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Assessment of Vitamin and Carotenoid Concentrations of Emerging Food Products: Edible Microgreens

Zhenlei Xiao,[†] Gene E. Lester,*[,‡] Yaguang Luo,[‡] and Qin Wang*[,†]

[†]Department of Nutrition and Food Science, University of Maryland, College Park, Maryland 20742, United States

[‡]Food Quality Laboratory, Beltsville Agricultural Research Center, Agricultural Research Service, U.S. Department of Agriculture, 10300 Baltimore Avenue, Beltsville, Maryland 20705, United States

ABSTRACT: Microgreens (seedlings of edible vegetables and herbs) have gained popularity as a new culinary trend over the past few years. Although small in size, microgreens can provide surprisingly intense flavors, vivid colors, and crisp textures and can be served as an edible garnish or a new salad ingredient. However, no scientific data are currently available on the nutritional content of microgreens. The present study was conducted to determine the concentrations of ascorbic acid, carotenoids, phylloquinone, and tocopherols in 25 commercially available microgreens. Results showed that different microgreens provided extremely varying amounts of vitamins and carotenoids. Total ascorbic acid contents ranged from 20.4 to147.0 mg per 100 g fresh weight (FW), while β-carotene, lutein/zeaxanthin, and violaxanthin concentrations ranged from 0.6 to 12.1, 1.3 to 10.1, and 0.9 to 7.7 mg/100 g FW, respectively. Phylloquinone level varied from 0.6 to 4.1 μg/g FW; meanwhile, α-tocopherol and γ-tocopherol ranged from 4.9 to 87.4 and 3.0 to 39.4 mg/100 g FW, respectively. Among the 25 microgreens assayed, red cabbage, cilantro, garnet amaranth, and green daikon radish had the highest concentrations of ascorbic acids, carotenoids, phylloquinone, and tocopherols, respectively. In comparison with nutritional concentrations in mature leaves (USDA National Nutrient Database), the microgreen cotyledon leaves possessed higher nutritional densities. The phytonutrient data may provide a scientific basis for evaluating nutritional values of microgreens and contribute to food composition database. These data also may be used as a reference for health agencies' recommendations and consumers' choices of fresh vegetables.

KEYWORDS: *Microgreens, phytonutrients, ascorbic acid, carotenoids, phylloquinone, tocopherols, HPLC*

INTRODUCTION

Epidemiological studies have shown that fruit and vegetable consumption is associated with reduction in the development of chronic disease, such as cancer and cardiovascular disease.[1,2] Diets rich in fruits and vegetables provide an abundance of human bioactive compounds,[3] such as ascorbic acid (vitamin C), carotenoids (provitamin A compounds), phylloquinone (vitamin K_1), and tocopherols (vitamin E), which are known to have protective benefits against cancers and cardiovascular disease.[4] The new Dietary Guidelines for Americans (2010) released by the U.S. Department of Agriculture (USDA) and the Department of Health and Human Services (DHHS) specifically recommends Americans to fill half of their plate with fruits and vegetables because they possess benefits for human health.

Microgreens are an exotic genre of edible greens, appearing in upscale markets and restaurants, that have gained popularity as a new culinary trend over the past few years. Microgreens are tender immature greens produced from the seeds of vegetables and herbs, having two fully developed cotyledon leaves with or without the emergence of a rudimentary pair of first true leaves. Microgreens are usually 2.5−7.6 cm (1−3 in.) in height, harvested at 7−14 days after germination, depending on the species, and sold with the stem and attached cotyledons (seed leaves). Although small in size, microgreens can provide a large array of intense flavors, vivid colors and tender textures. Therefore, microgreens can be served as a new ingredient in salad, soups, and sandwiches, enhancing their color, texture, and/or flavor, and also can be used as edible garnish to brighten up a wide variety of main dishes.[5−8]

Although microgreens have been claimed as nutritionally beneficial, to the best of our knowledge, no scientific data are available on the exact phytochemical content of microgreens. Limited studies have shown that some young seedlings may have much higher levels of vitamins, minerals, and other health-giving phytonutrients than the mature leaves. In a recent study from Lester et al.,[9] it was reported that the younger leaves of baby spinach (*Spinacia oleracea* L.) generally had higher levels of phytonutrients: vitamins C, B_9 and K_1, and the carotenoids (lutein, violaxanthin, zeaxanthin and β-carotene) than the more mature leaves. Oh et al.[10] also found that young lettuce (*Lactuca sativa*) seedlings, 7 days after germination, had the highest total phenolic concentration and antioxidant capacity in comparison to the older leaves. Therefore, the object of this study was to assess the vitamin and carotenoid concentrations of the 25 commercially available varieties of microgreens. The human bioactive compounds assayed include ascorbic acid (total, free, and dehydro), carotenoids (β-carotene, violaxanthin, and lutein/zeaxanthin), phylloquinone, and tocopherols (α- and γ-tocopherol).

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MATERIALS AND METHODS

Plant Materials. Twenty-five varieties of microgreens were purchased from Sun Grown Organics Distributors, Inc. (San Diego, CA) from May through July 2011. They were produced by the grower in an unheated greenhouse and under ambient light except etiolated golden pea tendrils and popcorn shoots, which were grown in the dark. All the microgreens were grown in soil and fertilized in a proprietary manner except China rose radish and green daikon radish microgreens, which were grown hydroponically. Samples were harvested without roots, packed in clamshell containers (113.4 g of each) and shipped overnight in a cardboard box which was filled with frozen-ice packs. When received, 3 g of fresh tissue was weighed for ascorbic acid analysis. Remaining tissue was frozen in liquid nitrogen and lyophilized for dry weight and other vitamin and carotenoid determinations. It is worth mentioning that golden pea tendrils and green pea tendrils are grown from the same seed source. Golden pea tendrils were grown in dark and green pea tendrils were grown under ambient light. Commercial names, scientific names, and plant colors of the 25 commercially grown microgreens assayed in this study are listed in Table 1.

Dry Weight Analysis. Dry matter was determined by freeze-drying according to a previous procedure.[11] Portions (10 g) of fresh microgreens were weighed into plastic tubes, frozen in liquid nitrogen, and lyophilized for 48 h (VirTis Freeze-mobile 35 ES Sentry 2.0 freeze-dryer, SP Scientific Corp., Warminster, PA), followed by holding at room temperature in a desiccator prior to weighing.

Nutrient Analysis. All chemicals and standards unless otherwise stated were obtained through Sigma–Aldrich Chemical Corp. (St. Louis, MO). Standards of lutein and zeaxanthin were obtained from ChromaDex (Irvine, CA).

Ascorbic Acid. Total ascorbic acid (TAA) and free L-ascorbic acid (FAA) were determined spectrophotometrically according to the procedure previously reported by Hodges et al.[12] Fresh tissue (3 g) was weighed into a 50 mL centrifuge tube, and 10 mL of ice-cold 5% (w/v) metaphosphoric acid was added, followed by homogenization at the speed of 15 000 rpm for 1 min in an ice–water bath by use of a polytron homogenizer (Brinkman Instruments, Westbury, NY). Homogenized tissue was centrifuged at 7000*g* (Beckman J2-MI, Beckman Coulter, Inc., Irving, TX) for 20 min at 4 °C, and supernatant was filtered through Whatman Grade No. 4 filter paper (Millipore Corp., Bedford, MA). Filtrate was used for FAA determination and TAA by converting dehydroascorbic acid (DAA) to FAA with dithiothreitol. TAA and FAA were determined spectrophotometrically (Genesys 20, Thermo Scientific Inc., Logan, UT) at 525 nm. Concentrations of TAA and FAA were calculated by use of an L-ascorbic acid standard curve (all $R^2 \geq 0.99$), and their difference was equal to the concentration of DAA.

Carotenoids and Tocopherols. Carotenoids and tocopherols were extracted under yellow light according to the modified method described by Lester et al.[9] Briefly, 0.05 g of lyophilized sample was weighed into a 15 mL screw-cap glass vial, and then 7.5 mL of 1% butylated hydroxytoluene (BHT) in ethanol and 500 μL of internal standard (86.82 μM *trans*-β-apo-8 carotenal) were added, followed by ultrasonic homogenization for 15 s, by using a Fisher Scientific model 300 sonic dismembrator (Pittsburgh, PA). The vials were capped under a stream of N_2 and placed in a 70 °C dry bath for 15 min, after which 180 μL of 80% KOH was added. After mixing and flushing with flow N_2, vials were capped again and placed in a 70 °C dry bath for 30 min. Vials were then removed and cooled for 5 min on ice and then the contents were transferred into 15 mL centrifuge tubes (Fisher), after which 3.0 mL of deionized water and 3.0 mL of hexane/toluene solution (10:8 v/v) were added. The mixture was vortexed for 1 min and then centrifuged at 1000*g* (Clay Adams Dynac II centrifuge, Block Scientific, Inc., Bohemia, NY) for 5 min. The top organic layer was collected into an 8 mL glass culture tube and immediately placed into a nitrogen evaporator (Organomation Associates, Inc., Berlin, MA) set at 30 °C and flushed with a stream of N_2. The bottom layer was extracted again with 3.0 mL of hexane/toluene solution (10:8 v/v) for further partition. This extraction was repeated at least four times until the top layer

Table 1. Twenty-five Commercially Grown Microgreens Assayed in the Nutrient Study

commercial name	scientific name: family	scientific name: genus and species	plant color
arugula	Brassicaceae	*Eruca sativa* Mill.	green
bull's blood beet	Chenopodiaceae	*Beta vulgaris* L.	reddish-green
celery	Apiaceae	*Apium graveolens* L.	green
China rose radish	Brassicaceae	*Raphanus sativus* L.	purplish-green
cilantro	Apiaceae	*Coriandrum sativum* L.	green
garnet amaranth	Amaranthaceae	*Amaranthus hypochondriacus* L.	red
golden pea tendrils[a]	Fabaceae	*Pisum sativum* L.	yellow
green basil	Lamiaceae	*Ocimum basilicum* L.	green
green daikon radish	Brassicaceae	*Raphanus sativus* L.var. *longipinnatus*	green
magenta spinach	Chenopodiaceae	*Spinacia oleracea* L.	red
mizuna	Brassicaceae	*Brassica rapa* L. ssp. *nipposinica*	green
opal basil	Lamiaceae	*Ocimum basilicum* L.	greenish-purple
opal radish	Brassicaceae	*Raphanus sativus* L.	greenish-purple
pea tendrils[a]	Fabaceae	*Pisum sativum* L.	green
peppercress	Brassicaceae	*Lepidium bonariense* L.	green
popcorn shoots	Poaceae	*Zea mays* L.	yellow
nutrient purple kohlrabi	Brassicaceae	*Brassica oleracea* L. var. *gongylodes*	purplish-green
purple mustard	Brassicaceae	*Brassica juncea* (L.) Czern.	purplish-green
red beet	Chenopodiaceae	*Beta vulgaris* L.	reddish-green
red cabbage	Brassicaceae	*Brassica oleracea* L. var. *capitata*	purplish-green
red mustard	Brassicaceae	*Brassica juncea* (L.) Czern.	purplish-green
red orach	Chenopodiaceae	*Atriplex hortensis* L.	red
red sorrel	Polygonaceae	*Rumex acetosa* L.	reddish-green
sorrel	Polygonaceae	*Rumex acetosa* L.	green
wasabi	Brassicaceae	*Wasabia japonica* Matsum.	green

[a]Golden pea tendrils and pea tendrils are grown from the same seeds. Golden pea tendrils are grown in dark and pea tendrils are grown under light, therefore, the colors are different (yellow and green, respectively). All the microgreens were grown organically except China rose radish and green daikon radish microgreens, which were grown hydroponically.

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was colorless, and all the supernatants were combined into a glass culture tube. After evaporation, the residue was reconstituted in 500 μL of mobile phase acetonitrile/ethanol (1:1 v/v), filtered into an HPLC amber vial through 0.22 μm nylon filter (Millipore, Bedford, MA) with a glass syringe, and 20 μL was injected for HPLC analysis. Carotenoid and tocopherol concentrations were simultaneously determined by isocratic reverse-phase high-performance liquid chromatography (RP-HPLC), which were separated on a C18 column (Adsorbosphere C18-UHS, 5 μm, 150 × 4.6 mm, Grace, Deerfield, IL) with a photo diode array detector (DAD) (G1315C, Agilent, Santa Clara, CA) and isocratic mobile phase acetonitrile/ethanol (1:1 v/v). The flow rate was 1.2 mL/min and the running time was 20 min. Absorbance was measured at 290 and 450 nm simultaneously for tocopherols and carotenoids, respectively. Quantification was based on a standard curve for each compound.

Phylloquinone. Phylloquinone was extracted from 25 microgreens under dim light and determined by RP-HPLC, as described by Booth et al.[13] Each sample (0.1 g of freeze-dried tissue) was homogenized (Brinkman Instruments, Westbury, NY) with 10 mL of H_2O and 0.4 mL of 200 μg/mL menaquinone (internal standard) at the speed of 15 000 rpm for 1 min, after which 15 mL of 2-propanol/hexane (3:2 v/v) was added. The sample was then vortexed for 1 min, and centrifuged (Beckman J2-MI, Beckman Coulter, Inc., Irving, TX) for 5 min at 1500*g*, 21 °C. The upper (hexane) layer was transferred into a glass culture tube and dried under a stream of N_2. The residue was dissolved in 4 mL of hexane. The sample extract was purified by loading 1 mL of redissolved extract onto preconditioned silica gel columns (4 mL of 3.5% ethyl ether in hexane, followed by 4 mL of 100% hexane), and then the column was washed with 2 mL of hexane. Phylloquinone was eluted with 8 mL of 3.5% ethyl ether in hexane, and the eluate was evaporated on a water-jacketed heating block (Pierce Reacti-Therm, Pierce Chemical Co., Rockford, IL) at 40 °C under N_2 flow and then reconstituted in 2 mL of mobile phase (99% methanol and 1% 0.05 M sodium acetate buffer, pH = 3.0) and filtered through a 0.22 μm nylon syringe filter (Millipore, Bedford, MA). Detection of phylloquinone was with a photodiode array detector (DAD) (G1315C, Agilent, Santa Clara, CA) on Agilent 1200 series HPLC system and absorbance wavelength was 270 nm. The extract (20 μL) was injected into HPLC and run through a C18 column (201TP, 5 μm, 150 × 4.6 mm, Grace, Deerfield, IL) with an isocratic mobile phase (described above) flowing at the rate of 1 mL/min. The phylloquinone content of the samples was quantified according to the internal standard method based on peak areas.

Statistical Analysis. Dry weight analysis and all assays were performed on three replicates. All phytonutrient analysis was conducted through one extraction of each replicate from each sample. All data are reported as the mean of three replicates ± standard error. Statistical separation of phytonutrient values per species is based on coefficient of variability (CV); this variability is in relation to the mean of the population from mature leaf data. A combined population of microgreens for each phytonutrient CV is listed in the tables.

RESULTS AND DISCUSSION

Dry Weight. Dry weight percentage of the 25 commercially available microgreens ranged from 4.6% to 10.2%, as shown in Table 2. Among them, pea tendrils had the highest dry weight percentage (10.2%) and red beet possessed the highest water content (95.4%). The overall average dry weight percentage of the 25 varieties of microgreens was 6.9% ± 0.1%.

Table 2. Mean Dry Weight Percentage and Phylloquinone Concentration in 25 Commercially Grown Microgreens[a]

microgreen name	dry weight (%)	phylloquinone (μg/g FW)
arugula	5.5 ± 0.0	1.6 ± 0.1
bull's blood beet	6.2 ± 0.1	2.0 ± 0.1
celery	6.8 ± 0.1	2.2 ± 0.1
China rose radish	8.1 ± 0.1	1.8 ± 0.1
cilantro	8.3 ± 0.1	2.5 ± 0.1
garnet amaranth	9.3 ± 0.1	4.1 ± 0.0
golden pea tendrils	9.8 ± 0.2	0.7 ± 0.0
green basil	7.3 ± 0.0	3.2 ± 0.1
green daikon radish	7.8 ± 0.1	1.9 ± 0.1
magenta spinach	5.1 ± 0.2	0.6 ± 0.0
mizuna	5.3 ± 0.0	2.0 ± 0.0
opal basil	6.8 ± 0.1	2.0 ± 0.1
opal radish	7.8 ± 0.1	2.2 ± 0.2
pea tendrils	10.2 ± 0.2	3.1 ± 0.2
peppercress	7.3 ± 0.1	2.4 ± 0.2
popcorn shoots	7.0 ± 0.1	0.9 ± 0.0
purple kohlrabi	6.1 ± 0.0	2.3 ± 0.1
purple mustard	5.7 ± 0.1	1.3 ± 0.1
red beet	4.6 ± 0.1	1.9 ± 0.1
red cabbage	7.7 ± 0.1	2.8 ± 0.1
red mustard	5.6 ± 0.1	1.9 ± 0.1
red orach	6.2 ± 0.2	0.7 ± 0.0
red sorrel	7.0 ± 0.1	3.3 ± 0.0
sorrel	4.9 ± 0.0	1.7 ± 0.1
wasabi	5.6 ± 0.0	1.9 ± 0.1
coefficient of variation		15%

[a]Values are expressed as means ± standard error (n = 3).

Phylloquinone. Vitamin K_1 is required for blood coagulation and is most abundant in photosynthetic tissues of dark-green vegetables, such as spinach (*Spinacia oleracea* L.), kale (*Brassica oleracea* L. var. *acephala*), and broccoli (*Brassica oleracea* var. *italica*) .[14] Among the 25 microgreens assayed, there was considerable variation in phylloquinone concentration, ranging from 0.6 to 4.1 μg/g freight weight (FW) as shown in Table 2. Among them, the most concentrated in phylloquinone was garnet amaranth (4.1 μg/g FW) (Figure 1), followed by red sorrel (3.3 μg/g FW), green basil (3.2 μg/g FW), pea tendrils (3.1 μg/g FW), and red cabbage (2.8 μg/g FW) microgreens. In contrast, magenta spinach, golden pea tendrils, red orach microgreens, and popcorn shoots had vitamin K_1 concentration ranging from 0.6 to 0.9 μg/g FW. Samples identified as rich in phylloquinone were generally green (e.g., pea tendrils) or bright red in color (e.g., garnet amaranth microgreens), while yellow-colored microgreens, such as popcorn shoots and golden pea tendrils, had relatively low concentration of vitamin K_1, which is in agreement with a previous report.[14] Surprisingly, magenta spinach, which has a similar appearance to the leading vitamin K_1 microgreen source, garnet amaranth (4.1 μg/g FW), had among the lowest vitamin K_1 concentrations. Comparison of fully grown and cotyledon leaves demonstrated that growth stage affected vitamin K_1 concentration, and for some of the varieties, the effect was obvious. For example, according to the USDA national nutrient database,[15] phylloquinone concentration in mature amaranth, basil, and red cabbage were 1.14, 0.41, and 0.04 μg/g FW,

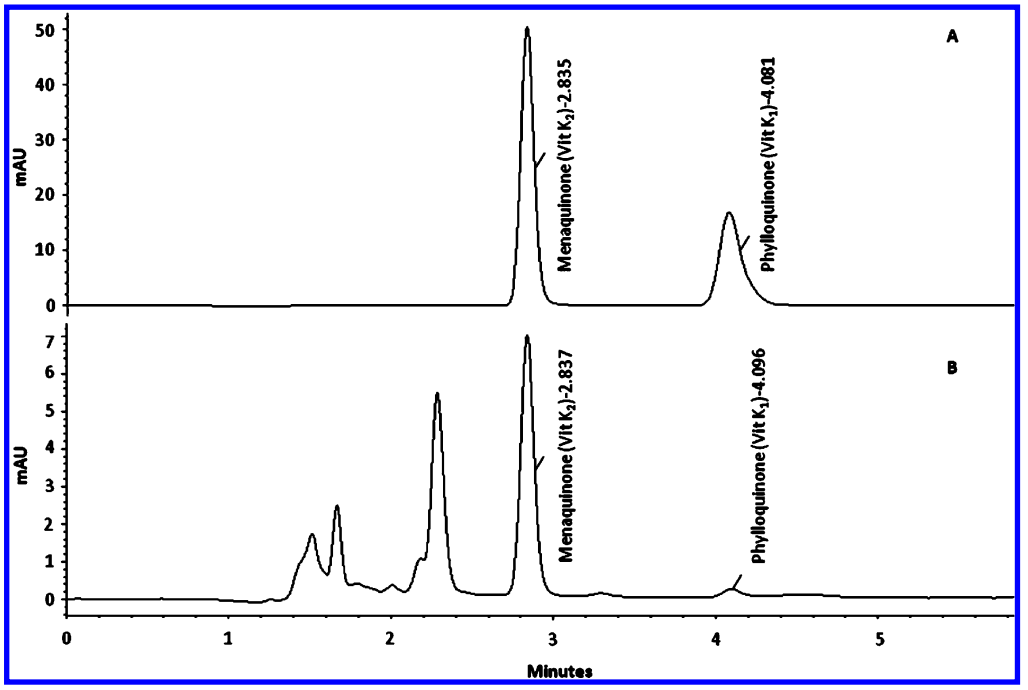

Figure 1. HPLC chromatograms of (A) vitamin K standards and (B) extraction of garnet amaranth microgreens. Menaquinone (vitamin K_2) is the internal standard. HPLC conditions are described under Materials and Methods.

Table 3. Mean Total Ascorbic Acid (TAA), Free Ascorbic Acid (FAA), and Dehydroascorbic Acid (DAA) Concentrations in 25 Commercially Grown Microgreens[a]

microgreen name	TAA (mg/100 g FW)	FAA (mg/100 g FW)	DAA (mg/100 g FW)
arugula	45.8 ± 3.0	32.7 ± 1.3	13.2 ± 2.8
bull's blood beet	46.4 ± 3.0	46.0 ± 3.3	0.5 ± 0.3
celery	45.8 ± 3.1	32.6 ± 1.3	13.2 ± 2.8
China rose radish	95.8 ± 10.3	73.2 ± 3.4	22.6 ± 7.4
cilantro	40.6 ± 2.4	24.5 ± 1.8	16.1 ± 2.2
garnet amaranth	131.6 ± 2.9	105.1 ± 3.1	26.5 ± 1.4
golden pea tendrils	25.1 ± 0.7	15.3 ± 1.7	9.8 ± 1.2
green basil	71.0 ± 2.7	59.0 ± 1.8	12.0 ± 1.1
green daikon radish	70.7 ± 2.7	58.8 ± 1.7	11.9 ± 1.1
magenta spinach	41.6 ± 0.8	36.0 ± 0.8	5.6 ± 0.2
mizuna	42.9 ± 1.6	32.3 ± 1.0	10.6 ± 0.7
opal basil	90.8 ± 2.7	81.8 ± 1.6	9.0 ± 2.0
opal radish	90.1 ± 2.7	81.1 ± 1.7	9.0 ± 1.9
pea tendrils	50.5 ± 0.9	27.9 ± 1.1	22.5 ± 0.3
peppercress	57.2 ± 1.6	33.0 ± 0.7	24.2 ± 1.8
pop corn shoots	31.8 ± 0.7	21.4 ± 2.5	10.4 ± 3.0
purple kohlrabi	62.8 ± 7.3	48.1 ± 3.7	14.7 ± 3.7
purple mustard	72.1 ± 4.6	53.6 ± 2.6	18.5 ± 4.4
red beet	28.8 ± 0.4	27.5 ± 0.3	1.3 ± 0.5
red cabbage	147.0 ± 3.6	103.3 ± 9.0	43.7 ± 5.4
red mustard	62.2 ± 2.6	40.8 ± 1.4	21.4 ± 1.3
red orach	45.4 ± 0.9	43.7 ± 0.9	1.7 ± 0.2
red sorrel	56.7 ± 1.4	41.9 ± 1.9	14.9 ± 0.7
sorrel	20.4 ± 0.5	17.9 ± 0.3	2.6 ± 0.2
wasabi	44.8 ± 1.9	35.0 ± 2.0	9.8 ± 0.1
coefficient of variation	12%	18%	35%

[a]Values are expressed as mean ± standard error (n = 3).

respectively, which were much lower than the values for their corresponding microgreens (4.09, 3.20, and 2.77 μg/g FW, respectively). Four of the 25 microgreen varieties assayed in this study had comparable amount of phylloquinone to mature leaf spinach, which is generally considered as an excellent source of vitamin K_1; and 18 out of 25 exhibited vitamin K_1 densities equal to or higher than that of broccoli, the most commonly consumed vegetable in the United States;[14,15] demonstrating that most of the 25 microgreens can serve as good natural sources of vitamin K_1.

Ascorbic Acid. Ascorbic acid (vitamin C) is an essential nutrient for the human body, acting as an antioxidant. When the plant is subject to physical or physiological stress (harvesting injury, chilling, irradiation, etc.), the FAA can be oxidized into DAA.[12] It was previously reported that the utilization of DAA is equivalent to that of FAA, although the metabolic turnover time is different.[16] In this study, TAA, FAA, and DAA concentration were determined and are listed in Table 3. The 25 microgreens exhibited TAA content ranging from 20.4 to 147.0 mg/100 g FW. Among samples tested, red cabbage and garnet amaranth microgreens had the highest TAA contents, followed by China rose radish, opal basil, and opal radish. The vitamin C concentration of red cabbage microgreens (147.0 mg/100 g FW) was 6-fold higher than previously published data for mature red cabbage (24.4 mg/100 g FW) [17] and 2.6 times greater than that (57.0 mg/100 g FW) recorded in the USDA National Nutrient Database for Standard Reference, Release 24,[15] and was determined to be 2.4 times greater than the estimated average requirement (EAR) for ascorbic acid. Garnet amaranth (131.6 mg/100 g FW) had much higher ascorbic acid content than reported concentration of mature leaf (11.6–45.3 mg/100 g FW).[18,19] China rose radish, opal basil, and opal radish microgreens also were relatively abundant sources of vitamin C with more than 90.0 mg/100 g FW, equal to 1.5 times the recommended dietary allowance (RDA).These microgreen varieties had higher

Table 4. Mean β-Carotene, Violaxanthin, and Lutein/Zeaxanthin Concentrations in 25 Commercially Grown Microgreens[a]

microgreen name	β-carotene (mg/100 g FW)	lutein/zeaxanthin (mg/100 g FW)	violaxanthin (mg/100 g FW)
arugula	7.5 ± 0.4	5.4 ± 0.2	2.6 ± 0.1
bull's blood beet	5.3 ± 0.8	4.3 ± 0.7	2.3 ± 0.1
celery	5.6 ± 0.1	5.0 ± 0.1	2.6 ± 0.1
China rose radish	5.4 ± 0.5	4.9 ± 0.4	1.9 ± 0.1
cilantro	11.7 ± 1.1	10.1 ± 0.3	7.7 ± 0.6
garnet amaranth	8.6 ± 0.3	8.4 ± 0.1	4.4 ± 0.1
golden pea tendrils	0.6 ± 0.0	2.7 ± 0.0	1.0 ± 0.1
green basil	8.4 ± 0.4	6.6 ± 0.3	2.7 ± 0.2
green daikon radish	6.1 ± 0.1	4.5 ± 0.1	1.7 ± 0.0
magenta spinach	5.3 ± 0.3	3.2 ± 0.2	3.7 ± 0.5
mizuna	7.6 ± 0.4	5.2 ± 0.3	2.4 ± 0.1
opal basil	6.1 ± 0.4	5.3 ± 0.3	2.0 ± 0.0
opal radish	6.3 ± 1.0	5.5 ± 0.9	2.3 ± 0.4
pea tendrils	8.2 ± 1.1	7.3 ± 1.2	3.9 ± 1.4
peppercress	11.1 ± 0.6	7.7 ± 0.4	3.1 ± 0.2
popcorn shoots	0.6 ± 0.1	1.3 ± 0.1	0.9 ± 0.1
purple kohlrabi	5.7 ± 0.2	4.0 ± 0.1	1.5 ± 0.0
purple mustard	5.6 ± 0.4	6.4 ± 1.9	1.0 ± 0.2
red beet	7.7 ± 0.1	5.5 ± 0.0	3.7 ± 0.0
red cabbage	11.5 ± 1.2	8.6 ± 1.0	2.9 ± 0.3
red mustard	6.5 ± 0.4	4.9 ± 0.3	1.7 ± 0.1
red orach	6.3 ± 0.3	3.9 ± 0.2	3.2 ± 0.2
red sorrel	12.1 ± 0.6	8.8 ± 0.2	3.6 ± 0.1
sorrel	5.2 ± 1.0	4.2 ± 0.8	1.3 ± 0.3
wasabi	8.5 ± 0.2	6.6 ± 0.3	2.2 ± 0.2
coefficient of variation	31%	18%	18%

[a]Values are expressed as mean ± standard error (n = 3).

ascorbic acid concentration than does broccoli (89.2 mg/100 g FW),[15] which is generally recognized as an excellent source of vitamin C. Even though some of the 25 microgreens tested had relatively low levels of total ascorbic acid, such as golden pea tendrils (25.1 mg/100 g FW) and sorrel microgreens (20.4 mg/100 g FW), they were comparable to spinach (28.1 mg/100 g FW),[15] which is one of the most commonly consumed leaf vegetables in the United States. Therefore, it was suggested that fresh microgreens are generally good to excellent sources of ascorbic acid and likely more concentrated with TAA than their mature plant counterparts, which is in accordance with the findings of Bergquist et al.[1] on baby spinach: that younger plants had higher ascorbic acid content than older harvested leaves.

Carotenoids. *β-Carotene.* β-Carotene (provitamin A) is an important fat-soluble antioxidant and can protect cellular membranes by scavenging free radicals.[17] As shown in Table 4, β-carotene levels ranged from 0.6 to 12.1 mg/100 g FW. Among the tested microgreens, red sorrel had the highest β-carotene concentration (12.1 mg/100 g FW), followed by cilantro, red cabbage, and peppercress (11.7, 11.5, and 11.1 mg/100 g FW, respectively). The lowest β-carotene concentration was found in golden pea tendrils and popcorn shoots (around 0.6 mg/100 g FW), with the other microgreens at intermediate values (5.2 to 8.6 mg/100 g FW). Compared with fully developed cilantro leaves, cilantro seedlings contained 3-fold more β-carotene. Red cabbage microgreens contained an average of 11.5 mg/100 g FW which is approximately 260-fold more than the value (0.044 mg/100 g FW) reported for mature red cabbage leaves.[17] Wasabi, green basil, pea tendrils, and garnet amaranth microgreens are also abundantly concentrated with β-carotene. The β-carotene concentration in these microgreens is comparable to that of carrot (*Daucus carota* L.) and sweet potato (*Ipomoea batatas* (L.) Lam), which are well-known β-carotene-rich vegetables.[15,18] In summary, almost all the microgreens tested can be considered as excellent sources of β-carotene, with the exceptions of popcorn shoots and golden pea tendrils.

Lutein/Zeaxanthin. Lutein and zeaxanthin are xanthophyll carotenoids, accumulating in the macula of human eyes. Numerous epidemiological studies have shown that lutein and zeaxanthin play a critical role in the prevention of age-related macular degeneration and cataract.[20] In the analysis of lutein and zeanthaxin, these two isomers were coeluted in our HPLC system, so all values were calculated on the basis of area under the curve of lutein standard and expressed in lutein equivalents but represent as the sum of lutein and zeaxanthin. While all 25 microgreens assayed in this study contained lutein and zeaxanthin (Table 4), cilantro had the highest lutein/zeaxanthin levels with 10.1 mg/100 g FW (Figure 2). Red sorrel, red cabbage, and garnet amaranth microgreens followed with lutein/zeaxanthin concentrations of 8.8, 8.6, and 8.4 mg/100 g FW, respectively. These values were higher than that of mature spinach (7.2 mg/100 g FW), which contains high quantities of lutein/zeaxanthin.[21] The lowest concentration of lutein/zeaxanthin, 1.3 mg/100 g FW was found in popcorn shoots. According to the USDA national nutrient database,[15] it was determined that the values of lutein/zeaxanthin in raw mature cilantro and red cabbage were 0.9 and 0.3 mg/100 g FW, respectively, which contrasted with the more abundant concentrations in their microgreen counterparts, which had 11.2 and 28.6 times greater lutein/zeaxanthin concentrations, respectively. These findings suggest that these immature leaves of the microgreens tend to possess higher lutein/zeaxanthin concentration than their fully grown plant counterparts.[15]

Violaxanthin. Violaxanthin is a natural orange-colored carotenoid found in photosynthetic organs of plants. The concentration of violaxanthin in the 25 microgreens varied considerably, with cilantro microgreens containing 7.7 mg/100 g FW violaxanthin while popcorn shoots and golden pea tendrils contained only 0.9 and 1.0 mg/100 g FW violaxanthin, respectively (Table 4). The rest of the microgreens had violaxanthin ranging from 1.3 to 4.3 mg/100 g FW. The maximum concentration of violaxanthin in cilantro microgreens was more than 5-fold than that of mature cilantro leaves (1.4 mg/100 g FW) and 2.8 times than that of mature spinach (2.7 mg/100 g FW), both of which are considered as good sources of violaxanthin.[22,23] Twenty-two out of the 25 microgreens assayed possessed violaxanthin concentration higher than mature cilantro, and 40% of them were at levels equal to or higher than commonly consumed mature-leaf spinach and baby-leaf spinach.[9]

Tocopherols. Tocopherols and tocotrienols are together summarized as "vitamin E", known as fat-soluble antioxidants.

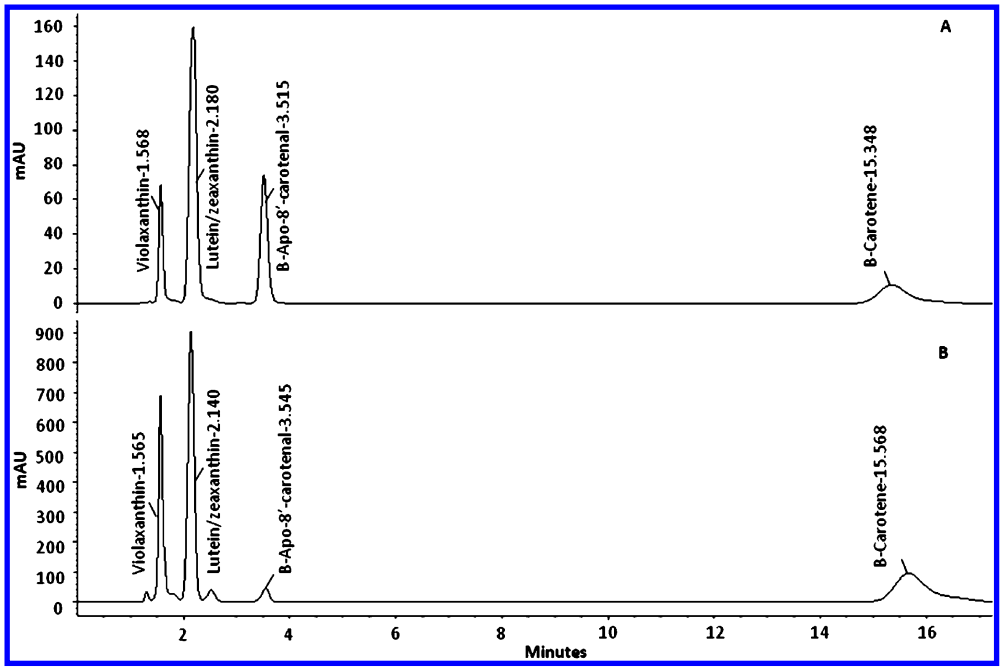

Figure 2. HPLC chromatograms of (A) carotenoid standards and (B) extraction of cilantro microgreens. β-Apo-8′-carotenal is the internal standard, and lutein and zeaxanthin are coeluted. HPLC conditions are described under Materials and Methods.

Table 5. Mean α- and γ-Tocopherol Concentration in 25 Commercially Grown Microgreens[a]

microgreen name	α-tocopherol (mg/100 g FW)	γ-tocopherol (mg/100 g FW)
arugula	19.1 ± 4.3	7.1 ± 2.4
bull's blood beet	18.5 ± 2.5	5.0 ± 0.7
celery	18.7 ± 5.1	6.1 ± 1.4
China rose radish	19.7 ± 3.1	7.5 ± 1.1
cilantro	53.0 ± 13.5	12.5 ± 2.0
garnet amaranth	17.1 ± 2.1	11.2 ± 1.3
golden pea tendrils	4.9 ± 0.3	3.0 ± 0.2
green basil	19.9 ± 0.3	6.0 ± 0.4
green daikon radish	87.4 ± 15.9	39.4 ± 7.8
magenta spinach	14.2 ± 3.3	5.1 ± 0.8
mizuna	25.0 ± 3.7	9.6 ± 1.4
opal basil	24.0 ± 2.1	8.3 ± 0.8
opal radish	47.7 ± 14.6	16.7 ± 5.3
pea tendrils	35.0 ± 6.8	8.3 ± 2.0
peppercress	41.2 ± 3.7	14.5 ± 1.4
popcorn shoots	7.8 ± 0.1	3.5 ± 0.0
purple kohlrabi	13.8 ± 1.0	5.6 ± 0.4
purple mustard	18.6 ± 1.3	7.0 ± 0.7
red beet	34.5 ± 2.3	8.3 ± 0.6
red cabbage	24.1 ± 5.5	10.3 ± 3.1
red mustard	22.1 ± 1.9	8.2 ± 0.7
red orach	18.3 ± 2.8	7.0 ± 0.9
red sorrel	21.8 ± 1.2	7.7 ± 0.5
sorrel	9.3 ± 1.5	3.1 ± 0.5
wasabi	18.7 ± 2.9	7.6 ± 1.0
coefficient of variation	20%	16%

[a]Values are expressed as mean ± standard error (n = 3).

Each group has four isomers (α, β, γ, and δ). The most active form of all the tocopherols is α-tocopherol, while γ-tocopherol is the most abundant form in plants.[24] In this study, α-and γ-tocopherol contents for the 25 different varieties of microgreens are summarized (Table 5). Green daikon radish has extremely high α- and γ-tocopherol contents of 87.4 and 39.4 mg/100 g FW, respectively (Figure 3). In addition, cilantro, opal radish, and peppercress microgreens are also excellent sources of α- and γ-tocopherol, with the α-tocopherol concentrations ranging from 41.2 to 53.1 mg/100 g FW and γ-tocopherol values from 12.5 to 16.7 mg/100 g FW. Even though the values of α-tocopherol (4.9 mg/100 g FW) and γ-tocopherol (3.0 mg/100 g FW) in golden pea tendrils were among the lowest of the 25 microgreens, their values were still markedly higher than those for more mature spinach leaves (2.0 and 0.2 mg/100 g FW, respectively).[15] Red cabbage microgreens contained over 40 times the vitamin E content of its mature counterpart (0.06 mg/100 g FW) reported by Podsedek et al.[25]

In summary, the essential vitamin and carotenoid concentrations of 25 commercially available microgreens varieties have been determined. In general, microgreens contain considerably higher concentrations of vitamins and carotenoids than their mature plant counterparts, although large variations were found among the 25 species tested. Maximum values of vitamin C, viamin K_1, and vitamin E were found in red cabbage, garnet amaranth, and green daikon radish microgreens, respectively. In terms of carotenoids, cilantro microgreens showed the highest concentration of lutein/zeaxanthin and violaxanthin and ranked second in β-carotene concentration. In contrast, popcorn shoots and golden pea tendrils were relatively low in vitamins and carotenoids, although they were still comparable nutritionally to some commonly consumed mature vegetables. It is also noted that golden pea tendrils, which are grown in the absence of light, processed much lower vitamin and carotenoid concentrations than pea tendrils grown under light, suggesting that light plays an important role on nutriential values during the growth of microgreens. The data generated by this research likely provide a scientific basis for evaluating the vitamin and carotenoid concentrations of microgreen cotyledon leaves. It

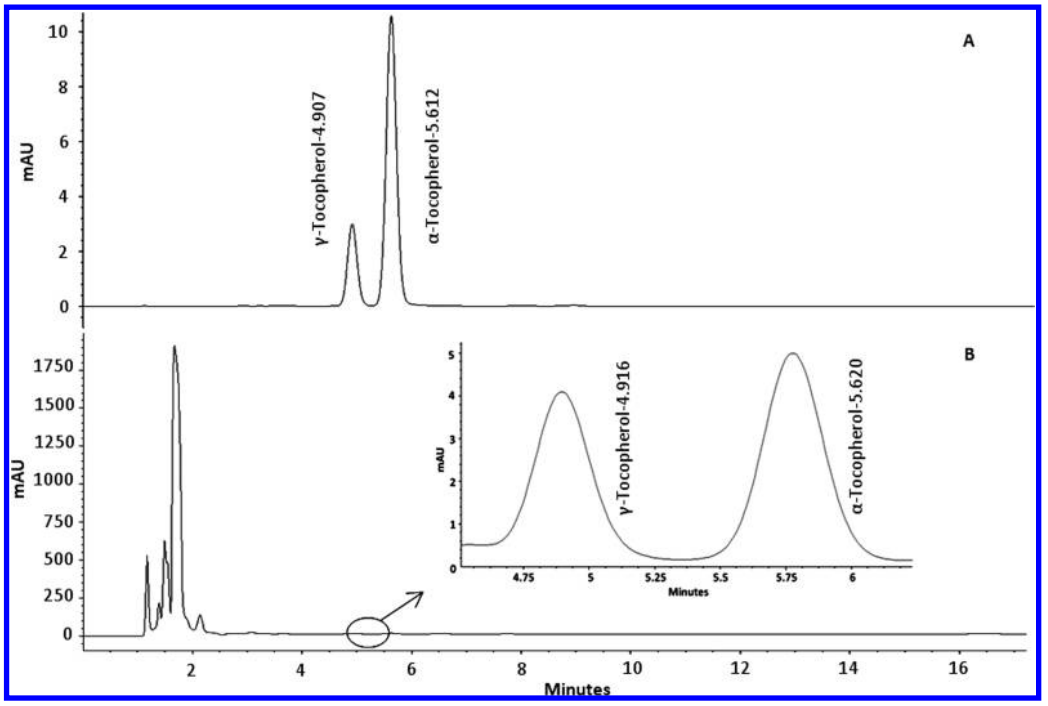

Figure 3. HPLC chromatograms of (A) tocopherol standards and (B) extraction of green daikon radish microgreens. HPLC conditions are described under Materials and Methods.

can also be used as a possible reference in estimating the dietary intake and adequacies of vitamins from microgreens. However, since growing, harvesting, and postharvest handling conditions may have a considerable impact on the synthesis and degradation of phytonutrients, including vitamins and carotenoids, additional studies may be needed to evaluate the effect of these agricultural practices on phytonutrient retention.

AUTHOR INFORMATION

Corresponding Authors

*(G.L.) Telephone (301) 504-5981; fax (301) 504-5107; e-mail gene.lester@ars.usda.gov. *(Q.W.) Telephone (301) 405-8421; fax (301) 314-3313; e-mail wangqin@umd.edu.

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