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Development and evaluation of functional property of guava leaf based herbal tea

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Abstract
Herbal teas have been gaining popularity as consumers believe that they are natural, safe and can promote health. In the present study, guava leaf based herbal teas were developed with the combination of supporting and activating herbs like guava leaf, coriander leaf, dry ginger and lemongrass. Guava leaves were subjected to two different processing conditions *viz.*, steam blanching and hot water blanching. The developed herbal combinations were packed as dip tea bags. Total polyphenol content (TPC), total flavonoid content (TFC), tannin, total antioxidant activity and oraganoleptic characteristics were investigated in fresh guava leaves as well as in herbal combinations by standard methods. Results indicated that the steam blanched herbal tea combinations exhibited highest total phenol content (113.5mg GAE/g), total flavonoid content (5.729mg QE/g) and tannin content (1.145mg CE/g) than hot water blanched herbal tea combinations. The antioxidant activity of the fresh guava leaves and developed herbal tea combinations ranged from 71.16-91.28% RSA. Organoleptic evaluation revealed that steam blanched guava leaf based herbal teas had highest overall acceptability. Based on the results of this study, it can be concluded that guava leaves have potential antioxidant activity and can be an alternative to commercial tea (*Camellia sinensis*) with various health benefits.

Keywords: Guava leaves, processing, herbal tea, functional properties

Introduction

Guava *(Psidium guajava)*, belonging to the family of Myrtaceae, grows in all the tropical and subtropical areas of the world, adapts to different climatic conditions but prefers dry climates [1]. Guava leaves contains quercetin, ferulic acid, protocatehuic acid, guavin B, asiatic acid and β-carotene that has been known to have antioxidant activity [2]. The pharmacological research *in vitro* as well as *in vivo* has been widely used to demonstrate the potential of the extracts from the leaves for the co-treatment of different ailments with high prevalence worldwide, upholding the traditional medicine in cases such as diabetes mellitus, cardiovascular diseases, cancer, and parasitic infections [3]. Coriander is a valuable herb in promoting digestion and treating gastro-intestinal disorders such as dyspepsia, flatulence, loss of appetite, griping pain and vomiting [4]. Ginger contains pungent constituents such as gingerols and shogaols which exhibit biological activities, ranging from anticancer, anti-oxidant, antimicrobial, anti-inflammatory and anti-allergic to various central nervous system activities [5]. Studies indicate that lemongrass possesses various pharmacological activities such as anti-amoebic, anti-bacterial, anti-diarrheal, anti-filarial, anti-fungal and anti-inflammatory properties [6].

Tea is one of the most widely consumed beverages worldwide, second only to water [7]. Herbal teas and medicinal plant formulations are produces from green and dried herbs, flowers, fruits, leaves, seeds, barks and roots of medicinal plants and sold in a loose form or packed in bags. The herbal teas and medicinal plant formulations are consumed in many parts of the world due to their therapeutic and healing properties [8]. In recent times, there is renewed interest in functional beverage because of growing consumer awareness of health benefits derived from tea consumption. Tea therefore belongs to a rapidly expanding market of 'wellness beverages' [9].

In this context, antioxidants especially derived from natural sources such as plants and herbs require special attention. Antioxidants neutralize the toxic and 'volatile' free radicals. There are many flavored green teas and popular flavored green teas are lemon green tea, ginger & mint green tea, lemon honey green tea, jasmine green tea, etc. It is thus imperative to research the potential of native plant materials in the expansion of new flavored green tea [10]. Green teas are normally prepared from young leaves, leaf buds and internodes of varieties of the tea plant *Camellia sinensis* or *Camellia assamica*. It contains caffeine and it has been used to increase alertness. Caffeine is a bitter substance that stimulates central nervous system that leads to harmful effects on human body.

Therefore, herbal teas were developed using guava leaves to improve the taste, aroma and health promoting properties which can complement commercial green tea.

Materials and Methods

Sample Collection

The fresh leaves of seven years old *Psidium guajava* plant leaves were collected from the orchard, Tamil Nadu Agricultural University, Madurai. They were middle age intense green leaves. The environmental conditions had mean maximum/minimum temperature of 31.5/18.5 °C, precipitation of 4mm, and saturated light duration of 8.31 hours. Dry ginger and coriander leaves and lemongrass leaves were purchased from the local shops in Madurai city.

Standardization of processing methods of guava leaves

Guava leaves were subjected to two different processing conditions to obtain maximum retention of bioactive compounds. Fresh guava leaves were carefully inspected and all foreign materials removed and then gently rinsed in tap water. In ordinary processing, the cleaned guava leaves were spread thinly on aluminium trays of the cabinet drier and dried at a temperature of 60 °C for 6 hours. In steam blanching, the cleaned guava leaves were subjected to steaming at 90 °C for 3 minutes followed by drying in cabinet drier (60 °C for 6 hours). In case of hot water blanching, the cleaned guava leaves immersed in hot water at 85 °C for 2 minutes followed by drying in cabinet drier (60 °C for 6 hours). The coriander leaves and lemongrass were also cleaned and dried in cabinet drier for 4 hours at 60°C. Herbal tea powder was prepared according to method described by Giao *et al.,* (2009) [11]. The above processed and dried guava leaves were powdered using blender and passed through aluminum sieve (1mm) to get uniform particle size for herbal tea powder.

Formulation of blended herbal tea powder

The guava leaf based functional herbal tea was standardized by preparing teas with different combination. The combination which had most acceptable sensory score was the combination of 75% of guava leaves, 15% of coriander leaves & dry ginger and 10% of lemongrass. The prepared herbal tea powder was made as dip tea bag. Generic empty tea bags with string heal seal filter paper, sold by Fashlady, China was purchased for the preparation of dip tea bag.

Preparation of herbal tea infusions

The herbal tea was prepared by infusing tea bag which contained 3g of herbal tea powder in 150 ml boiling water for 3 minutes according to method suggested by Horzic *et al.,*(2009) [12].

Analysis of bioactive compounds for antioxidant activity

Sample Extraction

The infused teas were filtered through a Whatman filter paper No. 41. Then the tea samples were extracted with 80% ethanol by centrifugation @3000rpm for 20 minutes. The supernatant thus was collected and stored at 4°C for further analysis.

Total Phenol Content (TPC)

The total phenol content in herbal tea sample was determined by using the Folin-Ciocalteu method [13]. The absorbance was read at 765 nm by UV-VIS Spectrophotometer. The TPC of herbal tea sample was expressed as mg of gallic acid equivalents (mg GAE)/ml of tea sample.

Total Flavonoid Content (TFC)

The total flavonoids content was analysed according to method as described by Singh *et al.* (2012) [14]. The absorbance was measured at 510 nm using UV-VIS Spectrophotometer. The results were expressed as mg of quercetin equivalents (mg QE)/ ml of tea sample.

Total anti-oxidant activity (DPPH method)

The DPPH assay was performed according to procedure as described by Nuengchamnong and Ingkaninan (2010) [15]. The absorbance was measured using UV-VIS Spectrophotometer at 515 nm where methanol was used as blank. The results were expressed as% of radical scavenging activity.

Tannin Content

Tannin content of the sample was determined by Vanillin Hydrochloride Method [16]. The absorbance was read in spectrophotometer at 500 nm.

Sensory Evaluation

Sensory evaluation was conducted as per the method described by Watts *et al*., (1989) [17]. Quality attributes of developed herbal tea combination were evaluated by twenty five semi trained panel members. The organoleptic evaluation sessions were conducted one hour before lunch under adequate conditions of temperature, humidity and illumination. The panelists were asked to score herbal teas on a scale of 9 to 1 hedonic scale.

Statistical Analysis

The assays were carried out in triplicate, and the results were expressed as mean values and the standard deviation (SD). Factorial Completely Randomized Design (FCRD) was applied for the analysis (Gomez and Gomez, 1984) [18].

Results and Discussion

Total Phenol Content (TPC)

The total phenolic content of fresh guava leaf was 99.25mg GAE/g. Steam blanched and hot water blanched herbal tea combination shows 113.5 and 109.5 (mg GAE/g) respectively. Chan *et al.,* (2010) [19] revealed that total phenol content in *C. sinensis* ranged from 60.60 to 141.20 mg GAE/g.

The results indicated that the total phenol content in steam blanched guava leaves tea combination is significantly higher. This may be due to the heat treatment which leads to degradation of tannins to simple phenolics compounds. Total phenol content is responsible to the antioxidant activity.

Total Flavonoid Content

The total flavonoid content of fresh guava leaf was 13.292mg QE/g. Steam blanched and hot water blanched herbal tea showed 5.522 and 4.604 (mg QE/g) respectively. Barreira *et al*., (2013) [20] revealed that total flavonoid content in *C. sinensis* tea 0.13mg CE/ml.

The results indicated that the total flavonoid content in steam blanched guava leaves tea combination is significantly higher. The steaming process of fresh guava leaves during the manufacturing of green tea will inactivate polyphenol oxidase enzyme, control the level of flavonoids compound of flavanol group or known by catechins and prevented from enzymatic

oxidation process to catechins to maintain polyphenols in monomer form [22].

Total Antioxidant Activity

The total antioxidant activity of fresh guava leaf was 71.16% RSA. Steam blanched and hot water blanched herbal tea combination shows 89.39 and 91.28 (% RSA) respectively. Chan *et al.*, (2012) [21] concluded that drying enhances the antioxidant property in leaves of white mulberry (*M. alba)* and laurel clockvine *(T. laurifolia).*

The results coincide with the present study where steam blanched guava leaf herbal tea combination showed highest antioxidant activity than the unblanched guava leaf herbal tea. It might be due to processing condition which influences the antioxidant activity of herbal tea combinations.

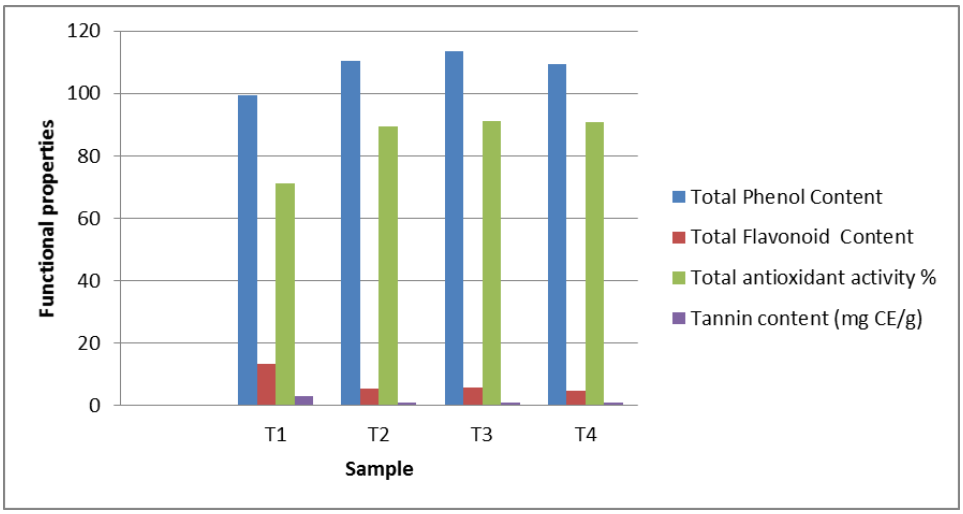
Tannin Content

The tannin content of fresh guava leaf was 2.962mg CE/g. Steam blanched and hot water blanched herbal tea combination showed 1.134 and 1.088 (mg CE/g) respectively. Mailoa *et al.*, (2014) [22] concluded that the ethanolic extract (30%) of guava leaves contains 2.351 mg/g of tannins. During processing tannin content of guava leaves decreased, which might be due to heat process leads to degradation of tannin content. However highest tannin content was found in steam blanched guava leaf tea combination.

Table 1: Functional properties of fresh guava leaves and tea samples

Sample	Total Phenol Content (mg GAE/g)	Total Flavonoid Content (mg QE/g)	Total antioxidant activity% (DPPH assay)	Tannin content (mg CE/g)
T1	99.25±2.514796	13.29±0.062659	71.16±1.299874	2.96±0.029671
T2	110.50±1.0526	5.52±0.116475	89.39±0.729866	1.13±0.032407
T3	113.50±3.166306	5.73±0.159822	91.28±1.117947	1.15±0.015581
T4	109.50±3.352739	4.60±0.031326	90.88±2.535276	1.09±0.011104

*Values are expressed as mean ± standard deviation.
T1- Fresh guava leaves
T2- Herbal teas without blanching
T3- Steam blanched herbal teas
T4-Hot water blanched herbal teas



T1- Fresh guava leaves
T2- Herbal teas without blanching
T3- Steam blanched herbal teas
T4-Hot water blanched herbal teas

Fig 1: Functional properties of fresh guava leaves and tea samples

Sensory Evaluation

The organoleptic evaluation of developed herbal tea combination was judged by a panel of semi trained members. Steam blanched guava leaf herbal tea (S2) had highest overall acceptability with the mean value of 9.1994±0.17 when compared to other treatments.

Table 2: Sensory evaluation of herbal tea

Sample	Sensory parameters				
	Appearance & Colour	Flavour	Consistency	Taste	Overall acceptability
S1	9.4	8.5	8.3	8.4	8.6494±0.1482
S2	9.3	9.7	8.3	9.5	9.1994±0.1706
S3	7.6	7.3	8.3	8.6	7.9494±0.0984

S1 Herbal tea without blanching
S2- Steam blanched herbal tea
S3-Hot water blanched herbal tea

Conclusion

This study concluded that steam blanching treatment of guava leaves could increase functional properties in terms of total polyphenol, total flavonoid, tannin and total antioxidant activity as well as sensory appeal of herbal tea. Based on these findings, it is recommended to use guava leaves for herbal tea preparation since it possessed high antioxidant activity which is beneficial for human health. Nowadays there are so many chronic degenerative diseases outbreak in industrialized and developing countries. The high cost of allopathic medicines direct to the search for herbal remedies to treat many ailments. Guava leaves are rich in bioactive compounds which can be used in the treatment of diabetes, cardiovascular disease, obesity and atherosclerosis. The guava leaf consumption is nil among common public even though it is rich in bioactive compounds. In order to exploit the bioactive compounds rich guava leaves, it can be used to prepare beverage. Hence it is suggested that guava leaf based herbal teas can be produced which will have high acceptability among all age groups of people.

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