Comparison of the phenolic composition and antioxidant activity of Korean black raspberry, Bokbunja, (Rubus coreanus Miquel) with those of six other berries

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ABSTRACT
Bokbunja (Rubus coreanus Miquel) and six other berries were analyzed for physico-chemical properties and antioxidant activities, using four different methods (DPPH·, ABTS−, FRAP, and reducing power). The total polyphenol, total flavonoid, and total anthocyanin contents of bokbunja were significantly higher than those of other berries, followed by mulberry, blackberry, cranberry, raspberry, wild strawberry, and strawberry. Antioxidant activity assays also showed that bokbunja’s antioxidant activity exceeded that of the other berries. Bokbunja had not only significantly higher levels of kaempferol, quercetin, and ellagic acid but also the highest ellagic acid content (408.57 mg kg−1 fresh weight). The potential antioxidant activities of bokbunja might be ascribed to its high total polyphenol, total flavonoid, and total anthocyanin contents. Ellagic acid may also affect antioxidant activity, although the concentrations of other phenolic compounds such as kaempferol, quercetin, and quercetin-3-O-rutinoside were high in other berries such as blackberry, cranberry, and mulberry.

INTRODUCTION

Bokbunja (Rubus coreanus Miquel), a Korean black raspberry, belongs to the Rosaceae family and is distributed in Southeast Asian countries, particularly Korea, Japan, and China. The general black raspberry (Rubus occidentalis) is native to midwestern and eastern North America (Lee, Dossett, & Finn, 2013). Bokbunja has been traditionally consumed and processed for traditional wine. Bokbunja has also been used in folk medicines for treatment of impotence, enuresis, spermatorrhea, asthma, and allergic disorders. Recently, based on the information of health beneficial effects of bokbunja, the growth area and the harvest quantity in South Korea have rapidly increased resulting in various bokbunja products, such as juices and concentrates, rice cakes, vinegar, and dietary supplements. Several studies have shown that bokbunja fruit and wine are positively related with various physiological functions, including antioxidative (Lee et al., 2012), anti-inflammatory (Lim, Hwang, & Shin, 2012; Yang et al., 2007), anticancer (Jung et al., 2009), anti-high cholesterol (Lee et al., 2015), and prevention of the memory impairment (Choi, Lee et al., 2012).

Berry fruits are rich sources of bioactive phytochemicals, the most prominent of which are phenolic compounds. Phenolic compounds, a diverse class of plant secondary metabolites, not only impart color and flavor to berries, but also beneficial effects against chronic diseases such as cardiovascular diseases, cancers, and certain degenerative diseases (Haminuik, Maciel, Plata-Oviedo, & Peralta, 2012; Zafra-Stone et al., 2007). The phenolic compounds of berries are classified into several structural and chemical classes such as phenolic acid (hydroxybenzoic acids and hydroxycinnamic acids), flavonoids (anthocyanins and flavonols), stilbenes (resveratrol), lignans, and tannins (hydrolyzed tannins and polymeric tannins).
condensed tannins) (Paredes-López, Cervantes-Ceja, Vigna-Pérez, & Hernández-Pérez, 2010). These components are, especially, associated with antioxidant activities. According to the antioxidant assays of raspberry and strawberry, they have about 10 times stronger antioxidant activity than apple and tomato (Beekwilder, Hall, & Vos, 2005). Therefore, they are excellent sources of dietary antioxidants. The diversity and concentration of their activities are highly dependent on the genetic factors, maturity stage, field condition, post-harvest management, and processing (Manganaris, Goulas, Vicente, & Terry, 2014). Commercially, the most common berries are from the genus Fragaria (strawberry), Rubus (blackberry, black raspberry, red raspberry, arctic raspberry), Sambucus (elderberry, red elderberry), and Vaccinium (blueberry, cranberry, bilberry).

In this study, we hypothesised that bokbunja may have higher levels of antioxidant compounds than other berries, such as blackberry, cranberry, mulberry, raspberry, strawberry, and wild raspberry. Hence, the objective of this study was to clarify the physicochemical characteristics of bokbunja and other six berries, and identify the concentrations of major phenolic compounds such as kaempferol, quercetin, quercetin-3-O-glucoside, and ellagic acid. Finally, the antioxidant activities of bokbunja were examined and compared to those of the other six berries using 2,2-diphenyl-1-picrylhydrazyl (DPPH·), 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid (ABTS·+), reducing power, and ferric reducing antioxidant power (FRAP) assays.

Materials and methods

**Chemicals and reagents and plant materials**

The chemicals Folin–Ciocăteu’s reagent, gallic acid, catechin, kaempferol, quercetin, quercetin-3-rutinoside, ellagic acid, DPPH·, ABTS·+, potassium persulphate, trichloroacetic acid (TCA), and potassium ferricyanide were purchased from Sigma-Aldrich Chemical Co. (St. Louis, Mo., U.S.A.). Analytical high-performance liquid chromatography (HPLC) grade solvents, methanol, water, acetic acid, and acetonitrile were purchased from Sigma-Aldrich and Fluka Biochemika (St. Louis, Mo., U.S.A.), respectively.

Samples of bokbunja (Gochang), mulberry (Daesungppong), raspberry (Mungduk), strawberry (Seolhyang), and wild raspberry (Kimhaewang) were picked from a farm located in Bundang (Gyeonggi-do, South Korea) in July 2015. And samples of blackberry (imported), cranberry (imported) were purchased from Bokbunja (Gochang), mulberry (Daesungppong), raspberry (Mungduk), strawberry (Seolhyang), and wild raspberry (Kimhaewang) were picked from a farm located in Bundang (Gyeonggi-do, South Korea) in July 2015. All samples were frozen and stored at −20°C until analysis.

**Determination of morphological characteristics and physicochemical properties**

Average fruit weight (g) was evaluated by Mettler PM460 DeltaRange Electronic Balance (Mettler, Greifensee, Switzerland), while a digital calliper (Traceable Digital Caliper 6”, WVR International, Milano, Italy) was used for measuring fruit size (mm). For each analysis, three replications were performed.

Moisture content was determined according to the method of Association of Official Analytical Chemists (AOAC, 2000). pH was determined by titrating 10 mL of pulp juice (in 100 mL final volume with Milli-Q water) using a pH meter (Model 320, Thermo Orion, Beverly, MA, U.S.A.). Total soluble solids (TSS, °Brix) were recorded with a digital refractometer N-1E (Atago, Tokyo, Japan). The UV/Vi absorbance spectra of each berry juice were determined over the range of 250–750 nm using a spectrophotometer (Epoch, Biotek, New York, U.S.A.). For the absorbance spectra measurements, raw berry was mixed using hand blender (Buwon, Korea), and centrifuged at 4000 rpm for 10 min at 4°C. After filtering using syringe filter (0.45 μm), the solution was used to the absorbance spectra measurement.

**Determination of the phenolic compound contents**

Total polyphenol content was determined according to the method described by Dewanto, Wu, Adam, and Liu (2002). A 1 mL sample was mixed with 1 mL of Folin–Ciocăteu reagent, followed by the addition of 5 mL of 14% sodium carbonate. The reaction was then allowed to proceed for 1 min. The absorbance at 750 nm was measured after 90 min at room temperature in dark. The results were expressed as milligram gallic acid equivalent (GAE) per kilogram fresh weight (mg GAE kg⁻¹ FW).

The total flavonoids content was analyzed according to the method described by Shen, Jin, Xiao, Lu, and Bao (2009). Two millilitres of deionized water and 0.6 mL of 50% sodium nitrate were mixed with 10 mL of sample, and then allowed to react at room temperature for 5 min. Then, 1.2 mL of 7.7% aluminium chloride hexahydrate was added and the samples were incubated for 5 min before the addition of 4 mL of 1 M sodium hydroxide. The absorbance was measured at 510 nm after 15 min incubation. The results were expressed as milligram (±)-catechin equivalent (CAE) per kilogram fresh weight (mg CAE kg⁻¹ FW). The total anthocyanin content was determined using a pH differential method (Association of Official Analytical Chemists (AOAC, 2006). The absorbance at 520 nm and 700 nm was measured and the absorbance at 700 nm was subtracted from the absorbance at 520 nm. The total anthocyanin content was calculated using the following equation:

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\text{Contents of anthocyanins (mg GAE kg⁻¹ FW)} = A \times MW \times 10^3 \times \text{dilution factor}(\epsilon \times 1) = A_{(A_{520} - A_{700})} \times \text{pH}1.0 - A_{(A_{520} - A_{700})} \times \text{pH}4.5, \quad \text{where} \ A \text{is absorbance,} \ \epsilon \text{is molar extinction coefficient for cyanidin-3-glucoside (26,900) and} \\
\text{MW is the molecular weight of cyanidin-3-glucoside (449.2 g mol⁻¹). The results were expressed as milligram cyaniding-3-glucoside equivalent per kilogram fresh weight (mg GAE kg⁻¹ FW).}
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HPLC analysis was performed according to Choi, Cho et al. (2012). The contents of kaempferol, quercetin, quercetin-3-rutinoside, and ellagic acid were analyzed by a HPLC system (Shimadzu, Tokyo, Japan). The Jasco HPLC system (Tokyo, Japan) comprised a bondapak C18 column (4 μm, 3.9 × 300 mm) coupled to a Jasco MD 2010 Plus photodiode array detector and double Jasco PU-980 pumps. Chromatograms were obtained from UV absorbance at 280 nm. The flow rate was maintained at 0.8 mL min⁻¹, the column temperature was set...
at 40°C; the mobile phase used was 2.0% acetic acid-containing water (solvent A) and 0.5% acetic acid-containing 50% acetonitrile (solvent B) were mixed in 100% solvent A in the beginning of the analysis, then after 70 min, the solution was mixed in 45% solvent A. To obtain specimens for analyses, the extracts were diluted with water to 1 mg mL\(^{-1}\), and filtered through a 0.45 μm syringe filter (Millipore, Billerica, U.S.A.). The contents of kaempferol, quercetin, quercetin-3-rutinoside, and ellagic acid were expressed as milligram per kilogram fresh weight (mg kg\(^{-1}\) FW).

**Determination of the antioxidant activity**

The DPPH\(^*\) free radical scavenging activity of bokbunja and other berries were measured using the method described by Gorinstein et al. (2004). A 0.2 mM solution of DPPH\(^*\) in methanol was prepared. One milliliter aliquot of the sample was added to 3 mL of this solution and kept in the dark for 30 min. The results were expressed as milligram Trolox equivalent (TE) per kilogram fresh weight (mg TE kg\(^{-1}\) FW).

The free radical scavenging activity of berries, based on the scavenging activity of the stable ABTS\(^+\) free radical, was also determined using the method described by Re et al. (1999) with slight modifications. The production of the radical cation was undertaken by preparing a stock solution of 7 mM ABTS\(^+\) in water. To this solution, ammonium persulfate (2.45 mM final concentration) was added and the solution was allowed to react for >16 h in the dark at room temperature to create a stable and dark blue-green radical solution. The solution was diluted with a phosphate buffered saline (PBS) solution to an absorbance of 0.70 ± 0.02 at 734 nm to prepare the working solution of the test reagent. A 0.03 mL of the diluted sample was added to 3 mL of the working solution. After incubating the solution at room temperature in the dark for 30 min, the absorbance was measured at 734 nm. The results were expressed as milligram TE per kilogram fresh weight (mg TE kg\(^{-1}\) FW).

The reducing power of the berries was determined as described previously (Oyaizu, 1986). A 1 mL aliquot of the sample was mixed with 2.5 mL of phosphate buffer (0.2 M, pH 6.6) and 2.5 mL of potassium ferricyanide (1%). The mixture was incubated at 50°C for 20 min, and then mixed with 2.5 mL of 10% TCA by vortexing. The mixture was centrifuged at 1000 g for 10 min, and then 2.5 mL aliquot of the supernatant was mixed with an equal amount of Milli Q water and 0.5 mL of 0.1% FeCl\(_3\). The absorbance was measured at 700 nm. The results were expressed as milligram TE per kilogram fresh weight (mg TE kg\(^{-1}\) FW).

The FRAP assay (Benzie & Strain, 1996) is based on the ability of phenolic compounds to reduce Fe\(^3+\) to Fe\(^2+\). To prepare the FRAP reagent, 0.1 M acetate buffer (pH 3.6), 10 mM 2,4,6-tripyridyl-s-triazine, and 20 mM ferric chloride (10:1:1, v/v/v) were mixed. Then, 0.2 mL of the previously diluted extract was added to 1.5 mL of the reagent. The absorbance was measured at 595 nm. The results were expressed as milligram TE per kilogram fresh weight (mg TE kg\(^{-1}\) FW).

**Statistical analysis**

Each determination was performed in triplicate, and results were expressed as mean ± standard deviation (S.D.) using SPSS version 11.5.0 (SPSS Statistics, IBM Corp., Version 20.0). Analysis of variance (ANOVA) and Duncan’s multiple-range test were used to determine the significance of the difference among samples with a significance level of 0.05.

**Results and discussion**

**The morphological characteristics and physicochemical property**

The morphological characteristics of bokbunja and other berries are given in Figure 1 and Table 1. The bokbunja fruit weight, length, and width were quite similar to the black berry. The physicochemical properties of bokbunja and other berries are shown in Table 2. The moisture content of bokbunja (81.44%) was similar to that of wild raspberry, whereas significantly lower than those of other berries, such as blackberry (86.13%), cranberry (86.03%), mulberry (86.00%), raspberry (85.84%), and strawberry (89.32%). The pH value of bokbunja (3.39) was similar to that of wild raspberry (3.39), whereas cranberry (2.36) had the lowest pH and mulberry (4.73) had the highest pH. The total soluble solid of bokbunja was 10.79 °Brix, which was higher than those of other raspberries, such as blackberry (9.44 °Brix), strawberry (9.86 °Brix), and cranberry (9.93 °Brix), and lower than those of raspberry (11.23 °Brix), mulberry (12.39 °Brix), and blackberry (13.05 °Brix).

**The phenolic compound contents in bokbunja and other six berries**

The total phenol, total flavonoid, and total anthocyanin contents of bokbunja and other berries are shown in Table 2. The total phenol contents of the berries ranged from 698.30 mg GAE kg\(^{-1}\) FW to 2911.35 mg GAE kg\(^{-1}\) FW. Bokbunja had the highest total phenolic content (2911.35 mg GAE kg\(^{-1}\) FW), followed by blackberry (2248.18 mg GAE kg\(^{-1}\) FW), mulberry

![Figure 1. Photo of the bokbunja (Rubus coreanus Miquel) and other six berries such as blackberry, cranberry, mulberry, raspberry, strawberry, and wild strawberry.](image)
Table 1. Morphological characteristics of the bokbunja (Rubus coreanus Miquel) and other six berries.

| Common name | Scientific name | Cultivated place | Fruit weight (g) | Fruit length (mm) | Fruit width (mm) |
|-------------|----------------|------------------|------------------|-------------------|-----------------|
| Blackberry  | Rubus fruticosus | U.S.A.            | 2.23 ± 0.10 cd   | 13.19 ± 0.29 b    | 15.95 ± 0.63 d   |
| Bokbunja    | Rubus coreanus   | Korea            | 2.44 ± 0.12 c    | 13.23 ± 0.24 b    | 16.02 ± 0.11 cd  |
| Cranberry   | Vaccinium macrocarpon | U.S.A. | 1.03 ± 0.05 e    | 13.16 ± 0.65 b    | 12.35 ± 0.29 f   |
| Mulberry    | Morus alba       | Korea            | 5.86 ± 1.21 b    | 31.46 ± 3.67 a    | 19.93 ± 1.68 b   |
| Raspberry   | Rubus idaeus      | Korea            | 2.02 ± 0.05 cd   | 13.56 ± 0.33 b    | 14.84 ± 0.32 e   |
| Strawberry  | Fragaria ananassa | Duch. Korea      | 12.63 ± 1.21 a   | 30.04 ± 2.59 a    | 29.92 ± 1.56 a   |
| Wild raspberry | Rubus crustatgillus Bunge | Korea | 1.63 ± 0.04 d    | 12.31 ± 0.47 b    | 16.82 ± 0.39 c   |

Values are mean ± SD; Values followed by different letters in the same column are significantly different by Duncan’s multiple range test (p < 0.05).

Los valores son medias ± desviación standard; valores seguidos de letras diferentes en la misma columna son significativamente diferentes según el test de rango multiple de Duncan (p < 0.05).

Table 2. Physicochemical properties of the bokbunja (Rubus coreanus Miquel) and other berries.

| Samples       | Moisture (%) | pH     | Total soluble solids (°Brix) | Total phenolics (mg GAE kg⁻¹ FW) | Total flavonoids (mg CAE kg⁻¹ FW) | Total anthocyanins (mg CGE kg⁻¹ FW) |
|---------------|--------------|--------|-------------------------------|----------------------------------|-----------------------------------|-------------------------------------|
| Blackberry    | 86.13 ± 0.12b| 3.49 ± 0.10bc| 13.05 ± 0.04a               | 2248.18 ± 118.14b                | 944.96 ± 43.35b                    | 827.06 ± 75.75c                      |
| Bokbunja      | 81.44 ± 0.16c| 3.39 ± 0.04b  | 10.79 ± 0.07d               | 2911.35 ± 243.62a                | 1273.88 ± 53.14a                   | 2742.35 ± 227.40a                    |
| Cranberry     | 86.03 ± 0.16b| 2.36 ± 0.00e  | 9.93 ± 0.05e                | 2178.32 ± 89.50b                 | 602.17 ± 19.32e                    | 465.90 ± 24.73d                      |
| Mulberry      | 86.00 ± 0.13b| 4.73 ± 0.11a  | 12.39 ± 0.07b               | 2074.45 ± 7.66b                  | 1267.88 ± 53.14a                   | 1115.67 ± 53.14d                     |
| Raspberry     | 85.84 ± 1.66b| 2.92 ± 0.03d  | 11.23 ± 0.25c               | 1420.75 ± 14.55c                 | 944.96 ± 43.35b                    | 343.20 ± 28.88d                      |
| Strawberry    | 89.32 ± 0.18a| 3.62 ± 0.04b  | 9.86 ± 0.02e                | 698.30 ± 29.94e                  | 193.87 ± 14.49e                    | 97.49 ± 8.98e                        |
| Wild raspberry | 81.44 ± 0.11c| 3.39 ± 0.11c  | 9.44 ± 0.02f                | 921.84 ± 18.20d                  | 225.60 ± 11.35e                    | 179.79 ± 8.20e                       |

Values are mean ± SD; Values followed by different letters in the same column are significantly different by Duncan’s multiple range test (p < 0.05).

Los valores son medias ± desviación standard; valores seguidos de letras diferentes en la misma columna son significativamente diferentes según el test de rango multiple de Duncan (p < 0.05).

(2178.32 mg GAE kg⁻¹ FW), cranberry (2074.45 mg GAE kg⁻¹ FW), raspberry (1420.75 mg GAE kg⁻¹ FW), wild raspberry (921.84 mg GAE kg⁻¹ FW), and then strawberry (698.30 mg GAE kg⁻¹ FW). The total phenolic content of blackberry, cranberry, and raspberry were found to be similar compared to those reported previously (Szajdel & Borowska, 2008), whereas strawberry was found to be lower. This may be due to the extraction methods, growing conditions, cultivars, and environmental condition. Bokbunja and mulberry contained the highest total flavonoids (1273.88 mg CAE kg⁻¹ FW and 1267.88 mg CAE kg⁻¹ FW, respectively), followed by blackberry (944.96 mg CAE kg⁻¹ FW), cranberry (602.17 mg CAE kg⁻¹ FW), raspberry (479.28 mg CAE kg⁻¹ FW), and wild raspberry (255.60 mg CAE kg⁻¹ FW). Strawberries had the lowest content of total flavonoid (193.87 mg CAE kg⁻¹ FW), which agreed with reported flavonoid content patterns by Mikulic-Petkovsek, Slatnar, Stampar, and Veberic (2012), which described 50 different flavonoids in 28 wild and cultivated berry species. Furthermore, bokbunja contained the highest anthocyanin content (2742.35 mg CGE kg⁻¹ FW), followed by mulberry (1115.67 mg CGE kg⁻¹ FW), blackberry (827.06 mg CGE kg⁻¹ FW), cranberry (465.90 mg CGE kg⁻¹ FW), raspberry (433.20 mg CGE kg⁻¹ FW), and wild raspberry (179.79 mg CGE kg⁻¹ FW). Strawberry also had the lowest content of anthocyanin (97.49 mg CGE 1 kg⁻¹ FW) consistent with the anthocyanins in related berries presented by other authors (Sariburun, Şahin, Demir, Türkben, & Uylaşer, 2010; Szajdel & Borowska, 2008). Anthocyanins, which have a typical flavonoid structure, are not only associated with water-soluble components but also responsible for the characteristic red to purple color of berries (Castañeda-Ovando, Pacheco-Hernández, Páez-Hernández, Rodríguez, & Galán-Vidal, 2009; Haminiuk et al., 2012).

Next, we focused on the impact of absorption differences on the bokbunja and other six berries using UV-visible spectroscopy. Typically, the UV-visible spectrum of anthocyanins shows its maximum band around 520 nm, related to the B-ring and C-ring (Halbwirth, 2010). In this result, bokbunja showed a maximum absorbance around 520 nm, followed by cranberry, raspberry, mulberry, blackberry, wild raspberry, and strawberry. These results were similar to the total anthocyanins patterns, except for mulberry and blackberry, which showed higher total anthocyanins contents than those of cranberry and raspberry (Table 2). According to Wrolstad, Durst, and Lee (2005), anthocyanins showed maximum absorbance in a solution with pH 1.0, whereas showed decreased absorbance in a solution with pH 4.5. Eiro and Heinonen (2002) showed that the absorbance of pelargonidin 3-glucoside with acid, such as ferulic acid or chlorogenic acid, gives a much narrower and higher spectrum than that of pelargonidin 3-glucoside without acid. In this study, cranberry and raspberry had pH 2.36 and pH 2.92, respectively, and blackberry and mulberry had pH 3.49 and pH 4.73, respectively. These pH conditions of the berries may be associated with the absorbance differences. Furthermore, bokbunja, cranberry, and mulberry showed additional shoulder around 350 nm, which is related to the glycoside form of flavonoids such as quercetin-3-rutinoside, quercetin-3-glucoside, and quercetin-3-arabinoside (Fecka, Kucharska, & Kowalczyk, 2015).

The contents and distribution of flavonoids and phenolic acids that show antioxidant activity vary in different kinds of berries. Kaempferol, quercetin with quercetin glycosides, and ellagic acid are the major phenolics, which are most concentrated within berries (Häkkinen et al., 1999; Iriwoharn & Wrolstad, 2004). Kaempferol, quercetin, quercetin-3-O-rutinoside, and ellagic acid contents from this study are shown in Figure 2.
Bokbunja had high levels of kaempferol, quercetin, and ellagic acid, while mulberry had the highest kaempferol and quercetin-3-O-rutinose contents. Blackberry had the highest of only kaempferol, whereas cranberry had the highest level of quercetin. According to Häkkinen et al. (1999), quercetin was the main flavonol in cranberry, whereas ellagic acid was the major phenolic compound in red raspberry and strawberry. In this study, the ellagic acid content was significantly higher in bokbunja (408.57 mg kg\(^{-1}\) FW), followed by mulberry (312.77 mg kg\(^{-1}\) FW), blackberry (282.50 mg kg\(^{-1}\) FW), cranberry (204.33 mg kg\(^{-1}\) FW), raspberry (181.00 mg kg\(^{-1}\) FW), wild raspberry (108.00 mg kg\(^{-1}\) FW), and strawberry (83.00 mg kg\(^{-1}\) FW). The ellagic acid contents of raspberry and strawberry was lower than that reported in the literature (Atkinson, Nestby, Ford, & Dodds, 2005; Choi & Kwak, 2014; Häkkinen, Kärenlampi, Mykkänen, Heinonen, & Törrönen, 2000; Wang & Zheng, 2005), but similar to the pattern where raspberry gad higher ellagic acid content than strawberry. Ellagic acid exist as a free form, a glycoside form, or linked ellagitannins with glucose which found mainly in foods (Abe, Lajolo, & Genovese, 2012; Koponen, Happonen, Mattila, & Törrönen, 2007; Vrhovsek, Giongo, Mattivi, & Viola, 2008). In this study, free ellagic acid content was detected

**Figure 2.** Absorption spectra of the bokbunja (*Rubus coreanus* Miquel) and other six berries.

**Figure 2.** Espectro de absorción de la bokbunja (*Rubus coreanus* Miquel) y otros seis frutos del bosque.

**Figure 3.** Ellagic acid, kaempferol, quercetin, and quercetin-3-O-rutinoside contents of the bokbunja (*Rubus coreanus* Miquel) and other six berries.

**Figure 3.** Contenido de ácido elágico, kaempferol, quercetina y quercetina-3-O-rutinósido de la bokbunja (*Rubus coreanus* Miquel) y otros seis frutos del bosque.

1\(^{\text{FW}}\): fresh weight.
without hydrolysis, that would be related lower content than that reported in the literature (Atkinson et al., 2005; Häkkinen et al., 2000; Wang & Zheng, 2005).

Antioxidant activity of bokbunja fruit and other berries

To evaluate the antioxidant activity of bokbunja and the other six berries, four types of antioxidant activity measurements such as DPPH radical scavenging activity, ABTS' radical scavenging activity, reducing power, and FRAP assays were performed because single assay is not sufficient to accurately assess antioxidant activity. These assays are characterized by their single electron transfer (SET) reactions (Huang, Ou, & Prior, 2005). Especially, DPPH' radical scavenging capacity and ABTS' radical scavenging activity assays are technically simple and easy to experimentally control (Macdonald-Wicks, Wood, & Garg, 2006). The DPPH' free radical scavenging activity of bokbunja was significantly higher than that of the other six berries in the following order: blackberry and mulberry, cranberry, raspberry, wild raspberry, and strawberry (Figure 4(a)). These results were similar to the DPPH' free radical scavenging effects. Our results are in general agreement with earlier studies (Ogawa et al., 2008; Pellegrini et al., 2003).

Reducing power and FRAP assays are also based on electron-transfer reaction, the reduction of ferric (Fe³⁺) to ferrous (Fe²⁺), which is accomplished in the presence of antioxidants. The FRAP assay requires acidic conditions (pH 3.6), whereas the reducing power assay requires at nearer neutral pH (Macdonald-Wicks et al., 2006). As shown in Figure 4(c), the reducing power of bokbunja was significantly higher than that of the other six berries, in the following order: mulberry, blackberry and cranberry, raspberry, wild raspberry, and strawberry. These results are similar to the DPPH', ABTS', and reducing power assays, and the highest value in the antioxidant activities of bokbunja may be due to the highest levels of total polyphenol, total flavonoids, and total anthocyanins contents. According to Fukumoto and Mazza (2000), the ellagic acid presented higher potential antioxidant activity, when compared to other phenolic compounds including...
benzoic acid derivatives, cinnamic acid derivatives, quercetin, and rutin. In this study, ellagic acid was significantly high especially in bokbunja, while the concentration of other phenolic compounds such as kaempferol, quercetin, and quercetin-3-O-rutinoside were high in other berries such as blackberry, cranberry, and mulberry, as were in bokbunja. Furthermore, antioxidant activities of bokbunja were significantly higher than those of other berries obtained from DPPH, ABTS\(^+\), reducing power, and FRAP methods.

Conclusions
In this study, the physicochemical properties and phenolic component contents with antioxidant activities of bokbunja and six other berries were analysed. Bokbunja had the highest total phenolic content (2911.35 mg GAE kg\(^{-1}\) FW), the highest total flavonoids (1293.88 mg CAE kg\(^{-1}\) FW), and the highest anthocyanin content (2742.35 mg CGE kg\(^{-1}\) FW). Furthermore, the four different antioxidant activity measurements also showed that bokbunja had the strongest antioxidant activities, followed by mulberry, blackberry, cranberry, raspberry, wild raspberry, and strawberry. Especially the ellagic acid content, which has been attributed to higher potential antioxidant activity compared to other 30 selected phenolic compounds (Fukimoto & Mazza, 2000), was the highest in bokbunja. Therefore, bokbunja may have valuable functional properties and could be very useful in developing bokbunja fortified food products in the food industry.

Disclosure statement
No potential conflict of interest was reported by the authors.

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