



Antioxidant Activity and Phenolic Content in Herbal Tea in Inflorescences and Mango (*Mangifera indica*) Leaves

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Received: 31 August 2023/ Revised: 10 December 2023/ Accepted: 15 December 2023

Abstract

The mango (*Mangifera indica*) fruit is an economic fruit. Thailand is the third-largest mango exporter after the Philippines and Mexico. Mango leaves are reported to have anti-inflammatory properties, blood sugar lowering effect, and blood lipid-lowering effect. Mango leaves are generally not being interested much and considered waste in agriculture despite their medicinal properties. To create a herbal tea for health enhancement extracts from mango leaves and inflorescences were examined in this study. The investigation of the antioxidant properties of crude mango inflorescence and leaf extracts was determined to examine the antioxidant activities of crude extracts, two tests were performed: 1) scavenging capacity, such as the ability to eliminate or suppress free radicals, as measured by the DPPH method, and 2) the ability to function as an electron donor or what is called a “reducing agent”. The FRAP test revealed that tea made from Man Duan Kao mango leaves had the highest anti-oxidant activity (53.65 ± 0.70 g FeSO₄/100 g tea powder), and it was discovered that mango inflorescences deliver more antioxidants than leaves (57.60 ± 1.49 g FeSO₄/100 g tea powder). The total phenolic and flavonoid contents of the crude extracts of tea from mango inflorescences and leaves revealed that tea from *Mangifera indica* leaves tended to have a higher total phenolic content. The largest levels of catechins and flavonoids were identified in both green and oolong tea, while mango inflorescence tended to provide overall phenolic content and more flavonoids than leaves, which results in consistent outcomes. Colorimetric examination (CIE-Lab) showed no variation in b*, C*, and h values, except an increase in illuminance (L*) values for samples cured for shorter treatment durations inversely related to the value of redness (a*) in samples undergoing a lengthy preparation procedure.

Keywords: Antioxidant activity, Phenol, Mango inflorescence, Mango leaves, Herbal tea

Introduction

Mango (*Mangifera Indica*) is a fruit that is cultivated as an agricultural crop. Thailand is the third largest mango exporter behind the Philippines and Mexico. Mangos are well-liked by both domestic and foreign consumers. Suphan Buri Province which is one of the campuses of Suan Dusit University has a lot of mango



plantations, namely a total area of 3,348,755 rai, the majority of which is utilized for agriculture 2,316,357 rai (69.17%), 19,684 rai (0.85%) of which are fruit orchards. Mango is the most widely planted fruit tree, with a plantation area of 35,002.50 rai, a total production of 25,292.49 tons, an average yield of 1.45 tons per rai, and a farm price per ton of 21,780.00 baht. Community enterprises in the district of Mueang Suphan Buri in the province of Suphanburi cultivate three varieties of mango, namely Nam Dok Mai, Khiew Sawoey (*Mangifera indica* L.), and Man Duan Kao (*Mangifera Indica*) on a plot of land comprising approximately 3,000 rai. In an interview with the enterprise's president, it was discovered that the output of mangoes was significantly lower than anticipated, particularly during the off-season. Throughout the production season, there is an issue with the product's lowering price. In addition, there is an issue with mold resulting in inadequate production for processing. In mango plantations, there are also several non-profitable parts: mango leaves are over 4,800 tons per year, mango inflorescence is over 2,400 tons per year, and mango branches are over 7,200 tons per year [1].

It has been reported that mango leaf has anti-inflammatory and antioxidant qualities, and hypoglycemic and hypolipidemic activities [2]. Between March and May, 5-7 young leaves from the shoots are soft and yellow-green and can be cooked in water [3]. Mango leaf tea has been used in Ayurveda medicine to treat diabetes [4]. Mango leaves have a bitter flavor and help lower body temperature, and are used to treat constipation. Coughing, asthma, sneezing, throat burning, hemorrhages, hemorrhoids, wounds, abscesses, gastritis, diarrhea, dysentery, liver diseases, tooth decay, throat, scorpion poisoning, and canker sores are also treated with mango leaves. Burnt-leaf ash is used to cure burns and scalds [5]. Mango leaf smoke is smoked to treat hiccups and sore throat. In Guangxi, fresh leaves were pulverized for gum toning [6] and used as a cough treatment [7]. Moreover, they help resolve diarrhea and sleeplessness [8]. Due to the presence of Mangiferin, Quercetin, Catechin, Gallic acid, 3-taraxerol, Ethyl gallate, Gallotannins, and Benzophenones, mango leaves have therapeutic qualities [9-11].

The leaves of the *Camellia sinensis* (L.) tea plant are used to produce the most commercially available tea [12]. The types of tea can be categorized into 3 types namely, 1) Green tea is a type of tea that does not undergo fermentation or non-fermented tea; By steaming or roasting on a pan firing, the enzyme polyphenol oxidase is deactivated in fresh tea leaves It can hasten the oxidation and polymerization of tea polyphenols in the leaves, causing dimers and complexes of polyphenols. After that, knead (rolling) to make the cells break and knead to make the tea leaves curl and dry. The color of this type of tea is green to yellowish green. 2) Oolong tea is semi-fermented tea before ceasing the enzymatic reaction with heat. The production process involves withering for about 20 to 40 minutes. This aeration is a fermentation process in which the enzyme polyphenol oxidase catalyzes the oxidation and polymerization of polyphenols to form dimers and complexes of polyphenols. Oolong tea is greenish yellow to greenish brown. 3) Black tea is a type of tea that has been completely fermented. The tea leaves are fully catalyzed by polyphenol oxidase, and the polyphenols are completely oxidized to form theaflavins and thearubigins, giving black tea a reddish-brown color.



Mango leaves receive little attention and are categorized as agricultural waste despite having therapeutic characteristics. In this study, the analysis from the extracts of inflorescences and mango leaves was studied in order to generate a herbal tea for health enhancement.

Objective

To study the antioxidant activity and the content of phenol in healthy herbal tea products made from mango inflorescences and leaves.

Material and Methods

1. Tea preparation

Mango (*Mangifera indica*) inflorescences and leaves were collected in January, 2022 from organic field of Sanam khli community enterprise area (14°33'N, 99°57'E), Tambon Sanam khli, Mueang District, Suphanburi Province (Thailand). The 3 different varieties of mango including Nam Dok Mai, Kiew Savoey, and Man Duan Kao mango were studied. The 3 tea processes including green tea (non-fermented tea), oolong tea (semi-fermented tea), and black tea (fermented tea) used for prepare mango (*Mangifera indica*) inflorescences and leaves tea as described previously [12]. The fine powder obtained (2 g) was blended with 50 ml of water (4% of final concentration), boiled for 10 min, and then filtered (Whatman No.4).

2. Determination of DPPH radical scavenging activity

Radical 2, 2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activity was measured according to the method of Xie *et al.* [13] with a slight modification. About 24 mg of DPPH was added to 100 ml methanol to prepare the stock solution, and then stored at -20 °C in the dark. About 8 ml stock solution was diluted with 40 ml methanol to prepare the working DPPH solution with an absorbance of 1.1 ± 0.02 units at 515 nm. The 0.1 mL tea extract was mixed with 0.1 mM DPPH solution for 0.9 mL. The resulting solution was incubated for 30 min at room temperature in the dark, and the absorbance was measured at 517 nm. Trolox was used as the standard and methanol as the blank, and results were expressed as mg Trolox equivalents (TE)/g db.

3. Determination of the Ferric Reducing Antioxidant Power (FRAP)

The 2, 4, 6-tripyridyl-s-triazine (TPTZ) of 2.5 mL was prepared in 40 mM of hydrochloric acid, 2.5 mL of 20 mM iron (III) chloride was prepared, and 300 mM of acetate buffer was prepared at the volume of 20 mL, and all the solution was mixed. Analyses were performed by pipetting 0.05 ml of the sample with 0.95 ml of FRAP solution and leaving it in the dark for 30 min. The absorbance was measured at a wavelength of 595 nm using ferrous sulfate as a standard.

4. Determination of total phenolic content

Total phenolic was estimated calorimetrically using the Folin-Ciocalteu reagent adapted from Tang, *et al* [14]. An aliquot of 60 μ l of the extract solution and 60 μ l methanol were mixed with 750 μ l of 10% Folin-Ciocalteu's reagent solution (v/v) and incubated for 8 min at room temperature. Then, 600 μ l of 7.5%

(w/v) sodium carbonate solution was added, and the mixture was incubated at room temperature for 1 h in the dark. The absorbance was measured by a UV-Vis spectrophotometer (M501 Single Beam Scanning, Camspec, Leeds, UK) at 765 nm. Gallic acid was used as the standard and methanol as blank, and the results were expressed as mg gallic acid equivalents (GAE)/g dry basis (db).

5. Determination of total flavonoid content

Total flavonoid content was measured by the aluminium chloride colorimetric method according to Hamrouni-Sellami, *et al* [15] with a slight modification. An aliquot of 250 μ l of the extract solution was mixed with 75 μ l of 5% NaNO₂ solution (w/v) and incubated for 6 min at room temperature. Then 150 μ l of 10% (Al(NO₃)₃)₃ solution (w/v) was added to the mixture and incubated for 6 min at room temperature. Next, 500 μ l of 1M NaOH solution was added, and the mixture was incubated for 30 min at room temperature in the dark. The absorbance was measured by the M501 UV-Vis spectrophotometer at 510 nm. An absorbance was measured relative to the Quercetin standard and the results were expressed as mg Quercetin equivalents (CAE)/g db.

Results and Discussion

In the investigation of the antioxidant activity of crude mango inflorescence and leaf tea extracts by determining the antioxidant properties of crude extracts, two tests were performed: 1) scavenging capacity, i.e., the ability to destroy or inhibit free radicals, as measured by the DPPH method; and 2) the property of being a substance that acts as an electron donor, called a reducing agent, was tested by FRAP method. The experimental results are shown in Table 1. Free radical scavengers were highest in both green tea and oolong tea and it was discovered that mango inflorescences tended to provide more antioxidants than leaves due to the presence of relatively high total phenol, flavonoids, as well as total carotene contents [16].

Table 1 Antioxidant activity of crude extracts of tea from mango inflorescences and leaves

Sample	DPPH	FRAP
	(A gram of Trolox equivalent/100 g tea powder)	(A gram of FeSO ₄ /100 g tea powder)
Nam Dok Mai mango green tea (leaves)	8.81±0.30 ^k	34.13±0.42 ^g
Kiew Savoey mango green tea (leaves)	14.92±0.48 ^f	38.23±0.24
Man Duan Kao mango green tea (leaves)	17.31±0.38^e	53.65±0.70^b
Nam Dok Mai mango oolong tea (leaves)	8.40±0.24 ^k	27.51±0.65 ^h
Kiew Savoey mango oolong tea (leaves)	11.72±0.39 ^j	43.94±0.96^e
Man Duan Kao mango oolong tea (leaves)	16.49±0.30^e	39.73±0.64 ^f



Sample	DPPH	FRAP
	(A gram of Trolox equivalent/100 g tea powder)	(A gram of FeSO ₄ /100 g tea powder)
Nam Dok Mai mango black tea (leaves)	4.49±0.06 ^l	10.93±0.23 ^k
Kiew Savoey mango black tea (leaves)	4.71±0.14 ^l	12.48±0.11 ^j
Man Duan Kao mango black tea (leaves)	5.02±0.15^l	13.67±0.08ⁱ
Nam Dok Mai mango oolong tea (inflorescences)	28.54±0.93 ^c	57.60±1.49^a
Kiew Savoey mango oolong tea (inflorescences)	31.93±1.10 ^b	50.72±0.40 ^c
Man Duan Kao mango oolong tea (inflorescences)	37.76±0.98^a	53.23±1.45 ^b
Nam Dok Mai mango black tea (inflorescences)	28.25±0.43^c	51.37±0.54^c
Kiew Savoey mango black tea (inflorescences)	13.02±0.44 ⁱ	27.35±0.33 ^h
Man Duan Kao mango black tea (inflorescences)	23.43±2.14 ^d	48.75±0.18 ^d

Remark: The results were expressed as average ± standard deviation. The difference letters represented significant difference at $p < 0.05$.

Results of the study on the total phenolic and flavonoid contents of tea crude extract from mango inflorescences and leaves according to the experiment are shown in Table 2. It was found that the phenolic content of the tea made from mango leaves was close to that of the whole plant. Both green tea and oolong tea had the most flavonoids, and the inflorescence of mango plants had the most phenolics overall and more flavonoids than leaves, which produce consistent results.

Table 2 Total amount of phenolic compounds and total flavonoid compounds of tea from mango inflorescences and leaves

Sample	Total phenolic content	Total flavonoid content
	(A gram(s) of Gallic acid equivalent/tea powder gram(s))	(A gram(s) of Quercetin equivalent/tea powder gram(s))
Nam Dok Mai mango green tea (leaves)	7.07±0.01 ^e	7.31±0.30 ^d
Kiew Savoey mango green tea (leaves)	9.11±0.41 ^c	9.04±0.30 ^b
Man Duan Kao mango green tea (leaves)	10.32±0.02^b	10.10±0.12^a



Sample	Total phenolic content	Total flavonoid content
	(A gram(s) of Gallic acid equivalent/tea powder gram(s))	(A gram(s) of Quercetin equivalent/tea powder gram(s))
Nam Dok Mai mango oolong tea (leaves)	6.33±0.09 ^f	6.45±0.23 ^f
Kiew Savoey mango oolong tea (leaves)	7.27±0.04 ^d	6.98±0.35 ^{de}
Man Duan Kao mango oolong tea (leaves)	9.01±0.24^c	8.57±0.20^c
Nam Dok Mai mango black tea (leaves)	2.14±0.02 ⁱ	1.15±0.05 ^h
Kiew Savoey mango black tea (leaves)	2.07±0.03 ⁱ	1.10±0.00 ^h
Man Duan Kao mango black tea (leaves)	2.54±0.03^h	1.21±0.04^h
Nam Dok Mai mango oolong tea (inflorescences)	10.05±0.47 ^b	5.85±0.12 ^s
Kiew Savoey mango oolong tea (inflorescences)	11.56±0.37^a	7.04±0.30^{de}
Man Duan Kao mango oolong tea (inflorescences)	11.42±0.10 ^a	5.52±0.12 ^s
Nam Dok Mai mango black tea (inflorescences)	8.42±0.03^d	6.86±0.40^e
Kiew Savoey mango black tea (inflorescences)	4.37±0.01 ^s	3.84±0.02
Man Duan Kao mango black tea (inflorescences)	6.87±0.05 ^e	3.89±0.04

Remark: The results were expressed as average ± standard deviation. The difference letters represented significant difference at $p < 0.05$.

Colorimetric analysis (CIE-Lab) showed that the b^* , C^* , and h values were the same. Only the illuminance (L^*) values were more likely to be different in the samples that were treated for less time; it is in a reverse direction of the redness (a^*) value in the samples that took longer to prepare, as shown in Table 3. Oxidation describes the process of enzymes in the tea leaves reacting with oxygen. During oxidation, leaves undergo changes in their chemical composition turning them brown and changing the flavor and aroma profile. Oxidation is an essential step in the processing of tea. Depending on the type of tea which is made, oxidation is either fully prevented or intentionally caused and stopped at a certain level. Whereas green tea are unoxidized or lightly oxidized, black teas are highly or fully oxidized. In between are oolong



teas, which cover a wide range of oxidation levels from lightly oxidized to highly oxidized. The process of oxidation is intentionally caused by crushing, rolling or tumbling the leaves. Thereby the walls of the cells are damaged, exposing the inside of the leaves with oxygen, resulting in a series of chemical reactions. In order to stop the oxidation at a certain level, leaves have to be heated or dried. Heat denatures the enzymes responsible for oxidation. The sensorial scores for overall acceptability indicated that the products were highly accepted by the panelist.

Table 3 Colorimetric values (CIE) of tea from mango inflorescences and leaves

Sample	Colorimetric Value (CIE-Lab)				
	L*	a*	b*	C*	H*
Nam Dok Mai mango green tea (leaves)	51.83±0.82 ^b	2.86±0.23 ^{abcd}	12.02±1.05 ^b	11.29±1.39 ^b	74.73±1.50 ^a
Kiew Savoey mango green tea (leaves)	54.77±1.29 ^a	2.64±0.10 ^{abcd}	13.74±0.22 ^a	13.99±0.23 ^a	79.14±0.29 ^a
Man Duan Kao mango green tea (leaves)	54.12±0.74 ^a	2.45±0.10 ^{abcd}	12.79±0.78 ^{ab}	13.03±0.79 ^a	79.11±0.36 ^a
Nam Dok Mai mango oolong tea (leaves)	46.88±1.05 ^d	4.16±0.40 ^{ab}	7.59±1.05 ^d	8.67±1.12 ^c	60.87±1.04 ^{bc}
Kiew Savoey mango oolong tea (leaves)	47.51±0.17 ^d	4.60±0.02 ^a	8.93±0.08 ^{cd}	10.05±0.08 ^{bc}	62.58±0.04 ^{bc}
Man Duan Kao mango oolong tea (leaves)	49.31±1.20 ^c	4.15±0.37 ^{ab}	9.87±1.13 ^c	10.74±1.21 ^b	65.75±1.90 ^b
Nam Dok Mai mango black tea (leaves)	40.95±0.55 ^{ef}	1.28±0.19 ^{cd}	1.67±0.65 ^{efgh}	1.82±0.24 ^{defg}	44.27±2.92 ^{ef}
Kiew Savoey mango black tea (leaves)	40.51±0.26 ^{ef}	3.56±4.31 ^{abc}	1.06±0.59 ^{fgh}	1.72±0.69 ^{efg}	27.73±9.45 ^h
Man Duan Kao mango black tea (leaves)	41.49±1.75 ^e	1.91±1.26 ^{bcd}	2.23±1.26 ^{efg}	3.21±2.70 ^{de}	42.38±12.32 ^{ef}
Nam Dok Mai mango oolong tea (inflorescences)	37.82±0.64 ⁱ	1.07±0.04 ^d	1.21±0.03 ^{fgh}	1.62±0.05 ^{efg}	48.44±0.39 ^{de}
Kiew Savoey mango oolong tea (inflorescences)	39.12±0.11 ^{fi}	1.56±0.14 ^{cd}	2.30±0.26 ^{ef}	2.78±0.29 ^{def}	55.69±1.01 ^{cd}



Sample	Colorimetric Value (CIE-Lab)				
	L*	a*	b*	C*	H*
Man Duan Kao mango oolong tea (inflorescences)	40.48±0.12 ^e	1.67±0.19 ^{cd}	3.10±0.27 ^e	3.52±0.32 ^d	61.75±1.22 ^{bc}
Nam Dok Mai mango black tea (inflorescences)	38.30±0.13 ⁱ	1.0±0.30 ^d	1.85±1.85 ^{efgh}	1.27±0.44 ^{fg}	36.90±3.66 ^{fg}
Kiew Savoey mango black tea (inflorescences)	39.00±0.13 ⁱ	1.22±1.16 ^{cd}	0.32±0.09 ^h	0.62±0.09 ^s	32.77±3.66 ^{gh}
Man Duan Kao mango black tea (inflorescences)	38.01±0.38 ⁱ	0.72±0.12 ^d	0.51±0.02 ^{sh}	0.87±0.24 ^s	32.87±5.78 ^{sh}

Remark: The results were expressed as average ± standard deviation. The difference letters represented significant difference at $p < 0.05$

Inhibitors of Advanced Glycation Endproducts (AGEs) have now been identified as food or pharmaceutical ingredients, containing polyphenols like flavonoids, curcumin, phenolic acids, terpenoids, unsaturated fatty acids, and vitamins [17]. Overall, natural products containing polyphenols have received considerable attention due to their superior antimicrobial properties and high food safety. The majority of polyphenols are effective antioxidants. In addition, there are additional biological effects, including a reduction in blood sugar levels, against cancer cells, anti-inflammatory, anti-aging, and heart disease prevention [18, 19].

Conclusion

The antioxidant activity, phenolic and flavonoid content of teas derived from mango inflorescences and leaves in three forms, namely green tea, oolong tea, and black tea, were evaluated in preliminary experiments. They differ in appearance, color, odor, and flavor. It depends on two key factors: the chemical composition of the tea leaves and the temperature at which the tea is brewed, and the tea manufacturing procedure. The various chemical compositions of tea leaves are the result of the various tea strains, climate and soil conditions in the planting area, abundant soil, water, and preservation. The various chemical compositions will influence the chemical reactions that occur during the manufacturing process. Consequently, different aromas and flavors are imparted to tea.

Suggestion

This is merely an exploratory study. Only antioxidant content was displayed. However, the results of the study indicate the potential health benefits of herbal tea made from mango inflorescences and



leaves. It increases the product's value. Therefore, mango plantation owners and herbal tea manufacturers earn more money from the sale of herbal tea made from mango inflorescences and leaves. Moreover, this can help reduce the amount of agricultural waste produced by the mango plantation while promoting the well-being of people in the pre-aging age group and the elderly. The main uses of natural food ingredients derived mango by-products, especially mango inflorescence (flower), were considered further study because of cheap sources of valuable food and nutraceutical ingredients.

Acknowledgments

This research was supported by the Foundation Work Grant of Suan Dusit University. Fiscal Year 2022 Thank you Sanam Khli Mango Community Enterprise Export Sanam Khli Sub-district, Suphan Buri Province (Registration Code 2-72-01-20/1-0008).

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