Alpha-Tocopherol Content in 62 Edible Tropical Plants

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Vitamin E was determined by the high-performance liquid chromatography (HPLC) method. All the plants tested showed differences in their α-tocopherol content and the differences were significant ($p < 0.05$). The highest α-tocopherol content was in *Sauropus androgynus* leaves (426.8 mg/kg edible portion), followed by *Citrus hystrix* leaves (398.3 mg/kg), *Calamus scipronum* (193.8 mg/kg), starfruit leaves *Averrhoa belimbi* (168.3 mg/kg), red pepper *Capsicum annum* (155.4 mg/kg), local celery *Apium graveolens* (136.4 mg/kg), sweet potato shoots *Ipomoea batatas* (130.1 mg/kg), *Pandanus odorus* (131.5 mg/kg), *Oenanthe javanica* (146.8 mg/kg), black tea *Camelia chinensis* (183.3 mg/kg), papaya *Carica papaya* shoots (111.3 mg/kg), wolfberry leaves *Lycium chinense* (94.4 mg/kg), bird chili *Capsicum frutescens* leaves (95.4 mg/kg), drumstick *Moringa oleifera* leaves (90.0 mg/kg), green chili *Capsicum annum* (87 mg/kg), *Allium fistulosum* leaves (74.6 mg/kg), and bell pepper *Capsicum annum* (71.0 mg/kg). α-Tocopherol was not detected in *Brassica oleracea, Phaeomeria speciosa, Pachyrrhizus speciosa, Pleurotus sajor-caju,* and *Solanum melongena.*

Keywords: *Edible tropical plants;* α*-tocopherol; vitamin E*

INTRODUCTION

Vitamin E is protective against approximately 80 diseases, such as cancer, cardiovascular diseases (*1*), cell membrane and DNA damage by free radicals, oxidation of low-density lipoproteins, and disorders of the skin, eye, lungs, and other lipid-rich body constituents (*2*−*9*). α-Tocopherol, the most common form of vitamin E present in nature, is the most biologically active (*10*), and is preferentially retained in large quantities and transported to body components (*11*).

Daood et al. (*12*) studied the antioxidant vitamin content of spice red pepper during ripening, processing, and storage. Konings et al. (*13*) developed a HPLC method for determination of tocopherols and tocotrienols in margarine, infant foods, and vegetables. Mazhar et al. (*14*) isolated 0.94 mg/100 g of α-tocopherol from garlic, and Abushita et al. (*15*) investigated the antioxidant vitamin (vitamin E, vitamin C, and β-carotene) content in tomato cultivars. John (*16*) determined α-tocopherol in some southeast Asian foods by the thin-layer chromatography (TLC) method, but recovery experiments showed very heavy losses, over 50%, occurred during saponification, re-extraction, drying, and chromatography stages. The richest natural sources of α-tocopherol are oils from corn, cottonseed, soybean, safflower, and wheat germ (*17*); whereas fruits, vegetables, and whole grains have smaller amounts (*18*). The purpose of this study was to screen 62 types of edible tropical plants for α-tocopherol content.

MATERIALS AND METHODS

Raw Material. The following edible plants were all obtained from Sri Serdang evening wet market or picked fresh from the plant: *Sauropus androgynus, Citrus hystrix, Calamus scipronum, Averrhoa belimbi* leaves and fruits, red and green *Capsicum annum, Apium graveolens, Ipomoea batatas* shoot, *Pandanus odorus, Oenanthe javanica,* black tea *Camellia chinensis, Carica papaya* shoots, *Lycium chinense* leaves, *Capsicum frutescens, Moringa oleifera, Allium fistulosum,* bell pepper C*apsicum annum, Brassica alboglabra, Momordica charantia, Piper sarmentosum, Sesbania grandiflora, Garcinia atroviridis, Mentha arvensis, Psophocarpus tetragonolobus, Piper betel, Parkia speciosa, Gynandropsis gynandra, Anacardium occidentale,* cabbage *Brassica oleracea, Hydrocotyle asiatica, Musa sapientum* flower, *Manihot utilissima* shoots, *Diplazium esculentum* shoots, *Glycine max* sprout, *Vigna sinensis, Allium odorum* shoots, *Trichosanthes anguina, Phaseolus aureus* sprout, *Phaseolus vulgaris, Colocasia esculentum* stalk, *Daucus carota, Amaranthus spinosus, Pisum sativum, Ipomoea aquatica, Polygonum minus, Raphanus sativus, Cymbopogon citratus, Amaranthus gangeticus, Curcuma longa, Brassica chinensis, Psidium guajava, Luffah acutangula, Brassica oleracea, Hibiscus esculentus, Cucurbita maxima, Allium sativum,* cauliflower *Brassica oleracea, Pachyrrhizus erosus, Pleurotus sajor-caju,* and *Solanum melongena.*

Analytical-grade reagents (AR) were used for sample preparation and extraction, and 0.45-μm regenerated-cellulose-membrane filtered and degassed solvents (HPLC grade) were used for HPLC. Ethanol (AR, 96% v/v, Merck, Germany), *n*-hexane (Merck, Germany; AR, Fisher Scientific, Fair Lawn, NJ), butylated hydroxytoluene (BHT, Sigma, Germany), α-tocopherol standard (Sigma, Germany), ethyl acetate (AR, Fisher Scientific, United Kingdom), L(+)-ascorbic acid (Sigma, Germany), phenolphthalein (BDH Chemical, England made to 0.1% solution with ethanol), potassium hydroxide pellets (Merck, Germany), and methanol (BDH Chemical, England) were used throughout.

(a) Extraction Solvent: *n*-Hexane−Ethyl Acetate. *N*-hexane (analytical grade, 900 mL) was mixed with 100 mL of ethyl acetate and 20 mg BHT was dissolved in this solvent mixture.

(b) α-Tocopherol Standard Solution. α-Tocopherol (50 mg/100 mL *n*-hexane) stock solution was used to prepare standard 20, 40, 60, 80, 100, and 200 μg/mL working solution, which were prepared fresh each day.

Sample preparation, saponification and extraction were done according to a modified Konings et al. (*13*) method. Washed vegetables were cut, dried at 40 °C oven, and ground, and 5 g was suspended in 30 mL of water in a 500-mL conical flask. To it was added 21 g of KOH dissolved in 100 mL of

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ethanol, and 0.25 g of ascorbic acid per gram test portion was added to protect against oxidation; it was mixed and then flushed with nitrogen. Saponification was done at 80 °C for 40 min; the sample was immediately cooled to room temperature, and 300 mL of water was added to bring the ethanol/water ratio to 0.3. *n*-Hexane/ethyl acetate, 9:1 (3 × 100 mL), was added, and the mixture was extracted 3 times in a separatory funnel. Organic phases were combined and washed with 100-mL portions of water until free of alkali, as determined by where the reaction of washes to phenolphthalein was neutral (no visible pink color). Organic phases were then filtered through anhydrous sodium sulfate into a beaker, and the filtrate was evaporated to dryness under reduced pressure (40 °C). The residue was dissolved in 20 mL of *n*-hexane (HPLC grade), flushed with nitrogen, and stored in a freezer at −20 °C. This material is hereafter referred to as the test solution.

The α-tocopherol content was determined on a Shimadzu HPLC system using Waters Bondapak C18 reverse-phase column, 10 μm, 3.9 × 300 mm. The mobile phase, methanol/water 94:6, was used at a flow rate of 1.5 mL/min and the UV detector was set at 292 nm.

The experimental data were analyzed using the analysis of variance (ANOVA) software and the significant differences among means were determined by Duncan's multiple range test (DMRT) using the Statistical Analysis System (SAS, 1987) computing program (SAS, Cary, NC). All values were expressed as means ± standard deviation on at least triplicate samples.

RESULTS AND DISCUSSION

Edible Plants Contain A High Value of Alpha-Tocopherol Content. The α-tocopherol content of most plant extracts investigated in this study (Table 1) has not been reported in previous literature. The current Recommended Dietary Allowance (RDA) for vitamin E intake is 10 mg (15 IU) for adult males and 7.6 mg (11.4 IU) for adult females (*19*).

Sauropus androgynus contained the highest α-tocopherol content of 79.65 mg α-tocopherol per 100 g edible portion (Table 1). *Sauropus androgynus* contained 74 Kcal of energy, 7.6 g protein, 1.8 g fat, and 6.9 g carbohydrate per 100 g of edible portions (*20*). It is one of the most popular leafy vegetables eaten by the Malays; the leaves when cooked are firm. The leaves and young shoots can be eaten raw or steamed. It is also used as a coloring matter in food in Indonesia.

Wild lime leaves (*Citrus hystrix*) contained the second highest α-tocopherol content of 66.00 mg per 100 g (Table 1). *Citrus hystrix* is a small tree with very distinctive leaves. The fruit is wrinkled, pear shaped, and dark green turning to yellow on ripening. Both its leaves and fruits have a pleasant lemon smell. Wild lime fruit and leaves are mainly used as a flavoring in both savory and sweet food. Malays use it as a hair shampoo and as a medicine (*21*). The leaves are a potent antioxidant. Othman (*22*) reported that the crude antioxidant extracts from *Citrus hystrix* were added to the fish burger during processing and all treatment showed better oxidative stability than control. Jayamalar and Mohamed (*23*) found that *Citrus hystrix* leaves showed a strong antioxidant activity in both linoleic acid and TBA test at 1000 ppm.

Semambu leaves (*Calamus scipronum*) contained the third highest α-tocopherol content of 32.87 mg per 100 g (Table 1). The buds of these canes are eaten as food and have medical and antiseptic properties. They are commonly used for treatment of fever and aches (*21*).

Carambola (*Averrhoa belimbi*) contained 29.25 mg α-tocopherol per 100 g leaves and 1.70 mg α-tocopherol per 100 g fruit (Table 1). The syrup of the fruit is useful in relieving thirst, febrile excitement, and also in some slight cases of hemorrhage from the bowels, stomach, and internal hemorrhoids. The sour fruit is used as an acidulant in curries, and functions as a dietary article in piles and scurvy (*24*). Carambola leaves contain linalool, nerol, geraniol, and α-terpineol, which could act as potent antioxidants (*25*). It is used medicinally as a paste applied hot for itch.

Capsicum Species. The many varieties of chili include red chili, green chili, bell pepper (*Capsicum annum*), and bird chili (*Capsicum frutescens*). Chilies are rich in vitamins, especially vitamins A and C. Red chilies contained the highest amount of α-tocopherol, 29.07 mg per 100 g. Bird chili (*Capsicum frutescens*) contained 16.84 mg, green chili contained 15.39 mg, and bell pepper contained 13.81 mg α-tocopherol per 100 g (Table 1). Daood et al. (*12*) reported a higher content of 40.5 mg α-tocopherol per 100 g of red chili. *Capsicum frutescens* are used in native practice in typhus, intermittent fevers, dropsy, gout, dyspepsia, and cholera. Externally, they are used as a rubefacient, and internally, they are used for stomach ache (*24*). *Capsicum annum* acts as an acid stimulant, and externally as a rubefacient. It is used in sore throat, scarlatina, hoarseness, dyspepsia, yellow fever, and occasionally in diarrhea (*24*).

Local celery (*Apium graveolens*) contained 26.62 mg α-tocopherol per 100 g (Table 1). The plant is used as a flavoring because of its characteristic smell. The volatile oil is most abundant in the seeds and these often serve as a spice (*20*). Celery contains a glucoside (apin) and its leaves can be used for medicinal purposes or as food: eaten raw or cooked.

Sweet potato shoots (*Ipomoea batatas*) contained 25.03 mg α-tocopherol per 100 g shoots (Table 1). The young leaves are consumed as a green vegetable. They contain oxalate, hydrocyanic acid, and phytic acid (*26*).

Wolfberry (*Lycium chinense*) contained 17.73 mg α-tocopherol per 100 g leaves (Table 1). The fruits and leaves of *Lycium chinense*, family Solanaceae, are used as foods, tea, and medicine in the Orient. *Lycium chinense* leaves reportedly contain vitamin C and tocopherols and are capable of abating or reducing the risk of certain diseases such as arteriosclerosis, essential arterial hypertension, diabetes, and nightblindness (*27*).

Cruciferous vegetables (broccoli, cabbage, kale, and cauliflower) contain high levels of vitamins (α-carotene, β-carotene, α-tocopherol, γ-tocopherol, and ascorbate) that can act as antioxidants (*6*). The α-tocopherol contents of broccoli, cabbage, and kale were found to be 5.65, 1.34 , and 13.65 mg/100 g, respectively, but in cauliflower α-tocopherol was not detected (Table 1). Kurilich et al. (*6*) reported that kale had the highest levels of vitamins, followed by broccoli and then by cabbage and cauliflower with comparatively low concentrations. Konings et al. (*13*) report on the α-tocopherol content in broccoli (6.32 mg/100 g) was quite close to the result in this study (5.65 mg/100 g). However, the α-tocopherol content in fresh broccoli reported by Bauernfeind (*17*) was very low (0.5 mg/100 g).

French bean (*Phaseolus vulgaris*) has a relatively low α-tocopherol content (3.4 mg/100 g seeds, Table 1) quite similar to the results of Tsuda et al. (*28*).

Winged bean (*Psophocarpus tetragonolobus*) plant is consumed in its entirety because each part has nutritional and medicinal properties (*21*). The beans

Table 1. Alpha-Tocopherol Content[a] of 62 Types of Edible Tropical Plants

plant	α-tocopherol	
	dry weight basis	fresh weight basis
Leaves		
Sauropus androgynus	79.65 ± 0.23[A]	42.68 ± 0.12
Citrus hystrix	66.00 ± 3.61[B]	39.83 ± 2.18
Calamus scipronum	32.87 ± 0.15[C]	19.38 ± 0.09
Averrhoa belimbi	29.25 ± 0.48[D]	16.83 ± 0.27
Apium graveolens	26.62 ± 1.55[E]	13.64 ± 0.80
Pandanus odorus	24.65 ± 0.12[F]	13.15 ± 0.06
Oenanthe javanica	24.22 ± 0.85[F]	14.68 ± 0.51
black *Camellia chinensis*	21.08 ± 0.58[G]	18.33 ± 0.51
Lycium chinense leaves	17.73 ± 0.28[H]	9.44 ± 0.15
Moringa oleifera leaves	16.21 ± 0.83[IJ]	9.00 ± 0.46
Allium fistulosum leaves	14.46 ± 0.32[KL]	7.46 ± 0.16
Brassica alboglabra leaves (kale)	13.65 ± 0.24[L]	7.27 ± 0.13
Piper sarmentosum leaves	10.12 ± 0.36[N]	5.97 ± 0.21
Sesbania grandiflora leaves	9.51 ± 0.08[N]	5.53 ± 0.05
Mentha arvensis leaves	9.18 ± 0.07[N]	4.86 ± 0.04
Piper betel leaves	7.63 ± 0.28[O]	4.32 ± 0.16
Gynandropsis gynandra leaves	6.74 ± 0.81[PO]	3.61 ± 0.43
Brassica oleracea (broccoli)	5.65 ± 0.19[PQ]	2.95 ± 0.10
Hydrocotyle asiatica	5.60 ± 0.41[PQ]	2.98 ± 0.22
Allium odorum leaves	3.31 ± 0.01[R]	1.74 ± 0.004
Colocasia esculentum leaf stalk	3.06 ± 0.13[T]	1.59 ± 0.07
Amaranthus spinosus leaves	2.91 ± 0.25[T]	1.57 ± 0.14
Ipomoea aquatica leaves	2.66 ± 0.10[V]	1.37 ± 0.05
Polygonum minus leaves	2.60 ± 0.11[V]	1.41 ± 0.06
Cymbopogon citratus	2.31 ± 0.04[V]	1.26 ± 0.02
Amaranthus gangeticus leaves	2.26 ± 0.29[V]	1.16 ± 0.15
Brassica chinensis leaves	1.75 ± 0.13[W]	0.89 ± 0.06
Brassica oleracea cabbage	1.34 ± 0.01[YZ]	0.69 ± 0.01
Shoots		
Ipomoea batatas shoots	25.03 ± 0.09[F]	13.01 ± 0.05
Carica papaya shoots	20.78 ± 0.54[G]	11.13 ± 0.29
Anacardium occidentale shoots	5.73 ± 0.02[PQ]	3.17 ± 0.01
Manihot utilissima shoots	4.93 ± 0.02[Q]	2.72 ± 0.01
Diplazium esculentum shoots	4.78 ± 0.03[Q]	2.68 ± 0.01
Glycine max sprout	3.45 ± 0.19[R]	1.82 ± 0.10
Phaseolus aureus sprout	3.19 ± 0.26[SR]	1.64 ± 0.13
Fruit		
red *Capsicum annum*	29.07 ± 0.91[D]	15.54 ± 0.49
Capsicum frutescens	16.84 ± 0.28[H]	9.54 ± 0.31
green *Capsicum annum*	15.39 ± 0.04[KJ]	8.70 ± 0.02
bell pepper *Capsicum annum*	13.81 ± 0.01[L]	7.10 ± 0.005
Momordica charantia fruit	11.79 ± 1.15[M]	6.06 ± 0.59
Garcinia atroviridis	9.39 ± 0.42[N]	7.56 ± 0.34
Trichosanthes anguina snake gourd	3.25 ± 0.39[R]	1.67 ± 0.20
Averrhoa belimbi starfruit	1.70 ± 0.10[WX]	0.87 ± 0.05
Psidium guajava guava	1.66 ± 0.01[X]	0.88 ± 0.006
Luffah acutangula angular luffa	1.44 ± 0.16[XY]	0.74 ± 0.08
Hibiscus esculentus lady fingers	1.28 ± 0.07[YZ]	0.66 ± 0.04
Cucurbita maxima pumpkin	1.27 ± 0.07[YZ]	0.70 ± 0.004
Solanum melongena brinjal	ND	ND
Seeds/beans		
Psophocarpus tetragonolobus (winged beans)	8.91 ± 0.05[N]	4.62 ± 0.03
Parkia speciosa petai beans	7.13 ± 0.31[O]	4.15 ± 0.18
Vigna sinensis beans	3.33 ± 0.14[R]	1.74 ± 0.07
Phaseolus vulgaris (beans)	3.13 ± 0.15[ST]	1.68 ± 0.08
Pisum sativum french peas	2.83 ± 0.04[U]	1.47 ± 0.02
Flowers		
Musa sapientum flower	5.13 ± 0.02[Q]	2.98 ± 0.22
Brassica oleracea cauliflower, broccoli	ND	ND
Phaeomeria specios torch ginger	ND	ND
Roots		
Daucus carota carrot	2.99 ± 0.22[T]	1.56 ± 0.12
Raphanus sativus white radish	2.38 ± 0.11[V]	1.22 ± 0.05
Curcuma longa turmeric	1.78 ± 0.05[W]	1.10 ± 0.017
Allium sativum	1.23 ± 0.002[YZ]	0.77 ± 0.001
Pachyrrhizus erosus sengkuang	ND	ND
Fungi		
Pleurotus sajor-caju oyster mushroom	ND	ND

[a] Mean ± SD of duplicate analysis; expressed as mg per 100 g edible portion of sample. Means within column with the same letters are not significantly different ($\alpha = 0.05$). ND shows non-detected value or no peak found. The response of the HPLC system was linear over the range 0 to 200 μg/mL for α-tocopherol, with correlation coefficients $r^2 = 0.9972$. The detection limit for these compounds was 1.23 mg/100 g.

have been used as a milk substitute in the treatment of kwashiorkor (*29*) in children, and the seeds are rich in tocopherol (126 mg/100 g fresh weight) and some minerals (*30*). However, this study showed immature green winged bean in the pod to contain only a small amount of α tocopherol (8.91 mg/100 g fresh weight, Table 1).

Garlic (*Allium sativum*)'s α-tocopherol content (1.23 mg/100 g, Table 1) was higher than the amount reported by Mazhar et al. (*14*). Garlic is used in all parts of the world, not only as a spice and food but also as a popular folk remedy for ailments such as heart disease, cancer, diabetes, infections, parasites, respiratory ailments, and digestive problems, and to delay blood clotting.

Alpha-Tocopherol Content In Different Types of Edible Plants. John (*16*) reported the α-tocopherol content of various southeast Asian foodstuffs as determined by thin-layer chromatography. The α-tocopherol values reported per 100 g were: bean sprout (*Phaseolus aureus*), 0.20 mg; chinese cabbage (*Brassica pekiniensis*), 0.049 mg; kangkong (*Ipomoea aquatica*), 3.7 mg; spinach (*Spinacea oleracea*), 1.6 mg; bitter gourd (*Momordica charantia*), 1.4 mg; lady's finger (*Hibiscus esculentus*), 1.0 mg; yam bean (*Pachyrizus erosus*), 0.12 mg; and brinjal (*Solanum tuberosum*), 0.39 mg. The values of α-tocopherol content reported were lower than those found in this study, except for that of kangkong. However, α-tocopherol was detected in yam bean and brinjal, but was not detected in this study.

Discrepancies between the α-tocopherol content reported in this study compared to values reported by John (*16*), Konings et al. (*13*), Tsuda et al. (*28*), and Mazhar et al. (*14*) may be due to differences in variety and growing conditions. Davis et al. (*31*) reported that Avon (a soft white winter wheat) had 490 μg/100 g of total tocopherol while Coulee (a hard white winter wheat) contained 14 010 μg/100 g, a difference of more than 28-fold). The concentration of vitamins in plants can vary much depending on the time of year they are grown and harvested (*32*). Variation in factors such as light (intensity and duration), chemical and physical properties of the soils, temperature, and rainfall from one year to the next may be among the most important factors. A given plant grown in different geographical locations may have very different vitamin contents. Ascorbic acid and BHT were added during the saponification and extraction to protect α-tocopherol from oxidation (*33*). The recovery data revealed that pure tocopherols, in the absence of ascorbic acid, are much more readily decomposed in alkali by heat. The extraction efficiency of α-tocopherol was improved by bringing the ethanol/water to 0.3 ratio after saponification and by using 10% ethyl acetate in the extraction solvent (*13*).

Table 1 shows that leaves and shoots contained the highest levels of α-tocopherol, followed by fruits, rhizomes and edible roots, and flowers. Some roots, most edible flowers, and the edible mushroom studied did not contain any detectable levels of α-tocopherol. All the edible leaves/shoots and fruits analyzed contained α-tocopherol at some level.

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