ELSEVIER

Chemical composition and antioxidant activity of *Strobilanthes crispus* leaf extract

Maznah Ismail,* Elizabeth Manickam,* Azlina Md. Danial,* Asmah Rahmat,* and †Asmah Yahaya

**Department of Nutrition & Health Science, Faculty of Medicine & Health Sciences, University Putra Malaysia, Serdang, Selangor, Malaysia and †Department of Chemistry, Faculty of Science & Environmental Studies, University Putra Malaysia, Serdang, Selangor, Malaysia*

This study investigated the components present in and the total antioxidant activity of leaves of Strobilanthes crispus *(L.) Bremek or* Saricocalyx crispus *(L.) Bremek (Acanthacea). Proximate analyses and total antioxidant activity using ferric thiocyanate and thiobarbituric acid methods were employed. Minerals content was determined using the atomic absorption spectrophotometer, whereas the water-soluble vitamins were determined by means of the UV-VIS spectrophotometer (vitamin C) and fluorimeter (vitamins B_1 and B_2). Catechin, tannin, caffeine, and alkaloid contents were also studied. All data were compared to the previously reported results of Yerbamate, green tea, black tea, and Indian tea. The dried leaves contained a high amount of total ash (21.6%) as a result of a high amount of minerals including potassium (51%), calcium (24%), sodium (13%), iron (1%), and phosphorus (1%). High content of water-soluble vitamins (C, B_1, and B_2) contributed to the high antioxidant activity of the leaves. The leaves also contained a moderate amount of other proximate composition as well as other compounds such as catechins, alkaloids, caffeine, and tannin, contributing further to the total antioxidant activity. Catechins of* Strobilanthes crispus *leaves showed highest antioxidant activity when compared to Yerbamate and vitamin E. Consumption of the leafy extract daily (5 g/day) as an herbal tea could contribute to the additional nutrients and antioxidants needed in the body to enhance the defense system, especially toward the incidence of degenerative diseases.* (J. Nutr. Biochem. 11:536–542, 2000) *© Elsevier Science Inc. 2000. All rights reserved.*

Keywords: *Strobilanthes crispus*; nutritional composition; antioxidants; catechin; total antioxidant activity

Introduction

Over the centuries, no fewer than 3,000 plant species have been used to treat cancer.[1] Many recent studies indicate that among most plants, tea extracts have the most potent anticarcinogenic agents, thereby raising much interest not only in Japan but also in other countries.[2] Tea has become a product of great value and its popularity as a nonintoxicating drink is well received world wide. It is a beverage consumed by more than half the population of the world because of its desirable taste and refreshing effect as well as its properties considered to be a remedy for many ills.[3] Subsequent to medicinal properties of tea, many new plants are introduced and studied to increase the discovery of natural products as cancer chemotherapeutic agents.[4] So far, knowledge about beneficial effects of certain plants is mainly transmitted by personal communication; therefore, it remains unknown to the general population. Many studies need to be done to promote the usage and application of these native plants.

The *Strobilanthes crispus* ZII 109 (L.) Bremek or *Saricocalyx crispus* ZII 109 (L.) Bremek (Acanthaceae) plant is native to countries from Madagascar to Indonesia, and is commonly known as “daun picah beling” in Jakarta or “enyoh kelo,” “kecibeling,” or “kejibeling” in Java.[5] It was first recorded by Thomas Anderson (1832–1870) who classified the plant under Spermatophyta (Flowering Plants and Gymnosperma).[6] *Strobilanthes* (cone-head) was named

Address correspondence to Dr. Maznah Ismail, Department of Nutrition & Health Science, Faculty of Medicine & Health Sciences, University Putra Malaysia, 43400UPM, Serdang, Selangor, Malaysia.
Received January 11, 2000; accepted July 24, 2000.

J. Nutr. Biochem. 11:536–542, 2000

655 Avenue of the Americas, New York, NY 10010
0955-2863/00/$–see front matter
PII S0955-2863(00)00108-X

from the combination of *strobilos*, which means flower,[7] and *crispus*, which is phyllostachyus or spike-like leaf (*phyllo* means leaf, and *stachyus* means spike).[8] The conjunction of the names leads to the meaningful definition of the physical plant. This bush-like plant can be found on riverbanks or abandoned fields; some Javanese use this plant as fence hedges. The leaves are oblong-lanceolate, rather obtuse, and shallowly crenate-crispate.[9] Top surfaces of the leaves are darker green in color and less rough compared to the below surface.[5] The leaves are very scabrid on both surfaces and covered with short hairs, whereas the flowers are short, dense, and are panicled spikes.[10] The plant can be easily propagated by using the stacks.[5]

Although there is very little record of this plant being used for medicinal purposes, a study in Indonesia found that an infusion of the dried leaves of *Strobilanthes crispus* has been used as antidiabetic, diuretic, antilytic, and laxative. This plant has many cystoliths of calcium carbonate, and an infusion is mildly alkaline,[11] which gives a slightly bitter taste.[5] A very recent study also indicated that the water extract of *Strobilanthes crispus* contained compounds with very high binding affinity to protein molecules, which may bind the active sites of reverse transcriptase, therefore inhibiting the proliferation of retroviruses—agents in viral diseases such as acquired immune deficiency syndrome and adult T-cell leukemia.[12]

Soediro and colleagues[13] isolated and identified an ester glycosidic compound of caffeic acid, a verbascoside in the leaves, by using thin layer chromatography (TLC) techniques. This compound is known to have analgesic effects internally, and antifungal and antibacterial effects when used externally. Later, seven phenolic acids—ρ-hydroxy benzoic, ρ-voumaric, caffeic, vanillic, gentinic, ferulic, and syryngic acid—were also identified by TLC, paper chromatography, and UV spectrophotometric techniques.[14] However, there has not been any study performed on the composition of other chemicals in this plant, nor has the chemical composition been related to the total antioxidant activity. This study was therefore carried out to evaluate the chemical composition of *Strobilanthes crispus*; proximate analyses, vitamin and mineral content, and other compounds that could contribute to its total antioxidant activity were determined.

Materials and methods

Materials

The leaves of *Strobilanthes crispus* were collected from the Horticulture Unit of University Putra Malaysia. The leaves were separated from the stalks, thoroughly washed with tap water, rinsed with distilled water, and were then dried in the oven at 60°C overnight. The dired leaves were ground to a fine powder and stored in an airtight container until further use. Fresh leaves that had been cleaned were used only to determine moisture and ascorbic acid content. For antioxidant activity, ethyl extract of *Strobilanthes crispus* leaves was prepared in order to extract most of the catechin present. Crushed dried *Strobilanthes crispus* leaves (25 g) were placed in a separating funnel, soaked in distilled water and shaken for 10 min. Chloroform was added and the funnel was further shaken until two layers were formed. The organic layer was removed and ethyl acetate (J. T. Baker, Phillipsburg, PA, USA) was then added with constant shaking. The upper ethyl acetate layer was collected and concentrated using a rotary evaporator (Heidolph, WB 2001 Tokyo Rikakikai Co., Tokyo, Japan) until a viscous layer was formed.

Yerbamate, a popularly consumed imported commercial herbal tea was purchased from a local supermarket and was treated in a similar manner according to the analyses. Vitamin E was purchased from Sigma Chemical Co. (St. Louis, MO USA).

Methods

Proximate analysis. Moisture, total ash, crude fiber, protein, and ether extract contents of the sample were determined by Association of Official Analytical Chemists[15] and Tee et al.[16] methods. The nitrogen content was estimated by micro-kjeldhal techniques and the crude protein was calculated as $N \times 6.25$. Ash samples underwent further analyses to determine water-insoluble ash, water-soluble ash, and alkalinity of soluble ash according to the International Standard ISO 1576–1975 (E)[17] and 1578–1975 (E)[18] method, whereas acid-insoluble ash was determined according to Pearson[19] procedures. Extractives of the leaves were also determined according to the Pearson[19] method. These analyses were carried out to investigate whether the leaves were suitable for herbal tea preparations. Total carbohydrate in the leaves was determined by the Association of Official Analytical Chemists[15] method. The change of the green color was determined in a spectrophotometer at 630 nm.

Mineral determination. Sodium, potassium, calcium, and iron content of the leaves was determined from ashed samples using the flame system of the Atomic Absorption Spectrophotometer (GBC, Model #908AA, USA). Phosphorus content in the triple acid digested sample was determined colorimetrically.

Water-soluble vitamins determination. Fresh leaf samples were needed to determine ascorbic acid (due to its sensitivity toward light and water), which was determined according to Association of Vitamin Chemists, Inc.[20] methods, whereas dry samples were used to determine riboflavin (B_2) and thiamin (B_1) using fluorimetry (Perkin Elmer, Model #LS-5, Victoria, Australia) methods. A sample containing riboflavin was prepared according to the Association of Vitamin Chemists, Inc.,[20] whereas samples containing thiamin were prepared according to the Association of Analytical Chemists.[15]

Determination of other chemical compounds. Other chemical components of the leaves including tannin, caffeine, alkaloid, and catechin were also determined. Caffeine was analyzed by Nollet[21] methods using reverse phase high pressured liquid chromatography (Hewlett Packard, Model HP #1100, Palo Alto, CA USA), whereas tannin was determined by titration of $KmnO_4$ (Hart and Fisher[22]) and percent tannin was calculated according to the formula, 1 mL $KmnO_4$ = 0.0416 g tannin (gallotannic acid) or 1 mL of 0.008M $KmnO_4$ = 0.001664 g tannin. Alkaloid content of the dried leaves was determined using Wagner et al.[23] methods with slight modification. In brief, *Strobilanthes crispus* leaves (1 g) were mixed thoroughly with 1 mL of 10% ammonia solution, or 10% Na_2CO_3 solution, then extracted by shaking for about 5 min with 5 mL methanol at 60°C in a shaking water bath. The filtrate was cooled and concentrated so that 100 μL (the maximal quantity that should be applied to the TLC plate) contained about 50–100 μg alkaloids. Catechin was extracted as mentioned earlier and was subjected to antioxidant activity analysis.

Determination of total antioxidant activity. Ferric thiocyanate (FTC) method. The methods of Mitsuda et al.[24] and Osawa and Namiki[25] were slightly modified by Kikuzaki and Nakatani.[26] FTC

Table 1 Chemical composition of *Strobilanthes crispus*, Yerbamate, green, black, and Indian teas

Analysis	Mean ± SD	Yerbamate[a]	Indian tea[a]	Green tea[b]	Black tea[b]
Proximate analysis					
Moisture content (%)	69.3 ± 0.1	9.00	9.3	—[c]	3.9–9.5
Total ash (%)	21.6 ± 0.1	6.7	—	6.1–9.2	4.9–6.5
Water-insoluble ash (%)	13.8 ± 1.0	—	—	5.2–7.2	—
Water-soluble ash (%)	7.9 ± 1.0	2.3	—	2.6–1.6	3.0–4.2
Alkalinity of soluble ash (mL acid/g)	6.5 ± 2.0	—	—	2.6–1.6	1.2–1.6
Acid-insoluble ash (%)	2.2 ± 0.8	—	—	0.05–0.9	0.2–0.4
Extractivities (%)	6.7 ± 0.4	33.1	—	33–45	30–50
Protein content	13.3 ± 0.9	9.8	14.1	—	5.0–6.2
Total carbohydrate (%)	4.3 ± 0.7	—	0.0	—	—
Crude fiber (%)	13.9 ± 0.6	15.5	—	9–15	—
Ether extract (%)	1.9 ± 0.7	2.1	—	—	14–18
Minerals (mg/100 g dried sample)					
Sodium	2,953 ± 60	1,112	44.5	—	—
Potassium	10,900 ± 498	2,227	2,160	—	—
Calcium	5,185 ± 359	664	426	—	—
Iron	255 ± 163	—	15.2	—	—
Phosphorus	201 ± 22	394	628	—	—
Vitamins (mg/100 g sample)					
Ascorbic acid (C)*	9.8 ± 1.2	—	0.0	—	—
Riboflavin (B_2)	0.11 ± 0.04	—	0.9	—	—
Thiamin (B_1)	0.14 ± 0.001	0.2	0.14	—	—
Other chemical components					
Caffeine (%)	0.0 (11)	0.6–1.4	2.5	1.5–4.3	10–11
Tannin (%)	1.0 ± 0.30	9.8	—	—	—
Alkaloid (%)	3.2 ± 0.60	4.4	—	—	—
Catechin (%)	1.18 ± 0.08	0.9	—	5.8	—

Each value represents the mean ± SD of 6 determinations.
[a]Reference: Hart and Fisher[22]
[b]Reference: Pearson[19]
[c]—shows no studies have been done
*Value refers to values on a wet weight basis.

method was used to determine the amount of peroxide at the initial stage of lipid peroxidation. The peroxide reacts with ferrous chloride to form a reddish ferric chloride pigment. In this method, the concentration of peroxide decreases as the antioxidant activity increases. In brief, a mixture of 4 mg weight sample in 4 mL absolute ethanol (Merck) 4.1 mL of 2.52% linoleic acid (Sigma) in absolute ethanol, 8 mL of 0.05 M phosphate buffer (pH 7.0), and 3.9 mL of water was placed in a vial ($\varnothing = 38$ mm, $h = 75$ mm) with a screw cap and then placed in an oven at 40°C in the dark. To 0.1 mL of this solution was added 9.7 mL of 75% ethanol and 0.1 mL 30% ammonium thiocyanate (Sigma). Precisely 3 min after the addition of 0.1 mL of 0.02M ferrous chloride in 3.5% hydrochloric acid to the reaction mixture, the absorbance was measured at 500 nm for every 24 hours until the absorbance of the control reached maximum. The control and standard were subjected to the same procedures as the sample except that for the control, only the solvent was added, and for the standard, 4 mg sample was replaced with 4 mg of vitamin E.

Thiobarbituric acid (TBA) method. The method of Ottolenghi[27] was used to determine the TBA values of the samples. The formation of malonaldehyde is the basis for the well-known TBA method used for evaluating the extent of lipid peroxidation. At low pH and high temperature (100°C), malonaldehyde binds TBA to form a red complex that can be measured at 532 nm. The increase of the amount of the red pigment formed correlates with the oxidative rancidity of the lipid. Two mL 20% trichloroacetic acid and 2 mL TBA aqueous solution were added to 1 mL of sample solution prepared as in the FTC procedure, and incubated in a similar manner. The mixture was placed in a boiling water bath for 10 min. After cooling, it was centrifuged at 3,000 rpm for 20 min and the absorbance of the supernatant was measured at 532 nm. Antioxidant activity was based on the absorbance on the final day.

Data analysis. All determinations were carried out in six triplicates and data were subjected to analysis of variance and Student's *t*-test.

Results

Chemical composition

The proximate composition, some water-soluble vitamins and mineral contents as well as other chemical components of the *Strobilanthes crispus* leaves are presented in *Table 1*. For comparison purposes, values of Yerbamate, Indian tea, green tea, and black tea, according to some previous studies, are also included. Moisture content formed the bulk of tissue weight in the fresh *Strobilanthes crispus* leaves with the mean value of 69.3%. High levels of total ash (21.6%) in the dried plant leaves, on the other hand, were the result of high content of minerals such as potassium (10,900 mg/100 g sample), followed by calcium (5,185 mg/100 g sample). This plant was also rich in sodium (2,953 mg/100 g sample) and iron (255 mg/100 g sample) but with lower amounts of phosphorus (201 mg/100 g sample). Yerbamate, however, contained only 2,227 mg/100 g potassium, 1,112 mg/100 g sodium, 664 mg/100 g calcium, and 394 mg/100

Table 2 Percent contribution of minerals and vitamins by two cups of *Strobilanthes crispus* leaves

Components	mg components in 100 g	mg components in 5 g	Minimum RDA* needed	% of RDA components in 2 cups (5 g)***
Minerals				
Sodium	2,953	148	1,100	13.4
Potassium	10,900	545	1,875	29.1
Calcium	5,185	259	800	32.4
Iron	255	12.8	10	127.5
Phosphorus	201	10.1	800	1.3
Vitamins				
Ascorbic acid (C)***	9.75	0.49	10	4.9
Riboflavin (B_2)	0.144	0.006	0.6	1.0
Thiamin (B_1)	0.138	0.007	0.5	1.4

*Recommended Daily Allowance (RDA) according to Mahan and Escott-Stump.[36]
**Actual absorption needs to be taken into consideration.
***Values per wet leaves basis; all others are on dry leaves basis.

g phosphorus; Indian tea contained 2,160 mg/100 g potassium, 628 mg/100 g phosphorus, 426 mg/100 g calcium, 44.5 mg/100 g sodium, and 15.2 mg/100 g iron. The potassium content in *Strobilanthes crispus* leaves was about 5 times higher than in Yerbamate or Indian tea, and calcium was present about 10 times higher in *Strobilanthes crispus* when compared to the other two commercial products. However, phosphorus content was about 2 to 3 times lower than that of Yerbamate and Indian tea. The leaves also contained 2.2% of acid-insoluble ash, which was less than the U.S. regulations (4%) but much higher than in the green tea (0.05–0.9%). Water-insoluble ash and water-soluble ash were determined as 13.8% and 7.9%, respectively. Green tea and black tea, on the other hand, contained water insoluble ash and water-soluble ash ranging between 5.2–7.2% and 3.0–4.2%, respectively. The *Strobilanthes crispus* leaves gave a value of alkalinity at 6.5 mL acid/g sample, whereas green tea and black tea contained only 1.2–1.6 mL acid/g. The extractive value of this plant was 6.7%, whereas the extractive values for green tea and black tea ranged from 30–50%.

Protein content of *Strobilanthes crispus* leaves was about 13.3%, which is quite similar to Indian tea (14.1%). Total carbohydrate and ether extract content was 4.3% and 1.9%, respectively. Previous data of ether extract of Yerbamate gave a value of 2.1%. *Strobilanthes crispus* leaves also contained high amount of crude fiber (13.9%), which was comparable to Yerbamate (15.5%) and also within the range reported for green tea (9–15%). The *Strobilanthes crispus* leaves contained high amounts of vitamins, especially ascorbic acid (9.8%), in the fresh leaves. Indian tea did not show any appreciable amount of ascorbic acid due to losses during processing. As for riboflavin and thiamin, the leaves contained 0.11% riboflavin and 0.14% thiamin in dried leaves. The value of thiamin was quite similar to Yerbamate (0.2%) and Indian tea (0.14%), whereas riboflavin in *Strobilanthes crispus* (0.11%) was very much less compared with Indian tea (0.9%).

Levels of other components of the *Strobilanthes crispus* leaves were much lower than those of green tea, Indian tea, or Yerbamate. *Strobilanthes crispus* leaves contained only 0.01% caffeine, 1.0% tannin, and 3.2% alkaloid, whereas Yerbamate contained 0.6–1.4% caffeine, 9.8% tannin, and 4.4% alkaloid. The amount of catechin, however, was higher in *Strobilanthes crispus* leaves (1.18%) than in Yerbamate (0.9%). Green tea, on the other hand, contained about 1.5–4.3% caffeine and 5.8% catechin.[19,20]

Table 2 showed the percent composition of minerals and vitamins in 5 g of dried *Strobilanthes crispus* leaves. Consumption of two cups (5 g) of the leaves as herbal tea (2.5 g per cup) daily could contribute appreciable amounts of minerals to the body. The table shows about 127.5% of the Recommended Daily Allowance (RDA) for iron being contributed from 5 g of sample, followed by calcium (32.4%), potassium (29.1%), and sodium (13.4%). Nevertheless, not all of the minerals (especially iron) present in the leaves can be absorbed. As much as 5% of vitamin C could be consumed daily if fresh *Strobilanthes crispus* leaves were used as tea brew. However, phosphorus, riboflavin, and thiamin were present only in minor amounts (1% each) in 5 g sample if consumed daily.

Antioxidant activity

The total antioxidant activity of *Strobilanthes crispus* and Yerbamate extracts was assessed by both FTC and TBA methods at a concentration of 0.02% and compared with vitamin E. The individual activity of these samples by the FTC method (*Figure 1*) showed low absorbance values, which indicated high levels of antioxidant activity, as shown in *Figure 2*. In general, *Strobilanthes crispus*, Yerbamate, and vitamin E markedly inhibited the oxidation of linoleic acid for a period of 6 days when compared to control. *Strobilanthes crispus* showed the least increase in absorbance values, followed by Yerbamate and vitamin E, but there was no significant difference among these samples. The control showed no significant difference from these samples until Day 2, but increased significantly on Day 3 ($P < 0.05$), reached maximum level on Day 4, and finally dropped on Day 5 due to the malonaldehyde content.

The absorbance values from TBA method (*Figure 3*) showed total peroxide values produced by the oxidation of linoleic acid. The higher the absorbance values, the lower the level of antioxidant activity. The control had the highest

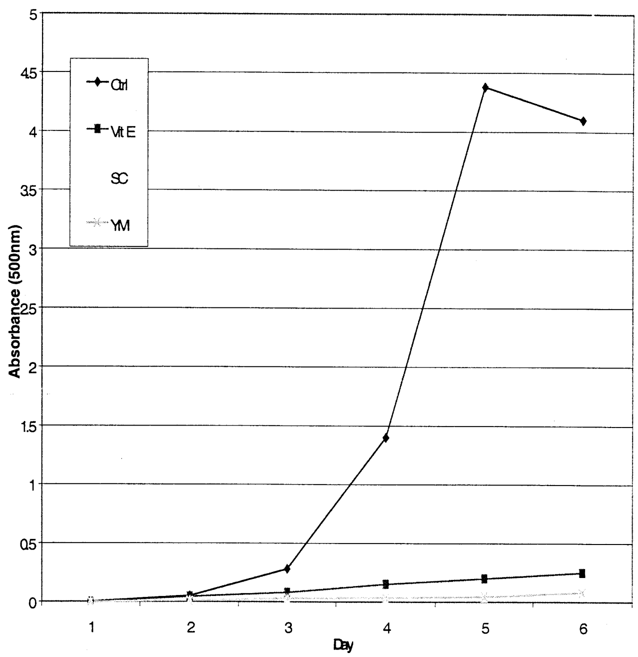

Figure 1 Absorbance values of samples at 0.02% concentration using FTC method. Ctrl: control; Vit E: vitamin E; SC: *Strobilanthes crispus*; YM: Yerbamate.

absorbance value (1.47) followed by vitamin E (0.23), Yerbamate (0.22), and *Strobilanthes crispus* (0.18), and they were significantly different within the samples. Based on the present result, *Strobilanthes crispus* had the highest antioxidant activity (96%), followed by Yerbamate (82%) and vitamin E (76%).

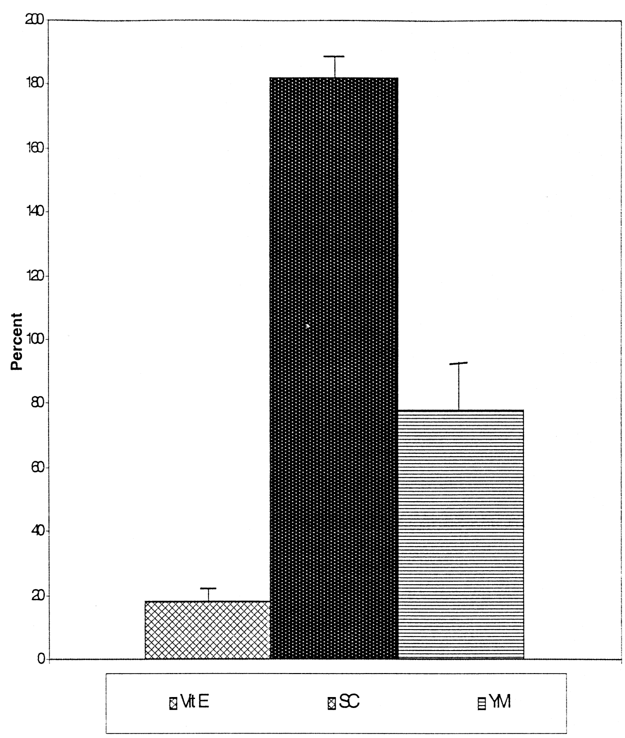

Figure 2 Antioxidant activity of samples at 0.02% concentration using FTC method. Vit E: vitamin E; SC: *Strobilanthes crispus*; YM: Yerbamate.

Discussion

Many plants are consumed not only as vegetables or used in food preparations but they are also utilized for medicinal purposes. *Strobilanthes crispus* is used widely in the herbal and traditional medicinal preparations in this part of the world, especially among the Indonesians and the Malays. It is also commonly consumed in the form of herbal tea. Although it has been used widely in the treatment of various ailments, scientific data on this plant is still lacking. Many studies need to be done before it can be promoted for utilization or be commercialized. The World Health Organization estimated that ~80% of the world's inhabitants rely on traditional medicine for their primary health care needs, and most of this therapy involves the use of plant extracts or their active components. The present study elaborates on the chemical composition of *Strobilanthes crispus* as well as on the total antioxidant activity of the leafy ethyl acetate extract.

The chemical composition was carried out to determine the nutrients and some of the active components in the leaves of the plant and, hence, compare them with some other commercial teas and an imported herbal tea, Yerbamate, which is highly consumed locally. Green tea is now one of the most widely consumed beverages in Asian countries not only because of its desirable taste but also due to its antioxidative, antimicrobial, and anticarcinogenic properties.[25] Similarly, black tea and Indian tea are consumed widely, although their effects toward disease prevention are very much less compared to green tea. Apart from these teas, there is now an increased interest to consume various herbal teas for similar purposes. All these teas contain numerous compounds including flavanoids and catechin, both of which have antioxidant and health-promoting properties. Yerbamate contains chlorophyll, iron, panthotenic acid, trace minerals, and vitamins C and E.[28] From the results, *Strobilanthes crispus* has been shown to have similar high mineral content, especially potassium, calcium, sodium, and iron. An earlier report by Sunarto[5] indicated that high amounts of the potassium is present as potassium silicate. High ash levels, as found in this study, could suggest the presence of an adulterant; however, acid-insoluble ash content was found to be lower than the maximum permitted value for herbs and spices.[29] Furthermore, the high values of both water-soluble ash and alkalinity of the ash indicated a high quality of *Strobilanthes* leaves. In addition to minerals, the leaves also contributed other nutrients such as fiber, ascorbic acid, riboflavin and thiamin, which correlate with the possible antioxidant activity. Proximate analysis composition is of a moderate amount. *Stribilanthes* leaves were found to be low in tannin and caffeine but contained an appreciable amount of catechin, which was lower than green tea but slightly higher than Yerbamate.

Daily consumption of the leafy water extract could contribute to the daily requirement of some of the nutrients. Normally, each cup of tea is recommended to have about 2.5 g of tea leaves. Consumption of about 5 g dried

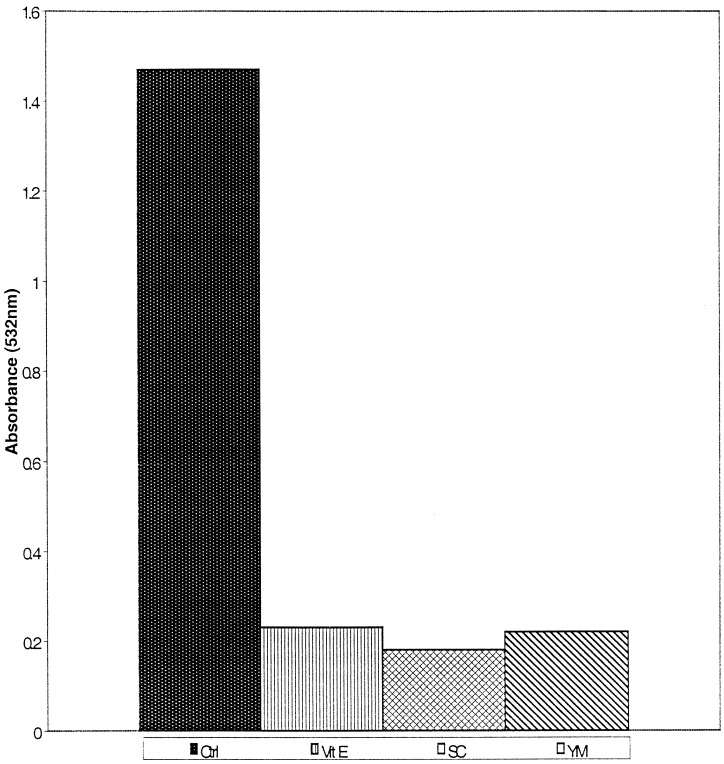

Figure 3 Absorbance values of samples at 0.02% concentration using TBA method. Ctrl: control; Vit E: vitamin E; SC: *Strobilanthes crispus*; YM: Yerbamate.

Strobilanthes crispus leaves or a minimum of two cups daily may contribute a high percentage of adult RDAs for iron, calcium, and potassium. Inorganic iron or nonheme iron is the most abundant form of iron in plant sources and the most prevalent form of iron in the diet; however, only about 3% of it can be absorbed. Nonheme iron absorption may be enhanced by ascorbic acid, certain animal proteins, and gastric acidity, but on the other hand, may be reduced by many natural substances including tannic acid, calcium, phytates, and oxalates. Low tannin and caffeine levels found in the leaves but high ascorbic acid content may be able to enhance the iron absorption. Catechin and alkaloid have been used in the treatment of many diseases for over 10 years.[30]

Catechin, which is present mostly in fruits, vegetables, tea, coffee, and cereal grains, was used to estimate the antioxidant activity of this plant. Antioxidants are known to alleviate oxidative stress by scavenging free radicals and protect biological macromolecules from their toxic effect. Because oxidative stress is generally perceived as one of the major causes for the accumulation of mutations in the genome, antioxidants are believed to provide protection against cancer.[30] During the oxidation process, peroxide is gradually decomposed to lower molecular compounds that are measured by FTC and TBA methods. FTC method is used to measure the amount of peroxide at the primary stage of linoleic acid peroxidation, whereas TBA measures at the secondary stage.[26] From the FTC result, *Strobilanthes crispus* showed the least increase in absorbance values, followed by Yerbamate and vitamin E without differences ($P < 0.05$) among them from Day 1 to Day 2, but levels increased significantly on Day 3, reached maximum levels on Day 4, and finally dropped on Day 5. This reduction is due to the accumulation of malonaldehyde compounds from linoleic acid oxidation, which is not stable. Further oxidation causes malonaldehyde to be converted to secondary products such as alcohols and acids that cannot be detected. Antioxidant activity in *Strobilanthes crispus* ethyl extract was found to be the highest, followed by Yerbamate and vitamin E. The high antioxidant activity of *Strobilanthes crispus* ethyl extract could be due to the catechin as well other flavanoids present in the extract, although their effect might not be as much as catechin due to their low levels. This result is supported by the report of Lunder,[31] who found catechins, especially epigallocatechin gallate, which represents 50–60% of all catechins, seem to be responsible for the total antioxidant activity; there seems to be a good correlation between its content and the antioxidant activity. Apart from these compounds, other phenolic acids that have been identified in the leaves[31] may also contribute to the antioxidant activity. The content of catechin somewhat varies in various teas in the following order; green tea > oolong tea > black tea. Serafini and co-workers[32] studied the in vitro antioxidant activity of green and black teas and established that both of them represented an excellent source of antioxidants, especially catechin, with green tea being about five times more potent than black tea. However, in another study, it was reported that oolong tea, which exhibited 73.6% inhibition of linoleic acid peroxidation, showed greater antioxidant activity than green tea, suggesting that the variable in antioxidant activity of these tea extracts may not be completely attributed to the content of catechins alone.[33] Other polyphenolic compounds such as those that have been isolated from the leafy extract as well as tocopherol, derivatives of cinnamic acid, phosphatide, and other organic acids could contribute to the total antioxidant activity.[34] However, further investigations are required to determine the specific components that may exhibit different degrees of activity.

It must be recognized that meaningful comparison of the data presented with those previously reported for other plants may be difficult because of different procedures used in the analyses, and that agronomic practices and environmental conditions can also influence the cellular contents of the plants.[35] Nonetheless, some of the values obtained for the different parameters for *Strobilanthes crispus* appear to fall within the range reported for other tea leaves and, in some cases, they are higher. Coupled with favorable amounts of minerals, fiber, vitamins, and the various phenolic compounds, this plant may present a potential use as an herbal tea worth promoting.

Acknowledgments

This study was supported by Intensifying Research Priority Areas (IRPA) Grant No. 06.02.04.0016 from the Ministry of Science, Technology, and the Environment, Malaysia.

References

1 Lewis, W.H. and Elvin-Lewis, M.P.F. (1977). *Medical Botany: Plants Affecting Man's Health*. Interscience Publications, New York, NY, USA

2 Reddy, B.S. and Rao, C.V. (1994). Chemoprevention of colon cancer by thiol and other organosulfur compounds. In *Food Phytochemicals for Cancer Prevention I: Fruits and Vegetables, ACS Symposium Series 546* (M.T. Huang, T. Osawa, C.T. Ho, and R.T. Rosen, eds.), pp. 164–172, American Chemical Society, Washington, DC, USA

3 Wickremasinghe, R.L. (1993). Biological activity of tea. *The Planter* **69(805),** 176–179

4 Suh, N., Lumonadia, L., Fong, H.H.S., Douglas, A.K., and John, M.P. (1995). Discovery of natural product chemopreventative agents utilizing HL-60 cell differentiation as a model. *Anticancer Res.* **15,** 233–240

5 Sunarto, P.A. (1977). *Materia Medika Indonesia*. Penerbit Ditektorat Jenderal Pengawasan Obat dan Makanan, Jakarta, Indonesia

6 Brummit, R.K. and Powerll, C.E. (1992). *Authors of Plant Names*. Royal Botanic Gardens, Kew, England

7 Plowden, C.C. (1968). *A Manual of Plant Names.* George Allen and Unwin Ltd., London, England

8 Jackson, B.D. (1960). *Index Kewensis: An Enumeration of the Genera and Species of Flowering Plants*, 3rd ed., Vol. II, Clarendon Press, London, England

9 Backer, C.A.D. and Bakhuizen, R.C.V. (1963). *Flora of Java* (Spermatophytes Only). Vol. II, Noordhoff Press, Groningen: 561–562

10 Heyne, K. (1987). Acanthaceae: Strobilanthes Crispus BL. *Tumbuhan Berguna Indonesia III*. Badan Litbang Kehutanan Jakarta ed., pp. 1752–1754, Yayasan Sarana Wana Jaya, Jakarta, Indonesia

11 Perry, L.M. and Metzger, J. (1980). *Medicinal Plants of East and South East Asia: Attributed Properties and Uses*. MIT Press, Cambridge, MA, USA and London, England

12 Kusumoto, J.T., Shimada, I., Kakiuchi, N., Hattori, M., and Namba, T. (1992). Inhibitory effects of Indonesian plant extracts on reverse transcriptase of an RNA tumour virus (I). *Phytother. Res.* **6,** 241–244

13 Soediro, L., Pellecuer, J., Andary, C., and Privat, G. (1983). *Strobilanthes crispus* (L.) Bl. I: Pemeriksaan senyawa turunan asam kafeat verbaskosid. *Acta Pharm. Indones.*, **VIII(I),** 1–10

14 Soediro, I., Pellecuer, J., Andary, C., and Privat, G. (1987). *Strobilanthes crispus* (L.) Bl.: Pemeriksaan Asam fenolat. *Acta Pharm. Indones.* **XII(I),** 1–7

15 Association of Official Analytical Chemists. (1984). *Official Methods of Analysis*, 14th ed., (S. Williams, ed.), Washington, DC, USA

16 Tee, E.S., Kuladevan, R., Young, S.I., Khor, S.G., and Omar, Z.H. (1996). *Laboratory Procedures in Nutrient Analysis of Foods*. Division of Human Nutrition, Institute for Medical Research, Kuala Lumpur, Malaysia

17 International Standard ISO 1576–1975 (E). Tea—Determination of water soluble ash and water insoluble ash. *International Organization for Standardization.* UDC 663.95, 543–548

18 International Standard ISO 1578–1975 (E). Tea—Determination of alkalinity of water soluble ash. *International Organization for Standardization.* UDC 663.95: 543.241

19 Pearson, D. (1976). *The Chemical Analysis of Foods*, 7th ed., The Macmillan Press Ltd., London, England

20 Association of Vitamin Chemists, Inc. (1966). *Methods of Vitamin Assay*, 3rd ed., Interscience Publishers, New York, NY, USA

21 Nollett, L.M.L. (1992). *Food Analysis by HPLC.* Marker Dekker Inc., London, England

22 Hart, L.F. and Fisher, H.J. (1971). *Modern Food Analysis*. W.B. Saundersw Company, London, England

23 Wagner, H., Bladt, S., and Zgainoki, E.M. (1984). *Plant Drug Analysis*. Berlin Heidelberg, Tokyo, Japan

24 Mitsuda, H., Yasumoto, K., and Iwani, K. (1967). Antioxidant action of indole compounds during the autoxiation of linoleic acid. *Eiyo to Shoduryou* **19(3),** 210

25 Osawa, T. and Namiki, M. (1981). A novel type of antioxidant isolated from leaf wax of Eucalyptus leaves. *Agric. Biol. Chem.* **45,** 735–740

26 Kikuzaki, H. and Nakatani, N. (1993). Antioxidant effects of some ginger constituents. *J. Food Sci.* **58(6),** 1407–1410

27 Ottolenghi, A. (1959). Interaction of ascorbic acid and mitochondria lipids. *Arch. Biochem. Biophys.* **79,** 355–359

28 Yen, G.C. and Chin, H.Y. (1995). Antioxidant activity of various tea extracts in relation to their antimutagenicity. *J. Agric. Food Chem.* **43(1),** 27–32

29 Bulch, J.F and Balch, P.A. (1997). *Presciption for Nutritional Healing*, 2nd ed., Avery Publishing Group, New York, NY, USA

30 Ames, B.N. (1983). Dietary carcinogens and anticarcinogens. *Science* **221,** 1256–1263

31 Lunder, T.L. (1992). Catechins of green tea: Antioxidant activity. In *Phenolic Compounds in Food and Their Effects on Health II: Antioxidants and Cancer Prevention, ACS Symposium Series 507.* (M.T. Huang, C.T. Ho, C.Y. Lee, eds.), pp. 114–120, American Chemical Society, Washington, DC, USA

32 Serafini, M., Ghiselli, A., and Luzzi, A.F. (1996). In vivo antioxidant effect of green tea and black tea in man. *Eur. J. Clin. Nutr.* **50(1),** 28–32

33 Yen, G.C. and Chen, H.Y. (1994). Comparison of antimutagenic effect of various tea extracts. *J. Food Prot.* **57,** 54–58

34 Pratt, D.E. (1992). Natural antioxidant from plant materials. In: Phenolic Compounds in Food and Their Effects on Health II: Antioxidants and Cancer Prevention, ACS Symposium Series 507 (M.T. Huang, C.T. Ho, C.Y. Lee, eds.), pp. 114–120, American Chemical Society, Washington, DC, USA

35 Yeoh, H.H. and Wong, P.F.M. (1993). Food value of lesser utilized tropical plants. *Food Chem.* **46,** 239–241

36 Mahan, L.K. and Escott-Stump S. (1996). *Krause's Food, Nutrition and Diet Therapy, 9th ed.* W.B. Saunders Co., Philadelphia, Pennsylvania, USA