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Variation in Major Antioxidants and Total Antioxidant Activity of Yuzu (*Citrus junos Sieb ex Tanaka*) during Maturation and between Cultivars

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Epidemiological studies suggest that a high consumption of fruits can reduce the risk of some cancers and cardiovascular disease, and this may be attributable to the antioxidant activity of vitamins and phenolic compounds. The present study investigated the variations in vitamin C, total phenolic, hesperidin, and naringin contents, and total antioxidant activity of yuzu (*Citrus junos Sieb ex Tanaka*)—which is a popular citrus fruit in Korea and Japan—between cultivars and during maturity. The amounts of phenolics and vitamin C and the antioxidant activity in all tested yuzu cultivars were higher in peel than in flesh. Ripening increased the total antioxidant activity and vitamin C content in both peel and flesh of yuzu. However, the amounts of all total phenolics, hesperidin, and naringin in peel increased with ripening, whereas they decreased slightly in flesh. There was a highly linear relationship between the vitamin C content and the total antioxidant activity in both peel ($r^2 = 1.000$) and flesh ($r^2 = 0.998$), suggesting that vitamin C plays a key role in the antioxidant activity of yuzu. In addition, the contribution of each antioxidant to the total antioxidant activity of yuzu was determined using a 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) radical scavenging assay and is expressed here in terms of the vitamin C equivalent antioxidant capacity (VCEAC). The means of vitamin C, naringin, and hesperidin in yuzu were 90.4, 63.8, and 65.7 mg/100 g fresh yuzu, respectively. The relative VCEAC values of these compounds were in the following order: vitamin C (1.00) > naringin (0.195) > hesperidin (0.162). Therefore, the estimated contribution of each antioxidant to the total antioxidant capacity of 100 g of fresh yuzus is as follows (in mg of VCEAC): vitamin C (90.36 mg) > naringin (12.44 mg) > hesperidin (10.64 mg). Our results indicate that mature yuzu contains higher amounts of vitamin C and phenolics than other citrus fruits and could therefore be used as a significant dietary source of antioxidants.

KEYWORDS: Yuzu (*Citrus Junos Sieb ex Tanaka*); antioxidant; vitamin C equivalent antioxidant capacity (VCEAC); vitamin C; hesperidin; naringin

INTRODUCTION

The results of epidemiological studies suggest that a high consumption of fruits reduces the risk of degenerative diseases such as cardiovascular disease, several types of cancer, and neurological disease (*1*−*3*). There is accumulating evidence that these effects of fruits are attributable to antioxidants such as vitamins and phenolic phytochemicals (*10*, *43*), because naturally occurring antioxidants have been reported to play a key role in ameliorating oxidative damage induced by the free radicals that cause several human diseases. Vitamin C is considered one of the most prevalent antioxidative components of fruits that exert substantial chemopreventive effects without apparent toxicity [reviewed in (*4*) and references therein]. Recent reports suggest that the chemopreventive effects of vitamin C are linked to its protective effects against epigenetic mechanisms such as the inflammation and inhibition of gap−junction intercellular communication as well as antioxidant activities (*5*−*9*). Phenolic substances have also been proposed as important contributors to the total antioxidant capacity (TAC) of fruits (*10*). Much attention has recently been paid to the possible health benefits of dietary phenolic phytochemicals that exhibit antioxidative, anticarcinogenic, antimicrobial, antiallergic, antimutagenic, and antiinflammatory activities (*11*−*14*).

The health benefits of citrus fruits have been largely attributed to the presence of the antioxidant vitamin C (*10*), and other

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studies have shown the additional roles of flavonoid components (*15*, *16*). Yuzu (*Citrus junos Sieb ex Tanaka*) is a citrus fruit native to northeast Asia, including Korea, China, and Japan (*17*). Particularly in Korea, it has been commonly used as a raw material for beverages and herbal medicines due to its unique flavor and effectiveness against colds (*18*, *19*). The high content of vitamin C and phenolic substances in yuzus might be associated with significant health benefits.

The TAC of fruits is mainly due to the combined activity of diverse antioxidants including vitamin C and phenolics, rather than being attributable to any particular antioxidant; hence, investigations into the antioxidant capacity of fruits should consider the overall concentrations and compositions of diverse antioxidants therein. Furthermore, the distribution and composition of phenolic phytochemicals are affected by maturity, cultivar, horticultural practices, geographic origin, growing season, postharvest storage conditions and duration, and processing procedures (*20*−*26*). Several studies have shown that citrus fruits such as oranges, lemons, and limes exhibit an antioxidant activity that is mainly attributable to vitamin C and phenolic substances (*16*), but the variations in the amounts of major antioxidants and the total antioxidant activity of yuzus between cultivars and during maturity have not been investigated. Therefore, the objective of this study was to determine the variations in vitamin C, total phenolic, hesperidin, and naringin contents, and total antioxidant activity of yuzu between cultivars and during maturity.

MATERIALS AND METHODS

Fruits. The Wando, Goheung, and Sadeung yuzus (*C. junos Sieb ex Tanaka*) fruits used in this study were grown at the Agricultural Research Centers in Jeolla province, South Korea. Wando yuzus were harvested in 2001 at various stages of maturity, with the degree of fruit maturity determined from the fruit surface color: unripe fruits picked on October 15 were green, fruits of intermediate ripeness picked on November 15 were yellow, and ripe fruits picked on December 15 were deep yellow. They were harvested under the auspices of the National Agricultural Cooperative Federation. After they were harvested, slices of yuzu prepared by hand were frozen at −80 °C and freeze-dried. The freeze-dried samples were ground to powder (FM-700W food mixer, Han Il, Korea) and then stored at −20 °C.

Chemicals. Chlorogenic acid, 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) as the diammonium salt, hesperidin, naringin, vitamin C, and Folin−Ciocalteu phenol reagent were obtained from Sigma (St. Louis, MO). 2,2′-Azobis-(2-amidino-propane)dihydrochloride (AAPH) was obtained from Wako Chemicals (Richmond, VA). All other chemicals used were analytical grade (Fisher, Springfield, NJ).

Extraction of Phenolics. The phenolics in yuzu (*9*, *14*) were extracted from 10 g of ground freeze-dried yuzu using 100 mL of 80% aqueous methanol in a 200 mL round-bottomed flask. The mixture of yuzu powder and aqueous methanol was sonicated for 30 min, and the mixture was filtered through Whatman no. 2 filter paper (Whatman International, Kent, United Kingdom) using a chilled Büchner funnel and rinsed with 50 mL of 80% aqueous methanol. The solid filter cake was reextracted by repeating the above steps under the same conditions. The two filter cakes obtained were transferred into a 1000 mL round-bottomed flask with an additional 50 mL of 80% aqueous methanol. The solvent was evaporated under reduced pressure at 40 °C. The phenolic concentrate (492.1 mg) was dissolved in 50 mL of 80% aqueous methanol and made to a final volume of 100 mL with 80% aqueous methanol. The solution was then centrifuged at 8000*g* for 15 min at 4 °C. The final extract solution was stored at −4 °C until analyzed.

Total Phenolics Contents. The total contents of phenolic phytochemicals were measured using the Folin−Ciocalteu method. Briefly, 1 mL of appropriately diluted samples or a standard solution of chlorogenic acid was added to a 25 mL volumetric flask containing 9 mL of ddH_2O. A reagent blank was prepared using ddH_2O. One milliliter of Folin−Ciocalteu phenol reagent was added to the mixture and mixed by shaking. After 5 min, 10 mL of 7% Na_2CO_3 solution was added with mixing. The solution was then immediately diluted to a volume of 25 mL with ddH_2O and mixed thoroughly. After incubation for 90 min at 23 °C, the absorbance relative to that of a prepared blank was read at 750 nm using a spectrophotometer (model DU 530, Beckman, United States). The total phenolics contents are expressed here in mg of chlorogenic acid equivalents (CAE) per 100 g fresh weigh as standard. Chlorogenic acid is the predominant phenolic acid in fruits, which possesses very little gallic acid (*41*, *42*).

Hesperidin and Naringin Contents. The contents of hesperidin and naringin were determined according to a method described elsewhere (*27*). Citrus fruits were divided into peel and flesh. The flesh—which consisted of juice sacs and segment epidermis—was ground, and 5 g of the sample was extracted three times with 50 mL of MeOH and dimethyl sulfoxide (1:1). The extracts were combined and made up to 5 mL with MeOH. Twenty microliters of the combined extract was injected into an high-performance liquid chromatography (HPLC) system (HP 1090 series II, Hewlett-Packard, MO), and the bioflavonoids were analyzed using a modified version of the method of Vandercook and Tisserat (*27*). The analysis of hesperidin and naringin utilized a TSK-gel ODS-80TS column (particle size 5 μm, 25 cm × 4.6 nm i.d.; Tosoh, Tokyo, Japan) in the mobile phase with a gradient elution program consisting of an initial 2 min of 80% 0.01 M H_3PO_4 and 20% MeOH followed by a linear gradient over 43 min to 55% 0.01 M H_3PO_4 and 45% MeOH. The UV diode array detector was set to measure spectra from 200 to 400 nm, and the eluent was monitored at 285 nm for hesperidin and naringin. The contents of hesperidin and naringin are expressed here in mg per 100 g of fresh weight or dry matter.

Ascorbic Acid Content. The ascorbic acid content of the yuzus was determined according to a procedure described elsewhere (*28*). Yuzus were homogenized and extracted with 2% (v/v) acetic acid for 3 min. An HPLC system (HP 1090 series II, Hewlett-Packard) equipped with a UV diode array detector was used. The separation was performed on a C18 column with a particle size of 5 μm, 25 cm × 3.0 nm (i.d.) (Shiseido, Tokyo, Japan) using 2% (v/v) acetic acid/acetonitrile at a flow rate of 0.5 mL/min at 40 °C. The ascorbic acid contents are expressed here in mg per 100 g of fresh weight or dry matter.

ABTS Radical Scavenging Activity. A method developed by van den Berg et al. was used with slight modifications to measure the ABTS radical scavenging activity (*30*). AAPH (1.0 mM) was mixed with 2.5 mM ABTS as the diammonium salt in phosphate-buffered saline solution (100 mM potassium phosphate buffer, pH 7.4, containing 150 mM NaCl) and heated in a 68 °C water bath for 13 min. The concentration of the resulting blue-green ABTS radical solution was adjusted to an absorbance of 0.650 ± 0.020 (mean ± SD) at 734 nm. Twenty microliter samples of the solution at various concentrations were added to 980 μL of the resulting blue-green ABTS radical solution. The mixture was incubated in darkness in a 37 °C water bath for 10 min, and the decrease in absorbance at 734 nm was measured. A control solution consisted of 20 μL of 50% methanol and 980 μL of ABTS radical solution. Stable ABTS radical scavenging activities of the samples are expressed here in milligrams per serving of vitamin C equivalent antioxidant capacity (VCEAC). The radical stock solution was freshly prepared each day.

Quantification of TAC. A method developed by Winston et al. (*31*) was applied with slight modifications for quantification of the antioxidant value of each compound tested. The area under the kinetic curve was calculated by integration. The TAC of each tested compound was then quantified according to

$$\mathrm{TAC} = 100 - \left(\int \mathrm{SA} / \int \mathrm{CA} \times 100\right) \quad (1)$$

where $\int$ SA and $\int$ CA are the integrated areas under the curves defining the samples and control reactions, respectively. The percentage increases in integrated areas were used to compare each phenolic compound and vitamin C content. The median effective dose (EC_{50}) of all samples tested was calculated from the dose−response curve.

Table 1. Weights and Relative Rations of Peel, Flesh, and Seed of Wando Yuzus (*C. junos Sieb ex Tanaka*) at Different Stages of Maturation[a]

	green (g)	yellow (g)	deep yellow (g)
peel	50.1 ± 4.0 (43.77%)	50.1 ± 4.7 (43.46%)	49.4 ± 4.3 (42.11%)
flesh	52.6 ± 3.4 (45.92%)	52.8 ± 5.1 (45.78%)	56.2 ± 3.5 (47.88%)
seed	11.8 ± 0.7 (10.31%)	12.4 ± 1.2 (10.76%)	11.7 ± 1.9 (10.01%)
total	114.5 ± 7.2 (100%)	115.3 ± 9.3 (100%)	117.4 ± 8.2 (100%)

[a] Data are expressed as means ± SEM ($n = 20$). Unripe green, intermediate mature yellow, and mature deep yellow yuzus were harvested at Wando on October 15, November 15, and December 15, respectively.

Antioxidant Capacity. The antioxidant capacity of the samples was measured and calculated as VCEAC according to the method described by Kim et al. (*29*). Briefly, vitamin C standard curves that correlate the concentration of vitamin C and the amount of absorbance reduction caused by vitamin C were obtained using the ABTS radical scavenging assay. This assay was also used to measure absorbance reductions (at 734 nm) of the samples at various concentrations. The determination of VCEACs of the samples at various concentrations was made using vitamin C standard curves. The EC_{50} values of the samples were calculated from the dose−response curves. The absorbance reduction of the samples was correlated to that of vitamin C standards, with the results calculated as VCEAC values. All data are presented as means ± SD and are from at least five replications for each sample.

Statistical Analysis. Significance was evaluated by analysis of variance (ANOVA) followed by Duncan's protected least significant difference test. Probability values of $p < 0.05$, $p < 0.01$, and $p < 0.001$ were used as the criteria for significant differences.

RESULTS AND DISCUSSION

The total weight and flesh weight, but not peel weight, increased as the yuzus ripened (**Table 1**). The total phenolics contents of the three yuzu cultivars are listed in **Table 2**, among which significant differences ($p < 0.001$) were found in their total phenolics contents: from 294.3 mg CAE/100 g in the peel of Wando yuzus to 206.3 mg CAE/100 g in the flesh of Goheung yuzus. There were also differences between the cultivars in the contents of vitamin C, hesperidin, and naringin of peel and flesh. In all cultivars, the levels of vitamin C were higher in the peel than in the flesh. The peel of Wando yuzus had the highest vitamin C content (107.1 mg/100 g fresh weight), while the flesh of Goheung yuzus had the lowest (60.6 mg/100 g fresh weight). Lemon peel contains 59.8 mg/100 g fresh vitamin C content, and that of orange peel contains 59.6 mg/100 g vitamin C content (*16*). Hence, the peel of Wando yuzus had two times more vitamin C than that of orange. The contents of hesperidin and naringin were in the following order: Wando yuzus > Goheung yuzus > Sadeung yuzus. The peel of Wando yuzus also contained the highest amounts of hesperidin and naringin (96.1 and 98.1 mg/100 g, respectively), while the lowest values were found in the flesh of Goheung yuzu (53.9 and 48.7 mg/100 g, respectively). These results indicate that Wando yuzu has more vitamin C and phenolic phytochemicals and a higher antioxidant capacity than Goheung and Sadeung yuzus.

In all cultivars tested, the levels of all antioxidants were higher in the peel than in the flesh. We measured the change of antioxidant contents in Wando yuzu during maturation. The total phenolic contents of whole yuzu except seed increased with maturity: In the peel, it increased from 246.5 to 294.4 mg/100 g, whereas in the flesh it decreased slightly from 231.5 to 227.2 mg/100 g (**Table 3**). The total phenolic contents in peel of lemons and oranges were 190 mg/100 g and 179 mg/100 g, respectively (*16*). Thus, the total phenolic content of yuzu is higher than that of other citrus fruits. The amounts of vitamin C, total phenolics, hesperidin, and naringin were also affected by maturity. There was a strong positive relationship between the vitamin C content and the maturity in both peel and flesh

Table 2. Total Phenolics, Vitamin C, and Bioflavonoids Contents in Mature Yuzu Cultivars[a]

cultivar	yuzu component	total phenolics[b]	vitamin C		hesperidin		naringin	
			WM	DM	WM	DM	WM	DM
Wando	peel	294.35 a	107.08 a	631.72 a	96.24 a	568.81 a	98.1 a	578.79 a
	flesh	227.21 d	94.79 b	588.64 b	61.33 d	380.85 d	55.79 d	346.45 d
Goheung	peel	286.38 b	91.13 c	546.78 c	81.52 b	489.12 b	84.86 b	509.16 b
	flesh	206.32 f	60.66 e	370.02 e	53.90 f	328.79 f	48.70 f	297.07 e
Sadeung	peel	239.57 c	89.92 d	541.53 c	74.47 c	419.92 c	81.51 c	430.75 c
	flesh	213.73 e	88.10 d	528.6 f	59.12 e	354.72 e	54.53 e	327.18 f

[a] Mean values for all determinations based on $n = 20$. [b] Total phenolics content, expressed in mg of CAE per 100 g of fresh fruit. The data showed significant differences at the level of $p < 0.001$. WM, fresh matter; DM, dry weight.

Table 3. Changes in Total Antioxidant Activity and Phenolics, Bioflavonoids, and Vitamin C Contents of Wando Yuzus with Maturity[a]

yuzu component	maturity	total antioxidant activity[e]	total phenolics[b]	vitamin C (mg/100 g)[c]		bioflavonoids (mg/100 g)[d]			
						hesperidin		naringin	
				WM	DW	WM	DW	WM	DW
peel	green	246.9 ± 1.2 c	246.5 c	81 ± 0.5 c	477 ± 1.2 c	84 ± 0.5 b	494 ± 6.0 b	73 ± 6.8 b	430 ± 7.0 b
	yellow	253.7 ± 0.5 b	256.1 b	90 ± 1.3 bc	534 ± 2.6 b	85 ± 8.1 b	501 ± 9.0 b	75 ± 0.4 b	444 ± 8.6 b
	deep yellow	289.4 ± 0.8 a	294.4 a	107 ± 3.1 a	631 ± 0.3 a	86 ± 2.4 a	507 ± 4.2 a	88 ± 1.0 a	519 ± 2.4 a
flesh	green	206.4 ± 1.3 e	231.5 c	66 ± 1.9 d	409 ± 2.7 d	65 ± 0.9 c	403 ± 6.5 c	57 ± 0.9 c	353 ± 9.7 c
	yellow	210.2 ± 2.4 de	229.8 d	69 ± 2.4 d	428 ± 2.1 d	64 ± 1.1 c	383 ± 4.4 d	55 ± 4.6 c	341 ± 5.5 c
	deep yellow	237.9 ± 0.6 d	227.2 d	94 ± 3.7 a	583 ± 0.9 b	61 ± 3.3 c	378 ± 1.8 d	55 ± 7.9 c	341 ± 3.5 c

[a] Mean values of triplicate determinations. Values in the same row (i.e., total antioxidant activity and total phenolics) that are followed by a different roman letter are significantly different ($p < 0.01$) using ANOVA. Data are expressed as means ± SEM. [b] Total phenolics content, expressed in mg of CAE per 100 g of fresh fruits. [c] Data expressed in mg of vitamin C per 100 g of WM or DM. [d] Data expressed in mg of bioflavonoids per 100 g of WM or DM. [e] Total antioxidant activity is expressed in mg per 100 g fresh yuzus of VCEAC.

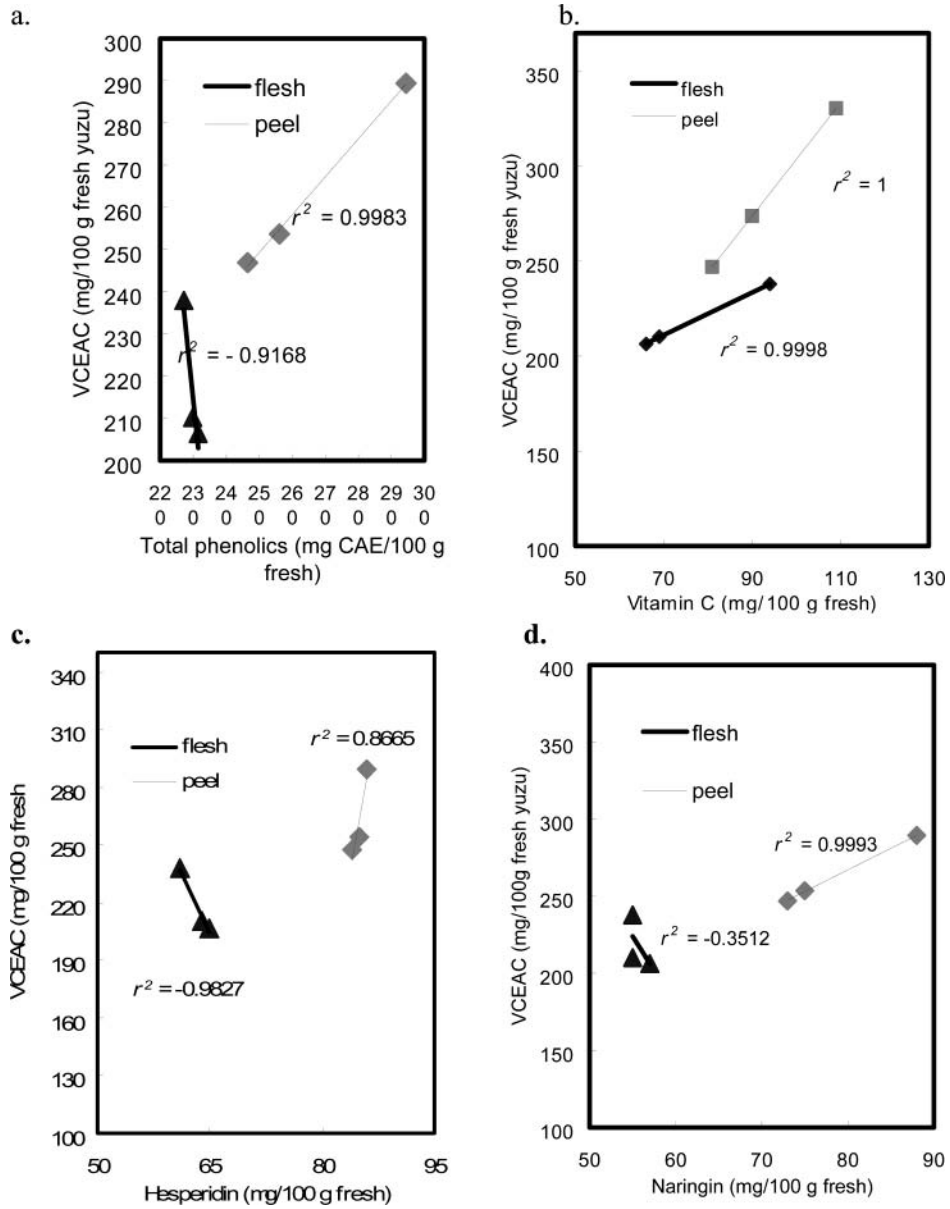

Figure 1. VCEAC values for each antioxidant factor. (**a**) Total phenolics contents, (**b**) vitamin C, (**c**) hesperidin, and (**d**) naringin.

Table 4. Contribution of Major Antioxidants to the Total Antioxidant Activity of Wando Yuzus[a]

antioxidant	concn of antioxidant (mg/100 g fresh yuzus)	EC_{50} values	relative VCEAC values	contribution to total antioxidant activity (mg VCEAC)
vitamin C	90.36	2.4	1	90.36
naringin	63.79	12.3	0.195	12.44
hesperidin	65.67	15.4	0.162	10.64

[a] Total antioxidant activity is expressed in mg per 100 g fresh yuzus of VCEAC.

(**Table 4**). However, the amounts of total phenolics, hesperidin, and naringin in peel increased with maturity, whereas those in flesh decreased with maturity. Similarly, the levels of naringin and hesperidin in immature citrus fruit flesh were higher than in the amounts of naringin and hesperidin in mature citrus fruit flesh (*44*, *45*). There was a strong positive relationship between the amount of vitamin C—but not of total phenolics and bioflavonoids—and the total antioxidant activity during maturation (**Figure 1**). These results suggest that the total content of vitamin C has a greater influence on the total antioxidant activity of yuzus than does total phenolics and bioflavonoids.

Investigations into the antioxidant capacity of food should consider the overall concentrations and compositions of diverse antioxidants, because the TAC of food is due to the combined activity of diverse antioxidants including phenolics, rather than being attributable to any particular phenolics. Wando yuzus contained higher levels of total vitamin C, phenolics, and bioflavonoids than did Sadeung and Goheung yuzus. Moreover, the peel contains more vitamin C and total phenolics than does the flesh in all of the tested yuzu cultivars. The contents of phenolics, bioflavonoids, and vitamin C in yuzus changed during ripening: The total antioxidant activity of the peel of Wando yuzus increased due to an increase in vitamin C content. However, there was an inverse relationship between the total antioxidant activity of yuzus and their phenolics content including hesperidin and naringin in flesh. Thus, the peel and flesh of ripe yuzus exhibited higher antioxidant activities than

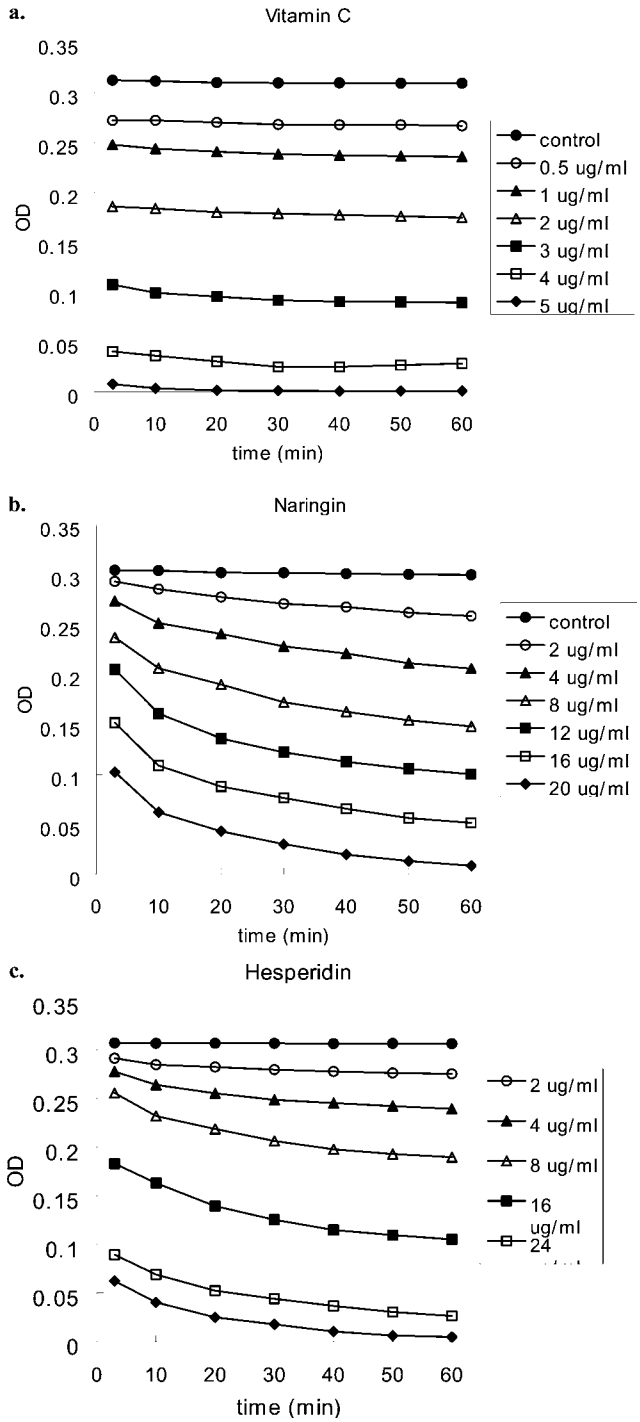

Figure 2. Kinetics of reactions of ABTS radicals with major antioxidants in yuzus. (**a**) Vitamin C, (**b**) naringin, and (**c**) hesperidin. Each antioxidant was reacted at different doses with 100 μM ABTS radicals.

unripe yuzu due to the content of vitamin C rather than that of phenolics (**Figure 1**). Therefore, to maximize the antioxidant properties, it may be desirable to harvest yuzus when they are ripe.

The scavenging rates of each antioxidant against ABTS radicals at different concentrations and reaction times are shown in **Figure 2**. The individual line plots of vitamin C, naringin, and hesperidin show the percentage decrease of ABTS radicals with the amount of tested compounds at different reaction times. There was a highly linear relationship between the concentrations of all antioxidants studied and the reduction in chromogen absorbance at 734 nm ($r^2 = 0.97$). A first-order highly linear

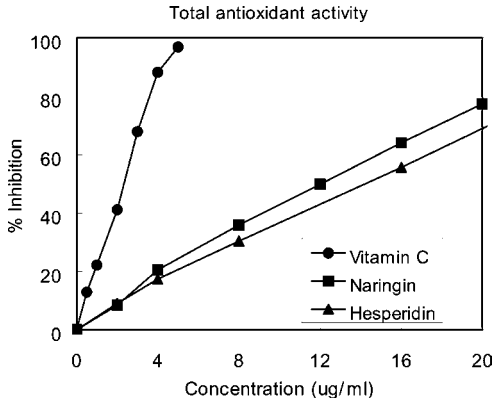

Figure 3. Total radical scavenging capacity of major antioxidants in yuzus. Values represent average percentage increases in the integrated area (eq 1) as compared to the control without antioxidants.

regression ($r^2 = 0.95$) was also obtained for the integrated area underneath each data point, which showed the time and dose dependency of each antioxidant at the same time as the antioxidant activity (**Figure 3**). The overall relative antioxidant capacity of yuzu compounds in VCEAC evaluated by the ABTS assay was in the following decreasing order: vitamin C > naringin > hesperidin. Normalizing the VCEAC value of vitamin C to 1, those of naringin and hesperidin were 0.195 and 0.162, respectively. The VCEAC value of these antioxidants evaluated by DPPH radical scavenging assay showed a linear dose−response relationship similar to that observed with the ABTS assay (data not shown). The average contents of vitamin C, naringin, and hesperidin in 100 g of fresh yuzus were 90.3, 63.7, and 65.6 mg, respectively. Therefore, the contributions of vitamin C, naringin, and hesperidin to the total antioxidant activity of the yuzus were 90.36, 12.44, and 10.64 mg VCEAC, respectively (**Table 4**); this indicates that the contribution of vitamin C to the TAC of yuzus is much greater than those of naringin and hesperidin.

Biomedical and epidemiological research suggests that antioxidant-rich fruits and vegetables play an important role in preventing disease (*1*, *2*, *11*). Antioxidants can inhibit reactive free radicals and, hence, can prevent the oxidation of other molecules and may have a health-promoting effect in the prevention of degenerative diseases including cancer. There have been many recent efforts to elucidate the potential health benefits of dietary phenolic phytochemicals, which exhibit stronger antioxidant activities than vitamin C. However, vitamin C is still an important bioactive constituent of fruits and vegetables. Recent reports suggest that the chemopreventive effects of vitamin C are linked to its protective effects against epigenetic mechanisms such as inflammation and the inhibition of GJIC as well as its antioxidant activities (*4*, *9*, *33*).

Gallic acid and epigallocatechin gallate (EGCG) are major antioxidants in tea, but they can also act as pro-oxidants (*33*, *34*). This pro-oxidant activity is thought to be directly proportional to the total number of hydroxyl groups, with the inclusion of multiple hydroxyl groups in gallic acid and EGCG, especially in the B ring, resulting in a significantly increased production of hydroxyl radicals in a Fenton system [see review (*36*)]. Some reports have suggested that high doses of gallic acid and EGCG induce cellular DNA damage (*34*−*37*). A recent study showed that the addition of gallic acid and EGCG to commonly used cell culture media leads to the generation of substantial amounts of H_2O_2, which may cause cellular DNA damage in humans (*38*). Furthermore, the antioxidant defense system needs many kinds of natural antioxidants to prevent diverse carcinogenic

processes. Therefore, the consumption of natural antioxidants through a balanced diet containing sufficient fruits and vegetables may be much more beneficial than supplementation of an individual antioxidant (*32*). Our results indicate that yuzus are a good source of antioxidants such as vitamin C and phenolic phytochemicals and may therefore provide health benefits to consumers.

ABBREVIATIONS USED

AAPH, 2,2′-azobis(2-amidino-propane)dihydrochloride; ABTS, 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid); CAE, chlorogenic acid equivalents; EGCG, epigallocatechin gallate; TAC, total antioxidant capacity; VCEAC, vitamin C equivalent antioxidant capacity.

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