditionally done by hand on a wok over and open flame. The contact between the leaves and the hot metal triggers the Maillard reaction to form molecules that create unique odors and flavors. The Maillard reaction is a reaction between reducing sugars and amino acids at high temperature, which produces brown pigments that can have a caramel flavor. Kawakami and Yamanishi [18], Choi [19] and Zheng *et al.* [20] reported that green teas, which are prepared by roasting or pan frying, contain high levels of Millard reaction products, such as 1-ethyl-3, 4-dehydropyrrolidone, pyrazines, pyrroles, pyrans and furans. Pyrazines and 1-ethyl-3, 4-dehydropyrrolidone play an important role in developing a roasted flavor in both roasted and pan-fried green teas.

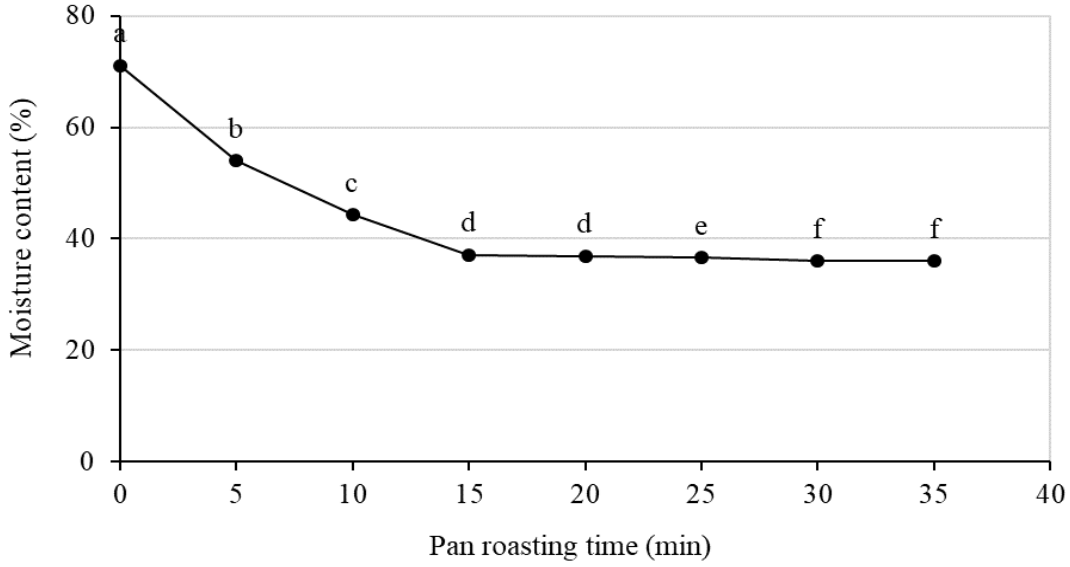


Figure 3: Moisture content in rice grass leaves during pan roasting at 70°C

Points with different letters were significantly different ($p \leq 0.05$)

Total phenolic content and antioxidant activity by FRAP, DPPH and ABTS assays of pan roasted samples progressively and continuously decreased significantly ($p \leq 0.05$) with increased heating time. The TPC decreased from 15.42 to 14.29 mg GAE/g sample, FRAP decreased from 533.67 to 516.57 mM TE/g sample, DPPH decreased from 53.96 to 47.81 mM TE/g sample and ABTS decreased from 178 to 150.22 mM

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

TE/g sample. (Table 2). Lorsuwan *et al*. [21] reported that there are many factors that directly affect the stability of phenolic compounds particularly temperature and acidity, which caused total phenolic content to decrease. Pan roasting and other thermal process can destroy antioxidant and phenolic compounds. Zzaman *et al*. [22] showed that roasting cocoa beans with superheated steam at temperatures of 150, 200 or 250°C and times over the range of 10 - 50 min, resulted in significantly ($p \leq 0.05$) lower free radical scavenging activity, antioxidant properties, total phenols and total flavonoids with the higher temperatures and longer times.

Dehydration step of rice grass leaves processing by tray dryer

The samples that had been pan roasting for 5, 10 or 15 min and then tray dried at 60 ± 2ºC for 30, 45 or 60 min all had moisture contents higher than 5% (Figure 4), which were higher than the standard for moisture content of dry tea leaves [23, 24]. However, when the drying time was increased to 90 min the moisture content fell below 5%. When samples were tray dried for 60 min combined with pan roasting for 20, 25 or 30 min, their moisture contents were 4.97, 4.85 and 4.14%, respectively. The reason why the longer drying time had little effect on the moisture content was because the equilibrium moisture content of the drying system had been reached. Previously, Keeratiburana and Srijesdaruk [25] showed the effect of tray drying at 40, 50 or 60°C for 10, 12 or 14 h on moisture content of Khao Dowk Mali 105 (KDML 105) rice grass tea resulted, as would be expected, in decreasing moisture content with increasing temperature and time. However, the dehydration step in DGTD tea processing resulted in stronger development of tea aroma and jasmine rice odor after 90 min of tray drying. Also, the optimum tray drying process achieved a moisture content below 5%, which gives stability and freedom from microorganisms [24].

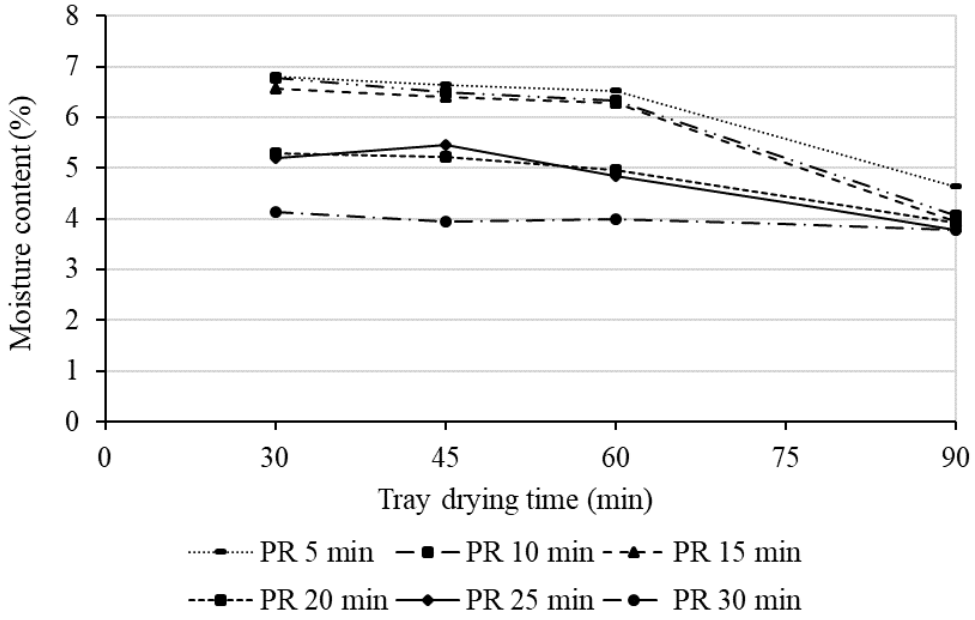


Figure 4: Moisture content of rice grass leaves pan roasted (PR) for 5, 10, 15, 20, 25 or 30 min prior to dehydration in an electrical cabinet tray dryer (TD) at 60ºC for 30, 45, 60 or 90 min

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

The effects of pan roasting rice grass leaves for 30 min and tray drying in an electrical cabinet tray dryer at 60ºC for 30, 45, 60 or 90 min on TPC, FRAP, DPPH and ABTS antioxidant activity were investigated. The TPC levels were 15.42, 13.09, 10.46 and 9.08 mg GAE/g, the FRAP levels were 516.57, 449.27, 424.64 and 369.90 mM TE/g, the DPPH levels were 47.81, 41.33, 36.73 and 29.17 mM TE/g, and the ABTS levels were 150. 22, 144. 89, 133. 78 and 100. 22 mM TE/g, respectively for the four dehydration times (Figure 5). The decreasing effect in TPC and FRAP with increasing drying time is consistent with the effects reported on mortino fruit (*Vaccinium floribundum*) by Lopez-Vidana *et al*. [26] who found that using temperatures of 40, 50 or 60°C for 15, 30, 45, 60, 90, 120 180, 240 or 300 min resulted in lower antioxidant capacity and TPC when drying with increasing time. Also, Keeratiburana and Srijesdaruk [25] showed that tray drying KDML (Khao Dawk Mali) rice grass at 40, 50 or 60° C for 10, 12 or 14 h resulted in decreased bioactive compounds (DPPH assay) when temperature and time were increased so the lowest temperature and shortest time gave the highest level of bioactive compounds. For chamomile tea, drying at 50°C for 4 or 5 h resulted in both decreased moisture content and decreased TPC with longer exposure time [27]. This confirmed that a high temperature for a longer time reduced the determined bioactive compounds.

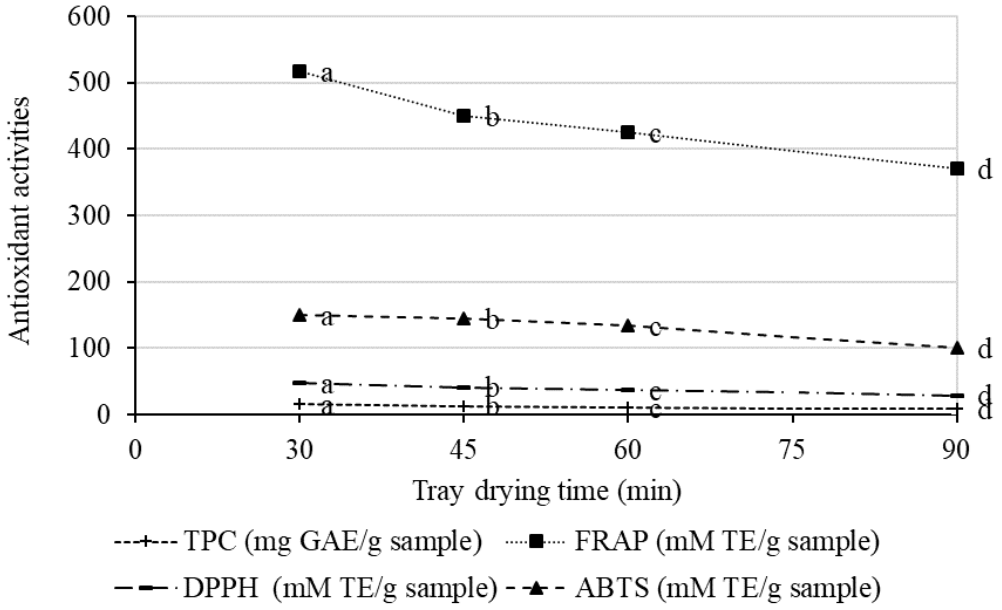


Figure 5: Total phenolic content, FRAP, DPPH and ABTS of rice grass leaves after pan roasting for 30 min and tray drying at 60 ± 2°C for 30, 45, 60 or 90 min

Proximate composition, mineral content, TPC and antioxidant activities of DGTD tea

The water activity, proximate composition, moisture content, ash, crude fiber, fat and protein of DGTD tea, which had been processed by steaming for 60 sec, pan roasting at 70 ± 2ºC for 30 min and tray drying at 60 ± 2ºC for 90 min were compared with several characters of tea leaves of *C. sinensis* [28, 29] (Table 3). There were considerable

differences, not only between DGTD tea and *C. sinensis* tea, but also within *C. sinensis*. For example, protein values were different, with levels of one of the *C. sinensis* teas reported to be 1.23% [29] and another at 18.60% [28], while DGTD tea was 17.73%. These differences in protein levels may be due to different varieties and cultivation conditions. There were also variations in mineral contents with DGTD tea, which were higher in Mg, K and Na, but lower in Ca, Fe and Mn compared to *C. sinensis* tea [28, 30] (Table 3). However, the mineral content in any crop reflects the soil in which it is grown so the mineral levels in the DGTD tea were comparably acceptable. Although TPC level in the DGTD tea was lower than *C. sinensis* tea [31, 32] (Table 3) and antioxidant activity as measured by FRAP, DPPH and ABTS, these levels were all considered acceptable [33] (Table 3).

Descriptive analysis of DGTD tea product

Ten trained panelists described the appearance, attributes and specified definition of all the DGTD tea products, which resulted in 11 different attributes quoted. These attributes were yellowness, clearness, tea odor, hay odor, green odor, seaweed odor, jasmine rice odor, tea flavor, seaweed flavor, bitter taste and astringent taste (Table 4). All the attributes were also present in the 7 brands of commercial *C. sinensis* tea that were tested except for jasmine odor which was found only in the DGTD tea. However, Lee and Chambers [34] described 18 attributes of *C. sinensis* tea produced from China, Japan and Korea as: green, asparagus, celery, green beans, green herb-like, parsley, spinach, brown, ashy/sooty, burnt/scorched, tobacco, animalic, musty/new leather, seaweed, straw like, bitter, astringent and toothetch. Additionally, Vittayaporn and Chompreeda [15] studied preference mapping of Thai consumers for commercial green tea with added roasted brown rice and also described the following 18 sensory attributes: yellow color, clear, tea aroma, dry aroma, green, seaweed, roast flavor, bitter, astringent, tea aftertaste, roast after - taste, bitter after-taste and astringent after - taste.

The intensity and comparison between reference standards, DGTD tea and *C. sinensis* tea for yellowness and clearness were very similar, but odor and tea flavor were different. This could be accounted for because oolong tea, which was made from *C. sinensis*, was used as the reference standard and, therefore, had a more pronounced tea odor and flavor as would be expected. In addition, the intensity of hay odor which was absent in *C. sinensis* was found as a mild odor in DGTD tea. Fresh grass was used as a reference standard for green odor, since DGTD tea is made from rice grass leaves, which are members of the grass family [35]. DGTD tea was found to have a stronger green odor than the fresh grass. DGTD tea was quite close to *C. sinensis* green tea for seaweed odor and seaweed flavor. Japanese green teas often exhibit a fresh and marine aroma, very similar to seaweed since dimethyl sulfide is a compound, found in Japanese green tea that has been described as having a smell of seaweed as well as cooked cabbage, asparagus and sweet corn [36]. Jasmine rice odor was found in the DGTD tea that gave it a pleasant smell but it was absent in *C. sinensis* tea. When DGTD tea and *C. sinensis* tea was compared for tea flavor, *C. sinensis* tea had a higher score than DGTD tea. For bitter taste and astringent taste, DGTD tea scored less than *C. sinensis* tea, which was consistent with levels of TPC of DGTD tea and *C. sinensis* tea which was found to have different phenolic compounds. The major polyphenols

ajfand SCHOLARLY, PEER REVIEWED
AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT
Volume 19 No. 4
November 2019
PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST
ISSN 1684 5374

components in *C. sinensis* tea were reported to be flavonols containing catechin that are responsible for the bitterness and astringency taste [37] of many foods and beverages. In addition, bitter taste and astringency in DGTD tea were less than *C. sinensis* tea, which may be because the latter are rich of tannins [38]. Boonwithawacharoen [39] studied tannins in green tea, oolong tea, black tea and mulberry tea and found that *C. sinensis* tea had a higher capacity than the others. However, on balance, the attributes of the DGTD tea were similar to those of *C. sinensis* tea but with the added advantage of the jasmine odor. Jasmine rice odor was absent in *C. sinensis* tea.

CONCLUSIONS

Damgatondam upland rice grass leaves, which had been sown one month earlier, were successfully made into green tea by steaming, pan roasting and tray drying. From the experiment it was possible to define exactly the optimum processing conditions, which were steaming at 98°C for 60 sec, pan roasting at 70°C for 30 min and tray drying in an electrical tray dryer at 60°C for 90 min. The chemical composition, antioxidant activity and the sensory characteristics, from descriptive analysis of the product, showed that DGTD tea was acceptable and its composition comparable to green tea made from *C. sinensis*, but had a unique distinctive fragrant jasmine odor. It was concluded that DGTD tea was an attractive alternative to other green teas and could possibly give a supplementary income to upland rice farmers.

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

Table 1: Characteristics of different amount of DGTD rice grass leaves with different steaming times

Tea weight (g)	Steaming time (sec)	Characteristics
100	60	Dark green, without damage
100	90	Damage
100	120	Damage
200	60	Dark green, without damage
200	90	Dark green, with damage
200	120	Damage
300	60	Irregularly color
300	90	Dark green, with damage
300	120	Dark green, with damage
400	60	Irregularly color
400	90	Dark green, with damage
400	120	Dark green, with damage

Table 2: Total phenolic content (TPC) and antioxidant activities (FRAP, DPPH and ABTS assays) of rice grass leaves during pan roasting

Time for pan roasting (min)	TPC (mg GAE/g sample)	FRAP (mM TE/g sample)	DPPH (mM TE/g sample)	ABTS (mM TE/g sample)
5	$15.42^{a}\pm0.27$	$533.67^{a}\pm5.17$	$53.96^{a}\pm1.46$	$178.00^{a}\pm4.16$
10	$15.36^{a}\pm0.07$	$529.14^{ab}\pm4.68$	$53.50^{a}\pm0.47$	$161.56^{b}\pm7.73$
15	$15.21^{a}\pm0.10$	$527.24^{ab}\pm5.07$	$51.94^{a}\pm0.49$	$160.89^{b}\pm2.14$
20	$14.74^{b}\pm0.06$	$521.76^{bc}\pm3.67$	$49.60^{b}\pm1.20$	$155.56^{bc}\pm1.02$
25	$14.62^{b}\pm0.12$	$521.53^{bc}\pm6.07$	$48.58^{b}\pm0.84$	$151.78^{c}\pm1.68$
30	$14.29^{c}\pm0.45$	$516.57^{c}\pm3.60$	$47.81^{b}\pm1.56$	$150.22^{c}\pm3.29$

In each column, different superscripts represent significant differences ($p\leq0.05$)

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

Table 3: Proximate composition, mineral content, antioxidant activities and TPC of the DGTD tea and *C. sinensis* tea as determined in other publications

Parameters	DGTD content	*C. sinensis* content Ahmad *et al.* [28]	*C. sinensis* content Adnan *et al.* [29]
Water activity	0.49	-	-
Moisture content (%)	3.77	4.88	6.46
Ash (%)	7.94	5.60	4.57
Crude Fiber (%)	22.59	15.35	16.16
Fat (%)	2.58	2.49	1.68
Protein (%)	17.73	18.60	1.23
Parameters	**DGTD Content**	***C. sinensis* Content Ahmad *et al.* [28]**	***C. sinensis* Content Ramdani *et al.* [30]**
Calcium (Ca) (mg/kg)	3400	3747.5	6699
Iron (Fe) (mg/kg)	66.4	205.3	119
Magnesium (Mg) (mg/kg)	3200	652.2	1993
Manganese (Mn) (mg/kg)	139.1	-	663
Potassium (K) (mg/kg)	20400	17031.2	8095
Sodium (Na) (mg/kg)	113.8	75.5	78.2
Parameters	**DGTD content**	***C. sinensis* content Nor and Fadzelly [31]**	***C. sinensis* content Sheikh *et al.* [32]**
Total phenolic content (mg GAE/g)	9.53	72.70	74.51
Parameters	**DGTD content**	***C. sinensis* content Lee *et al.* [33]**	***C. sinensis* Content**
FRAP (mM TE/g)	357.52	1555.06	-
DPPH (mM TE/g)	34.08	1423.22	-
ABTS (mM TE/g)	108.89	4293.33	-

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

Table 4: Descriptors and definitions used by the ten trained panelists to describe sensory attributes of DGTD tea product

Descriptors	Definition
Appearance	
Yellowness	The intensity of yellowness from light yellow to dark yellow
Clearness	The clearness concerned with turbidity of the samples
Odor	
Tea odor	The aromatics concerned with oolong tea
Hay odor	The aromatics concerned with rice straw
Green odor	The aromatics concerned with fresh grass
Seaweed odor	The aromatics concerned with natural dry seaweed (*Kappaphycus alvarezii*) from a Chinese restaurant
Jasmine rice odor	The aromatics concerned with Thai steamed Khao Dawk Mali 105 rice (*Oryza sativa*)
Flavor	
Tea flavor	The aromatic and taste concerned with oolong tea
Seaweed flavor	The aromatic and taste concerned with natural dry seaweed (*Kappaphycus alvarezii*) from a Chinese restaurant after steeping in boiled water for 3 min
Taste	
Bitter taste	A basic taste factor of paracetamol in water is typical
Astringent taste	The taste compared with raw cultivated banana peel

ajfand SCHOLARLY, PEER REVIEWED AFRICAN JOURNAL OF FOOD, AGRICULTURE, NUTRITION AND DEVELOPMENT Volume 19 No. 4 November 2019 PUBLISHED BY AFRICAN SCHOLARLY SCIENCE COMMUNICATIONS TRUST ISSN 1684 5374

Table 5: Standard references of sensory attributes and intensity scores given by ten trained panelists in the descriptive analysis of DGTD tea and *C. sinensis* tea bag from Japan

Attributes	Reference standards	Intensity reference standard	Intensity DGTD (SD)	Intensity *C. sinensis* (SD)
Appearance				
Yellowness	- Munsell book scales	-	6.5 (0.68)	6.3 (0.47)
	- 5Y8/2	0	-	-
	- 5Y8/4	7.5	-	-
	- 5Y8/6	15	-	-
Clearness	- C29 Distilled water	0	8.5 (0.64)	8.1 (0.55)
	- Wheat flour solution 0.01g: 100mL water	13	-	-
Odor				
Tea odor	- Oolong tea bag steeping 3 min in 150 mL water (98°C)	10	3.2 (0.67)	6.7 (0.58)
Hay odor	- Dry rice straw	13	0.68 (0.33)	0
Green odor	- Fresh grass	7	2.5(0.56)	0.7 (0.33)
Seaweed odor	- Natural dry 'Sakol' seaweed (*Kappaphycu salvarezii*)	15	12.7 (0.84)	10.9 (0.91)
Jasmine rice odor	- 30 g of cooked Khao Dawk Mali 105 rice (*Oryza sativa*) warm at 70°C	9	3.7 (0.90)	0
Flavor				
Tea flavor	- Oolong tea bag steeping 3 min in 150 mL water (98°C)	10	2.4 (0.61)	5.6 (0.68)
Seaweed flavor	- Natural dry 'Sakol' seaweed (*Kappaphycu salvarezii*) for 5 g steeped for 3 min in boiled water	15	7.5 (0.89)	8.2(0.75)
Bitter taste	- Paracetamol solution 0.5g: 100 mL water	10	3 (0.72)	7 (0.30)
Astringent taste	- Cultivated banana peel (2 g of totally green banana peel)	7.5	0.7 (0.43)	6.4 (0.58)

Values in parenthesis were standard deviation (SD) of panels mean scores

REFERENCES

1. **Cabrera C, Artacho R and R Gimenez** Beneficial effects of green tea - a review. *J. Am. Coll. Nutr*. 2006; **25:**79-99.

2. **Food and Drug Administration Guideline for considering herbal tea** (in Thai). http://www.rayongfda.org/photo_re/News_20160318111747.pdf (Accessed 20 March 2016).

3. **Ravikumar C** Review on herbal teas. *J. Phar. Sci & Res*. 2014; **6:**236-283.

4. **Somrit B** Upland rice Phrae (in Thai). Phrae Rice Research Center: Department of Agriculture Thailand Ministry of Agriculture and Cooperatives. 1988.

5. **Nokkoul R** Upland black waxy rice var. Ka Ton Dam (in Thai). http://upland-rice.blogspot.com/2016/03/upland-rice.html/ (accessed 26May 2016).

6. **Sudtasarn G, Jongdee S and R Kenlhaem** Research and development of green tea from fragrant (in Thai). 2008. https://dspace.tarr.arda.or.th/ handle/6622815955/ 9860 (Accessed 5 December 2016).

5. **Yun P** Freeze dried powder of selected Thai upland-rice grass juice. Thesis for Master of Food Science, Faculty of Agro-Industry, King Mongkut's Institute of Technology Ladkrabang, Thailand. 2016.

8. **Alvarez MV, Moreira MR and A Ponce** Peroxidase and sensory quality of ready to cook mixed vegetables for soup: combined effect of biopreservatives and refrigerated storage. *J. Food Sci. Tech.* 2015; **35:**86-94.

9. **Ministry of Public Health** Tea (In Thai). http://taxclinic.mof.go.th/pdf/ 417D 2E7B _DDE9_AC5E_A773_B5F37A9733E8.pdf 2000. (Accessed 18 Oct 2016).

10. **Yun P, Thamakorn P, Nokkoul R, Suwapanich R and K Huangrak** Antioxidant activity and bioactive compounds of freeze-dried product from Thai upland rice grass juice. In: Proceeding of the 17th Food Innovation Asia Conference, Innovative ASEAN Food Research towards the World, Bangkok, Thailand. 2015: 35-41.

11. **Wu HC, Chen HM and CY Shiau** Free amino acids and peptides as related to antioxidant properties in protein hydrolysates of mackerel (*Scomber austriasicus). Food Res. Int.* 2003; **36:**949-957.

12. **AOAC**. Official Methods of Analysis, Association of Official Analytical Chemist Inc. 19th Ed. Washington, DC, USA. Official methods 978.18, 925.10, 945.38, 981.10, 922.06, 978.10, 984.27, 984.27, 984.27, 999.10, and 984.27. 2012.

13. **Meilgaard M, Civille GV and TB Carr** Sensory evaluation techniques. In: Raton, B. (edition). 3rd edition. CRC press. http://dx.doi.org/10.1201/9781439832271 1999. (Accessed 18 September 2016).

14. **Stone H and JL Sidel** Sensory evaluation practices. Academic Press Inc., Tragon Corporation, Redwood City. 2004: 408.

15. **Vittayaporn V and P Chompreeda** Preference mapping of Thai consumers for commercial green tea with roasted brown rice. *Kasetsart J. (Natural Science).* 2010; **44:** 652-663.

16. **Wang H, Helliwell K and X You** Isocratic elution system for the determination of catechins caffeine and gallic acid in green tea using HPLC. *Food Chem.* 2000; **68:**115-121.

17. **Uhl JW** The art and craft of tea: An enthusiast's guide to selecting, brewing, and serving exquisite tea. Quarto Publishing Group USA Inc. 2016; 160.

18. **Kawakami M and T Yamanishi** Formation of aroma component in roasted or pan-fired green tea by roasting or pan-firing treatment. *J. Jpn. Soc. Biosci. Biotechnol. agrochem.* 1999; **73;**893- 906.

19. **Choi SH** Effect on pyrazines, reaction products of Maillard reaction in flavor of green tea and substation tea. *J. Kor. Tea Soc*. 2010; **16:**1-7.

20. **Zheng XQ, Li QS, Xiang L and YR Liang** Recent advances in volatiles of teas: a review. *Molecules*. 2016; **21:**1-12.

21. **Lorsuwan P, Rachtanapun C and S Chantanawaranggoon** Total phenolics, radical scavenging capacity and antimicrobial property of fruit peels. In: Conference of Kasetsart University 46th (in Thai), Bangkok, Thailand. 2008: 554-561.

22. **Zzaman W, Bhat R and TA Yang** Effect of superheated steam roasting on the phenolic antioxidant properties of cocoa beans. *J. Food Process Pres*. 2013; **38:** 1932-1938.

23. **Temple SJ and AJ B Boxtel** Thin layer drying of black tea. *J. Agri. Eng. Res.*1999; **74:**167-176.

24. **Winyangkul S, Saleesongsom S and N Moolcharoen** Estimation of moisture content in tea leaves, in the drying process. *Agri. Sci. J.* 2007; **38:**255-258.

25. **Keeratiburana T and V Srijesdaruk** Effect of tray drying of germinated brown KDML 105 on bioactive compounds. In: Proceeding of the 11th KhonKaen University Graduate Research Conference, KhonKaen, Thailand. 2011:729-734.

26. **Lopez-Vidana EC, Figueroa IP, Cortes FB, Rojano BA and AN Ocana** Effect of temperature on antioxidant capacity during drying process of mortino (*vacciniummeridionale Swartz*). *Int. J. Food. Prop.* 2016; **20:**294-305.

27. **Rungsirisakun R, Techavutthiporn C and P Boonyarithongchai** Optimization of drying process condition of chamomile (*Matricaria chamomile* L.) using developed prototype dryer on bioactive compound (in Thai). *Agri. Sci. J.* 2015; **46:**861-863.

28. **Ahmad RS, Butt MS, Huma N, Sultan MT, Arshad MU, Mushtaq Z and F Saeed** Quantitative and qualitative portrait of green tea catechin (gtc) through hplc. *Int. J. Food Prop.* 2014; **17:**1626-1636.

29. **Adnan M, Ahmad A, Ahmad A, Khalid N, Hayat I and I Agmed** Chemical composition and sensory evaluation of tea (*Camellia sinensis*) commercialized in Pakistan. *Pak. J. Bot.* 2013; **45:** 901-907.

30. **Ramdani D, Chaudhry AS and CJ Seal** Chemical composition, plant secondary metabolites, and minerals of green and black teas and the effect of different tea-to-water ratios during their extraction on the composition of their spent leaves as potential additives for ruminants. *J. Agric. Food Chem.* 2013; **61:**4961-4967.

31. **Nor QhairulIzzreen MN and AB Fadzelly** Phytochemicals and antioxidant properties of different parts of *Camellia sinensis* leaves from Sabah tea plantation in Sabah, Malaysia. *Int. Food Res. J.* 2013; **20:**307-312.

32. **Sheikh RE, Amin AS, Atwa MA, Gouda AA and AA Abdullah** Determination of phenolic components and antioxidant activity of some Egyptian tea samples. *Int. J. Pharm. Pharm. Sci.* 2015; **7:**198-202.

33. **Lee LS, Kim SH, Kim YB and YC Kim** Quantitative analysis of major constituents in green tea with different plucking periods and their antioxidant activity. *Molecules.* 2014; **19:**9173-9186.

34. **Lee J and DH Chamber** Descriptive analysis and U.S. consumer acceptability of 6 green tea samples from China, Japan and Korea. *J. Food Sci.* 2010; **75:**141-147.

35. **Ricepedia** (The online authority on rice). Rice as a plant. Program on Rice. http://ricepedia.org/rice-as-a-plant (Accessed 22 November 2017).

36. **Caicedo R** Why does Japanese Green Tea Smell like Seaweed. https:// www.myjapanesegreentea.com/why-japanese-green-tea-smells-like-seaweed (Accessed 22 December 2017).

37. **Drewnowski A and C Gomes-Carneros** Bitter taste, phytonutrients, and the consumer: a review. *Am. J. Clin. Nutr.* 2000; **72:**1424-1435.

38. **Asok PK and K Upadhyyaya** Tannins are Astringent. *J. Pharm. Phy.* 2012; **1:**45-50.

39. **Boonwithawacharoen O** Affecting antioxidant contents in pasteurized tea (In Thai). Thesis for Master of Food science, Faculty of Agro-Industry, King Mongkut's Institute of Technology Ladkrabang, Thailand. 2006.