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ARTICLE

Comparison of the Content of Bioactive Substances and the Inhibitory Effects against Rat Plasma Oxidation of Conventional and Organic Hot Peppers (*Capsicum annuum* L.)

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The aim of this study was to evaluate the chemical compositions and antioxidative activities of hot pepper fruits cultivated with strict management by organic and conventional agricultural practices. The ascorbic acid content in the organically grown hot pepper (OGP) was significantly higher than that of conventionally grown hot pepper (CGP) in both green and red fruits. The content of other bioactive compounds such as flavonoids (apigenin, luteolin, quercetin) and total phenolics in OGP was typically higher than in CGP regardless of fruit color. In addition, the $ABTS^{+}$ radical-scavenging activity of OGP red fruits was significantly higher than that of CGP red fruits. Moreover, regardless of the color of the fruits, a higher antioxidative activity was observed in blood plasma from rats administered the OGP fruit extracts than in blood plasma from rats administered the CGP fruit extracts. It was hypothesized that the higher antioxidant activity of the OGP fruits may have resulted from the higher antioxidant content in the OGP fruits. These results suggest that consumption of pepper fruits may increase antioxidant activity in the blood, and OGP fruits may be more effective in increasing this antioxidant activity than CGP fruits.

KEYWORDS: *Capsicum annuum*; hot pepper; organic agricultural products; bioactive compounds; antioxidant activity

INTRODUCTION

Organic foods are widely believed by the public to be healthier than the corresponding conventional foods (*1*−*3*). Therefore, the market for organic agricultural products has rapidly grown worldwide, which was as high as U.S. $46 billion in 2007 (*4*), and this demand has driven a similar increase in organically managed farmlands. Some studies have examined the content of nutrients (*3*, *5*), minerals (*3*), bioactive compounds (*6*−*11*), and antioxidative activities (*6*−*8*) in organic foods and reported that organic foods are superior to conventional foods. However, clear experimental evidence supporting these findings is still lacking, and therefore assessment of the nutritional and functional potentials of these products requires further research (*1*, *12*, *13*). Sustainable evaluation of the properties of conventional versus organic agricultural products requires highly systematic evaluation under identical conditions such as cultivation history, suitability of growing conditions, and methods used to evaluate the biological function and corresponding compounds of the food products. Among these parameters that must be controlled, the suitability of the cultivation conditions may be the most difficult because too many factors may affect growing and characterization of agricultural products (*14*, *15*). Therefore, it is thought that several additional case studies will be required for a clear comparison of the beneficial properties of organically versus conventionally grown plants (*1*, *16*). Recently, a few studies have very successfully compared the contents of bioactive compounds and antioxidative activities between organically and conventionally grown peppers (*8*, *17*). However, these studies did not focus on only peppers. Therefore, elevation of various other properties of only pepper fruits may provide an important case study to compare the quality of conventionally and organically cultivated plants. The hot pepper is a very important plant and is used as a vegetable, in spices, and as an external medicine worldwide. In particular, this fruit is widely used in Korean traditional fermented foods such as *kimchi* (*18*) and

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Table 1. Comparison of Soil Components of Conventionally (Conv) and Organically (Org) Grown Hot Peppers

				Ex[c] K	Ex Ca	Ex Mg		
	pH (1:5)	EC[a] (dS/m)	Av[b] P_2O_5 (mg/kg)		$cmol_c/kg$		NO_3-N (mg/kg)	NH_4-N (mg/kg)
org	6.8	2.50	1801	1.72	16.7	3.7	42	8.5
conv	6.9	3.41	1221	1.15	14.2	3.3	3	4.7

[a] EC, electrical conductivity. [b] Av, available. [c] Ex, exchangeable.

kochujang (*19*). In the present study, for a clearer comparison, organic and conventional peppers were simultaneously cultivated on neighboring farms, and the organically farmed peppers were cultivated under strict management at a farmhouse that was certified by a national certification institution to cultivate organic agricultural products. The constituent contents and antioxidant activities of conventionally and organically farmed pepper fruits were then compared.

MATERIALS AND METHODS

Plant Materials. Conventionally and organically farmed hot peppers (*Capsicum annuum* L. var. Nokgwang) were simultaneously cultivated on neighboring farms in Jangsa-ri (35.13° N, 128.08° E), Geumsan-myeon, Jinju City, Gyeongnam, Korea. Both red peppers were transplanted on October 2, 2007, and harvested on May 30, 2008. Organically farmed hot peppers were cultivated at a farmhouse that was certified by the National Agricultural Products Quality Management Service of Korea to cultivate organic agricultural products (Certified No. 17-03-1-1). The organic and conventional farming systems were managed in a plastic film house of silt loam soil. The experimental plots were arranged in a randomized block design at a size of 100 m^2 (10 m × 10 m), and three plots were replicated. To enhance soil health, the organic farming system was directly sowed with 0.2 Mg of rice seed ha^{-1} as a source of organic matter around July every year. The rice sprouts in the organic farming system were mixed with soil by rotating to a depth of 15 cm in early September before plowing prior to planting every year. The soil chemical properties for both the conventional and organic farming systems are shown in **Table 1**. Immediately after removal of the seeds from the hot pepper fruits, all samples were stored at −70 °C until analyzed.

Chemicals. Ascorbic acid, apigenin (4′,5,7-trihydroxyflavone), luteolin (3′,4′,5,7-tetrahydroxyflavone), and potassium persulfate were purchased from Wako Pure Chemicals Industries Ltd. (Osaka, Japan). Quercetin (3,3′,4′,5,7-pentahydroxyflavone) was obtained from Sigma-Aldrich Co. (St. Louis, MO). Folin−Ciocalteu's phenol reagent and gallic acid (3,4,5-trihydroxybenzoic acid) were purchased from Nacalai Tesque Inc. (Kyoto, Japan) and Acros Organics (Morristown, NJ), respectively. 2,2′-Azinobis(3-ethylbenzothiazoline-6-sulfonic acid ammonium salt) (ABTS) was obtained from Tokyo Chemical Industry Co., Ltd. (Tokyo, Japan).

Quantitative Analysis of Ascorbic Acid. The ascorbic acid content in hot peppers was measured by ODS-HPLC analysis (*20*). Finely cut fresh fruits (2 g) were mixed with 10% metaphosphoric acid (20 mL) at 4 °C. After homogenizing (BM-2 Nissei biomixer, Nihonseiki Co., Ltd., Osaka, Japan) for 1 min, the mixture was added to 100 mL of 5% metaphosphoric acid at 4 °C. An aliquot (5 mL) of the solution was centrifuged at 3000 rpm for 15 min, and the supernatant was filtered through a membrane filter (0.45 μm, Millex-FH 13MM, Millipore Co., Bedford, MA). The filtrate was analyzed by HPLC, which was equipped with an ODS-80Ts column (4.6 mm i.d. × 250 mm, 5 μm, Tosoh, Kyoto, Japan). The mobile phase consisted of 25 mM metaphosphoric acid (isocratic elution), and the flow rate was maintained at 1.0 mL/min. The sample was monitored at a wavelength of 254 nm using a photodiode array detector (PDA, SPD-M20A, Shimadzu, Kyoto, Japan). The experiments were conducted in quintuplicate. The ascorbic acid in each sample was quantified by the chromatographic peak area of an external standard. The calibration curve was plotted in the concentration range of 0.01−10 μg.

Quantitative Analysis of Flavonoids. The finely cut fresh hot pepper (2 g) was homogenized (BM-2 Nissei biomixer, Nihonseiki Ltd.) with MeOH (30 mL) and filtered (no. 2, Whatman International Ltd., Maidstone, U.K.). The extraction and filtering steps were repeated twice. The filtrates were combined and concentrated by vacuum evaporation at 38 °C. The concentrate was dissolved in 10 mL of MeOH. A portion (5 mL, fresh wt 1.0 g equiv) of the solution was added to 4.5 mL of distilled water and 0.5 mL of a concentrated H_2SO_4 solution. The mixture was allowed to react at 90 °C for 30 min to hydrolyze the flavonoid glycosides. After cooling, the reaction solution was evaporated until all MeOH was removed. Ten milliliters of distilled water was then added to the reaction solution. This solution was partitioned with EtOAc (10 mL, three times). The combined EtOAc layer was concentrated under vacuum. The concentrate was completely dissolved in 2 mL of MeOH. After filtering through a Millipore membrane (0.45 μm), an aliquot of the filtrate was injected into an HPLC system that was equipped with an ODS column (ODS-80Ts, 4.6 mm i.d. × 250 mm, Tosoh). The samples were monitored at a wavelength of 360 nm, and the flow rate was 1.0 mL/min. The sample was eluted using two solvents (35% MeOH containing 2% AcOH, solvent A; 70% MeOH, solvent B) with the following gradient: A/B (v/v) was increased to 0:100 over 30 min and then changed to 0:100 for 10 min. Each flavonoid aglycone was quantified using external standards (apigenin, 0.002−1 μg; luteolin, 0.002−1 μg; quercetin, 0.004−2 μg). The experiments were conducted in quintuplicate.

Determination of Total Phenolic Contents. The total phenolic content in each sample was measured using Folin−Ciocalteu's method (*21*). Briefly, each sample (fresh wt 2.0 g) was homogenized (BM-2 Nissei biomixer, Nihonseiki Ltd.) with MeOH (30 mL) for 5 min and filtered through filter paper (no. 2, Whatman). The residue was extracted with 80% MeOH (30 mL) and then filtered again (no. 2, Whatman). The remaining residue was washed with MeOH (30 mL). The combined filtrate was concentrated under vacuum. The concentrates were dissolved in 4 mL of MeOH. A portion (0.5 mL, fresh wt 0.25 g equiv) of the solution was evaporated with nitrogen gas and then the concentrate was dissolved in 0.5 mL of 30% MeOH. This solution was mixed with Folin-Ciocaltue's phenol reagent (2.5 mL) and a saturated Na_2CO_3 solution (0.5 mL). The mixture was incubated at room temperature for 60 min and then analyzed at a wavelength of 700 nm using a spectrophotometer (V-550, JASCO, Tokyo, Japan). The total phenolic content of the samples was quantified from a calibration curve of gallic acid, which was used as a standard compound. The experiments were conducted in quintuplicate.

Determination of $ABTS^+$ Radical-Scavenging Activity. $ABTS^+$ radical-scavenging activity was determined using the method described by Dudonne et al. (*22*) with slight modifications. Briefly, an $ABTS^+$ radical ($ABTS^{\bullet+}$) solution was prepared by reacting a 7 mM ABTS solution and 2.5 mM potassium persulfate (95:5, v/v) for 12 h at 4 °C in darkness. The $ABTS^+$ radical solution was diluted to an absorbance of approximately 0.7 ± 0.15 at 735 nm by the addition of ethanol. The MeOH extract (0.5 mL containing 15 mg in fresh wt equiv) was added to the diluted $ABTS^+$ radical solution (0.5 mL). After the mixture had been incubated for 30 min in darkness at room temperature, the absorbance (RS) was measured at 735 nm using a spectrophotometer. The $ABTS^+$ radical-scavenging activity of the sample was calculated as follows:

$$ABTS^+ \text{ radical-scavenging activity } (\%) = [(\text{control} - \text{RS})/\text{control}] \times 100$$

The experiments were conducted in quintuplicate.

Determination of the Inhibition Effect of the Hot Pepper MeOH Extracts against Copper Ion-Induced Oxidation of Rat Plasma. The antioxidative activity of samples was evaluated by measuring their inhibitory effects against cholesteryl ester hydroperoxide (CE-OOH) formation in copper ion-induced oxidation of diluted rat blood plasma (*23*). Sprague−Dawley rats (male, 6 weeks age, 180−200 g, Samtako Bio Korea, Osan, Korea) were kept at 22 ± 2 °C under a 12 h dark/light cycle and fasted for 12−15 h prior to blood collection. After anesthesia with diethyl ether, the abdomen wall was opened, and blood was collected into heparinized tubes from the abdominal aorta. Rat plasma was isolated by

centrifugation (1500*g*) at 4 °C for 20 min and used immediately for experiments or stored at −40 °C for no longer than 1 week. The plasma was diluted 4-fold with PBS (pH 7.4) and mixed with the MeOH extract of the hot pepper (fresh wt 50 μg equiv) in EtOH solution (final volume, 1%). The samples were then oxidized by the addition of 0.1 mL of $CuSO_4$ PBS solution (final concentration, 100 μM). The reaction mixture was incubated at 37 °C for 7 h with continuous shaking. The concentration of CE-OOH was determined using the method described by Arai et al. (*24*). Briefly, aliquots (100 μL) were withdrawn from the solutions and mixed with 3 mL of MeOH containing 2.5 mM 2,6-di-*tert*-butyl-4-methylphenol (BHT). The mixture was sonicated (Power Sonic 4200, Hwashin, Ulsan, Korea) for 1 min, and then neutral lipids were extracted with 3 mL of *n*-hexane by vortexing vigorously for 1 min. The upper layer (*n*-hexane) was collected, and extraction of the lower layer with 3 mL of *n*-hexane was repeated. The combined *n*-hexane phase was evaporated in a rotary evaporator at room temperature. The remaining lipids were dissolved in 100 μL of MeOH/$CHCl_3$ (95:5, v/v), and aliquots were subjected to CE-OOH analysis by RP-HPLC using a TSK-gel Octyl-80Ts column (Tosoh). The effluent was monitored by UV detection at 235 nm (Shimadzu SPD-10A, Shimadzu). The mobile phase consisted of MeOH/H_2O (97:3, v/v), and the flow rate was constant at 1.0 mL/min. The concentration of CE-OOH was calculated by comparison to a standard curve of cholesteryl linoleate hydroperoxide. Detailed procedures for the preparation of the cholesteryl linoleate hydroperoxide standard have been published elsewhere (*24*).

Copper Ion-Induced Oxidation of Rat Plasma after Oral Administration of Hot Pepper MeOH Extract. Male Sprague−Dawley rats (6 weeks of age; body weight, 180−200 g) were purchased from Samtako Bio Korea (Osan, Korea). Twenty-five rats were divided into five groups and housed in plastic cages at a temperature of 22 ± 2 °C with a 12 h light/dark cycle. Rats were fed a standard laboratory diet (Purina rodent chow, Cargill Agri Purina Inc., Seongnam, Korea) and water *ad libitum* for 3 days. The rats were fasted overnight (15 h), and then a propylene glycol solution (1 mL) of hot pepper MeOH extract (ca. 7 mg, fresh wt 14 g equiv) was administered orally. Five different samples were administered to the rats: (1) a control (propylene glycol solution, 1 mL); (2) a conventionally grown green pepper; (3) a conventionally grown red pepper; (4) an organically grown green pepper; and (5) an organically grown red pepper. After 1 h, whole blood was withdrawn from the abdominal aorta, and the blood plasma was obtained using the same methods described above. The plasma was pooled in the same volume from five rats of each group and then diluted 4-fold with PBS buffer (pH 7.4) and reacted with 100 μM (final concentration) $CuSO_4$ to induce CE-OOH formation. The reaction mixture was incubated at 37 °C with continuous shaking. The amount of CE-OOH induced by copper ions was measured at 30 min intervals for 12 h. The extraction and ODS-HPLC analysis of CE-OOH were performed using the same method described above.

Statistical Analysis. Results were expressed as mean ± standard deviation by SPSS (Statistical package for social sciences) 17.0 package programs. All data were measured by one-way ANOVA, followed by the Scheffe test. Significant differences were taken at $p < 0.05$.

RESULTS AND DISCUSSION

Ascorbic Acid Contents in Conventionally and Organically Grown Peppers. The ascorbic acid content in the green and red fruits of conventionally and organically grown peppers were quantitatively determined by ODS-HPLC analysis. Ascorbic acid was detected in the HPLC chromatogram at 6.2 min, which was in good agreement with the HPLC retention time (t_R) and UV−vis spectrum (data not shown) of the standard in HPLC-PDA system. The recovery of ascorbic acid was 92.7 ± 1.1%. As shown in **Figure 1**, the organically grown peppers (OGP, 239.5 ± 44.7 mg/100 g of fresh red fruit, 128.8 ± 22.6 mg/100 g of fresh green fruit) had a significantly higher ascorbic acid content than the conventionally grown peppers (CGP, 158.6 ± 24.1 mg/100 g of fresh red fruits, 77.6 ± 2.7 mg/100 g of fresh green fruit). In OGP, the ascorbic acid content of the red fruits (239.5 ± 44.7 mg/100 g of fresh fruit) was higher than that of the green pepper (128.8 ± 22.6 mg/100 g of fresh wt). The amount of ascorbic acid in red fruits was about twice that in green fruits, which was in agreement with ascorbic acid content reported in *Capsicum annuum* L. var. Mesilla (*25*).

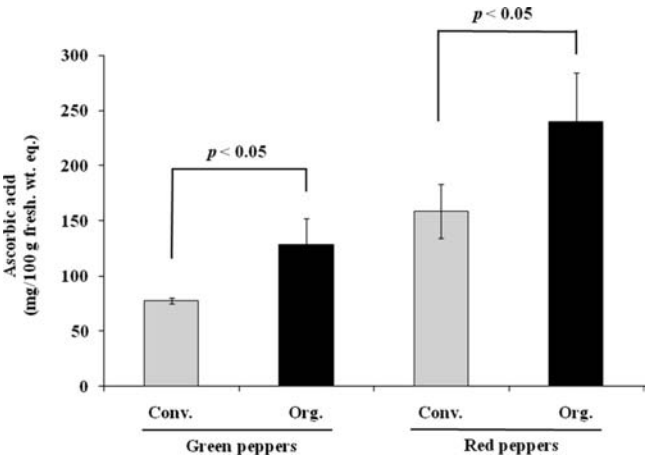

Figure 1. Comparison of the ascorbic acid content of conventionally (Conv.) and organically (Org.) grown hot pepper fruits. Values are means ± standard deviation, $n = 5$.

Flavonoid and Total Phenolic Contents in CGP and OGP. To determine the flavonoid content in CGP and OGP fruits, flavonoid aglycones obtained after acidic hydrolysis were quantitatively analyzed by ODS-HPLC analysis. Apigenin, luteolin, and quercetin have been reported to be the main constituents among the flavonoids contained in pepper fruits (*19*, *26*). They were detected at 6.1, 25.5, and 23.5 min, respectively, on the HPLC chromatogram (data not shown), which was confirmed by comparisons to the retention times and UV−vis spectra of each standard in the PDA system (data not shown). The recovery ratios of apigenin, luteolin, and quercetin were 95.8 ± 4.9, 90.5 ± 4.7, and 83.2 ± 2.5%, respectively. Luteolin was detected as the main constituent among the three flavonoid aglycones in the green and red pepper fruits (**Figure 2B**). The contents of apigenin, luteolin, and quercetin in green CGP fruits were higher than those in red CGP fruits (**Figure 2**). In addition, the contents of these compounds in the green and red OGP fruits were similar to those in the CGP fruits. These results are in agreement with previously studies that examined changes in the phenolic acid and flavonoid contents in hot pepper cultivars as a function of maturity (*27*). Therefore, it was suggested that the flavonoid content may decrease with the maturity of the hot pepper fruits regardless of farming practice. However, in some hot pepper cultivars, such as *C. annuum* var. Mesilla and *Capsicum frutescens* var. Tabasco, an increase in the luteolin, quercetin, and total flavonoid content with maturation has been reported (*28*). In addtion, fluctuations in flavonoid content with the maturation of peppers may be dependent on differences in cultivars, cultivation methods, and harvest time. Therefore, the flavonoid content in the CGP and OGP fruits may be also dependent on differences between these various factors. Nevertheless, in the present study, the contents of apigenin, luteolin, and quercetin in each green and red OGP fruit were similar to those in each green and red CGP fruits (**Figure 2**).

The total phenolic compound content in the MeOH extracts of CGP and OGP fruits was determined using Folin−Ciocalteu's method (*21*). In the green pepper fruits, the total phenolic compound content in OGP (34.20 ± 4.73 mg/100 g of fresh wt) was significantly higher than that in CGP (23.8 ± 3.1 mg/100 g of fresh wt), and the pattern of contents (CGP, 29.9 ± 2.0 mg/100 g of fresh wt; OGP, 46.7 ± 5.7 mg/100 g of fresh wt) in the red

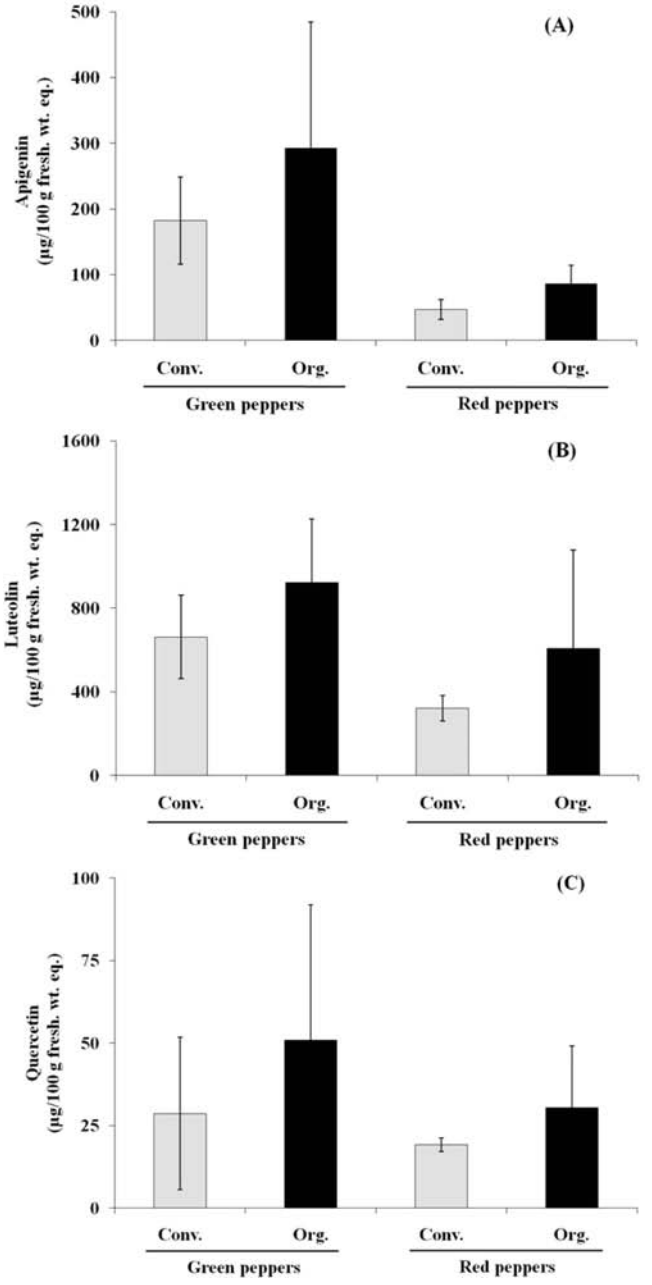

Figure 2. Comparison of the flavonoid (apigenin, luteolin, quercetin) content of conventionally (Conv.) and organically (Org.) grown hot pepper fruits. Values are means ± standard deviation, $n = 5$.

pepper fruits was similar ($p < 0.05$) to that in the green CGP and OGP fruits (**Figure 3**). The total phenolic compound content may reflect the flavonoid content as well as various other phenolic compounds, although the content of all phenolic compounds was not analyzed in this study. Several studies have reported that the flavonoid and total phenolic compound content in organic agricultural products was significantly higher than that in conventional agricultural products (*6*, *8*, *9*). Especially, a previous study on green peppers reported significantly higher levels of quercitrin and apigenin in the organic agricultural products relative to the content in conventional agricultural products (*8*). It is generally believed that organic agricultural products are released during cultivation due to environmental stresses such as insects, microorganisms, and other abiotic stress because the use of synthetic fertilizers and synthetic pesticides, plant growth regulator, etc. is severely limited (*29*). In addition, many studies have reported

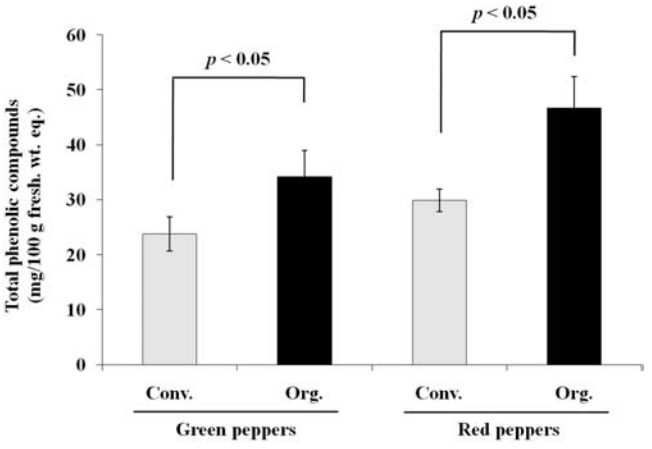

Figure 3. Comparison of the total phenolic content of conventionally (Conv.) and organically (Org.) grown hot pepper fruits. Values are means ± standard deviation, $n = 5$.

that an increase in stress response compounds such as volatiles, phenolics, and flavonoids was induced by biotic and abiotic stresses in various plants (*29*−*33*). However, in this study organically cultivated peppers appeared to have a higher amount of phenolic compounds containing flavonoids compared to conventionally cultivated pepper.

$ABTS^+$ Radical-Scavenging Activities of CGP and OGP. The antioxidant activity of each sample MeOH extract was evaluated using the $ABTS^+$ radical-scavenging assay (*22*). In the green pepper fruits, the $ABTS^+$ radical-scavenging activity did not differ significantly between the CGP (4.9 ± 1.5%) and OGP (6.9 ± 2.1%) fruits. A study (*17*) on the DPPH radical-scavenging activity of hot pepper fruit (*C. annuum* var. *annuum* L.) MeOH extract in relation to the maturity stage reported that the activity decreased with increased maturity. This decrease in activity may have occurred due to an interference in the absorption of light for the detection of DPPH by the pepper pigments. On the other hand, when Ren et al. (*8*) compared the antioxidative activity between conventionally and organically cultivated vegetables using microchemiluminescence, no significant differences in the antioxidative activity of green pepper (*C. annuum* L.) juices between conventionally and organically cultivated samples were observed, which was similar to the results observed this study (**Figure 4**). However, in the case of the red pepper fruits, the $ABTS^+$ radical-scavenging activity of OGP (43.6 ± 13.1%) was significantly higher than that of CGP (15.3 ± 2.9%). These results suggest that the antioxidative activity may increase with the maturity of the pepper fruits over a limited period and that this increase may be more obvious in OGP fruits than in CGP fruits.

Inhibitory Effect of CGP and OGP MeOH Extracts against Copper Ion-Induced Lipid Peroxidation of 4-fold Diluted Rat Plasma. Oxygen radicals, free radicals, and transition metal ions induce lipid peroxidation in human plasma lipoprotein, and this event has been shown to be an important aspect of the progress of various pathologic disorders such as atherosclerosis (*34*, *35*). Various antioxidants are present in human blood plasma, and therefore antioxidants have frequently been mentioned in connection with their physiological function in the cardiovascular system (*36*−*38*), because oxidative modification of plasma lipoprotein is strongly believed to participate in the initial event of atherosclerosis leading to coronary heart disease (*39*). It has been reported that CE-OOH is stable in human plasma when incubated at 37 °C (*36*), and CE-OOH is present in healthy human plasma at a concentration of ca. 3 nM (*40*). Therefore, CE-OOH was selected as an index of lipid peroxidation in blood plasma and

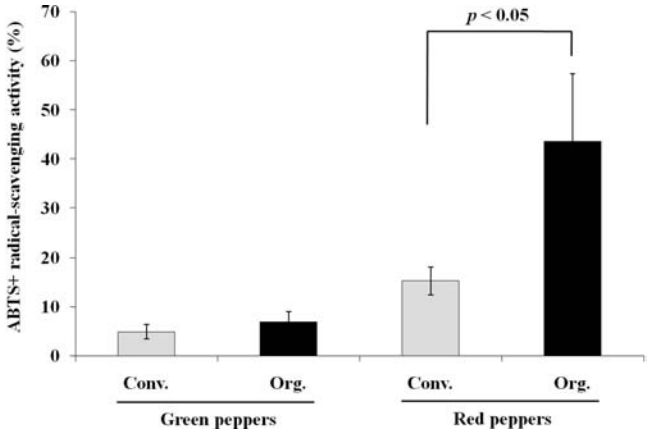

Figure 4. Comparison of $ABTS^+$ radical-scavenging activities of conventionally (Conv.) and organically (Org.) grown hot pepper fruit extracts. $ABTS^+$ radical solution (0.5 mL, absorbance = 0.7 ± 0.15 at 735 nm) was reacted with the MeOH extract (ca. 15 mg sample in fresh wt equiv/0.5 mL), and the mixture was then incubated for 30 min in darkness at room temperature. Values are means ± standard deviation, $n = 5$.

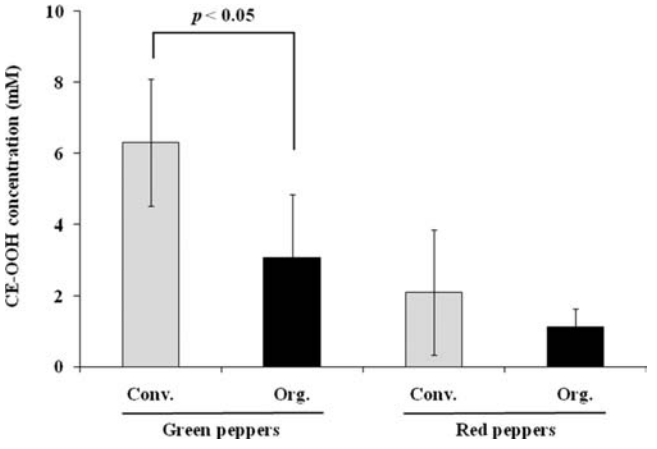

Figure 5. Comparison of the inhibition effects against CE-OOH formation in copper ion-induced rat plasma oxidation of conventionally (Conv.) and organically (Org.) grown hot pepper fruit extracts. 4-Fold dilutions of plasma with PBS (pH 7.4) were mixed with the MeOH extracts (fresh wt 50 μg equiv) of hot pepper fruits in an EtOH solution (final volume, 1%) and then oxidized through the addition of $CuSO_4$ (final concentration, 100 μM). The reaction mixture was incubated at 37 °C for 7 h with continuous shaking. Values are means ± standard deviation, $n = 5$.

was used to evaluate the antioxidant activity of CGP and OGP extracts in copper ion-induced rat blood plasma oxidation system by measuring the CE-OOH content. The amount of CE-OOH decreased in the following order: CGP green fruit (6.3 ± 1.8 mM) > OGP green fruit (3.1 ± 1.8 mM) > CGP red fruit (2.1 ± 1.8 mM) > OGP red fruit (1.1 ± 0.5 mM) (**Figure 5**). For both green and red fruits, the OGP extracts more effectively inhibited the formation of CE-OOH than the corresponding CGP extracts. However, the $ABTS^+$ radical-scavenging activity of the green pepper extracts was not significantly different between CGP and OGP fruits. This result suggests that the differences in the antioxidative activity may be related to the phenolic compound content due to the ability of phenolic compounds to chelate metal ions.

Comparison of Antioxidative Activity of Rat Plasma after Oral Administration of CGP and OGP MeOH Extracts. Blood plasma collected from five groups of rats ($n = 5$) that were administered

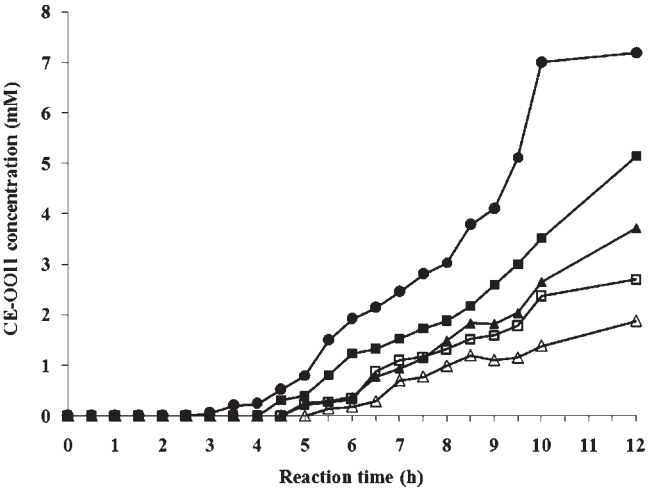

Figure 6. Comparison of the inhibition effects against CE-OOH formation in copper ion-induced oxidation of rat plasmas 1 h after oral administration of CGP and OGP fruit extracts: ●, control; ▲, green CGP; ▲, red CGP; □, green OGP; Δ, red OGP. The pepper fruit extracts (ca. 7 mg, fresh wt 14 g equiv) were orally administered as a propylene glycol solution (1 mL). The plasmas pooled in the same volume from rats ($n = 5$) of each group were diluted 4 times with PBS buffer (pH 7.4) and incubated with 100 μM (final concentration) $CuSO_4$ to induce CE-OOH formation. The reaction mixture was incubated in 37 °C with continuous shaking. The data are representative of two experiments.

the green and red CGP and OGP fruit extracts (fresh wt 14 g equiv) was pooled to the same volume from the five rats of each group. Oxidation of the 4-fold diluted plasma was initiated by adding copper ions (100 μM, final concentration), and the inhibition effect was evaluated by measuring the CE-OOH concentration (**Figure 6**). All groups administered the pepper fruit extracts more effectively suppressed the accumulation of CE-OOH than the control in regard to the lag times: control (3 h) < green CGP (4 h) < red CGP = green OGP (4.5 h) < red OGP (5 h). Moreover, the amounts of accumulated CE-OOH after 12 h of incubation were as follows: control (7.2 mM) > green CGP (5.1 mM) > red CGP (3.7 mM) > green OGP (2.7 mM) > red OGP (1.9 mM). In this experiment, the antioxidative activity in the blood plasma from rats administered the OGP fruit extracts was significantly higher than that of those administered the CGP fruit extracts regardless of the color of the fruit. It is believed that the higher antioxidant activity of the OGP fruits was related to the fact that it contained higher concentrations of various antioxidant compounds such as ascorbic acid, and phenolics containing flavonoids, relative to CGP fruits. These results suggest that consumption of pepper fruits may increase antioxidant activity in the blood, and OGP fruits may be more effective in increasing this activity than CGP fruits.

Perspectives. Organic agricultural products during cultivation are exposed to various environmental stresses because synthetic fertilizers, synthetic pesticides, etc. are not utilized (*29*). Therefore, organically farmed products may be significantly more undernourished due to the sterility of soil and stresses by insects and microorganisms compared to conventional agricultural practices. A number of studies have reported that an increase in stress response compounds such as volatiles, phenolics, and flavonoids is induced by biotic and abiotic stresses in various plants (*29*−*33*). The increase in these stress compounds is due to the strategy of plants to survive under various environmental stresses. In the present study, OGP fruits had significantly higher levels of most biological active compounds and higher antioxidative activities

than CGP. However, it is worth noting that the results of this study can be applied only to pepper fruits and may vary according to the methods use during cultivation and the environmental conditions of organic farming. Although an organically grown agricultural product was cultivated under the same soil conditions, numerous factors such as the types of green manure, treatment method and treatment time of green manure, environment, honesty of the farmer, etc., would affect the quality of the products. Previous results on the biological characteristics of an organically grown agricultural product have sometimes been generalized to all organically grown agricultural products. However, such generalization can confuse consumers as well as researchers. It may be too early to definitely state that organic food products are superior to conventional food products because the amount of given scientific data on organic farming is still small in comparison to the data obtained on conventional agriculture. In fact, synthetic pesticides and herbicides have only recently been shown to have negative health effects. Organically grown agricultural products are considered to be superior to conventional agricultural produce simply because they are not cultivated in the presence of chemicals; however, the situation is much more complicated than what is implied in this assumption. When we considered that many years of scientific study were needed to understand the process of conventional agriculture, it is apparent that much more scientific work will be needed to understand the parameters important to increasing the nutrition level, safety, and function of organically grown agricultural products.

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