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Research Report

# Physicochemical Properties and Antioxidant Activity of Korean Cactus (*Opuntia humifusa*) Cladodes

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**Abstract.** Physicochemical properties and antioxidant activity of Korean native cactus (*Opuntia humifusa*) cladodes (KCC) were investigated to evaluate the possibility for its application in new health functional foods. The KCC showed a high amount of crude ash (201.2 g·kg$^{-1}$ DW), characterizing Ca (1,967.8 mg·100 g$^{-1}$ DW), Mg (1,411.2 mg·100 g$^{-1}$ DW), K (1,269.6 mg·100 g$^{-1}$ DW), and P (1,110 mg·100 g$^{-1}$ DW) as the major minerals, as well as a high total dietary fiber (503.3 g·kg$^{-1}$ DW) including soluble dietary fiber (233.2 g·kg$^{-1}$ DW). The water holding capacity (WHC), oil holding capacity (OHC), and swelling power of KCC were 20.6 g·g$^{-1}$, 2.6 g·g$^{-1}$, and 16.8 mL·g$^{-1}$, respectively. The viscosity of a 100 g·L$^{-1}$ KCC suspension showed a non-Newtonian flow behavior with shear thixotropy and time dependency. The ethyl acetate fraction from an 80% ethanol extract of KCC showed the highest antioxidant activity as well as the highest total phenolic (112.8 g·kg$^{-1}$ fraction) and flavonoid (89.7 g·kg$^{-1}$ fraction) contents compared to other fractions. The most abundant phenolic compound within the ethyl acetate fraction was taxifolin (13.97 g·kg$^{-1}$ fraction). The results suggest that KCC could be used as potential sources of Ca and dietary fiber as well as a natural antioxidant. It might be possible to manufacture functional food products using the unique physical properties of KCC.

***Additional key words*:** dietary fiber, phenolic compounds, physical properties

## Introduction

Interest in health care among consumers is increasing steadily and has expanded to dietary intake, and as a result, the food industry has started to produce new food types to reflect this change in consumerism. Indeed, industries are manufacturing processed food products from various agricultural raw materials with the aim to develop new functional food sources that serve as high value-added products for their end-use application (Saenz, 2000). Of these, plant-derived functional foods are receiving much attention. The *Opuntia* spp. cladodes are one of the best raw materials for manufacturing value-added food. Previous reports have shown that *Opuntia* spp. provide numerous benefits to improve health such as preventing cancer, diabetes, and cardiovascular disease and inhibiting viral infections, inflammation, and hypercholesterolemia because they contain high levels of nutrients such as betalanins, phenolic compounds, polysaccharides, vitamins, and minerals (Feugang et al., 2006; Stintzing and Carle, 2005).

The *Opuntia* spp. could grow well across the globe due to their high ecological adaptability. However, their commercial use in the food industry is still restricted to a few areas such as Africa, the Middle East, and South America (Stintzing and Carle, 2005). In Korea, interest in the one of *Opuntia* spp., known as the Korean native cactus cultivar (*Opuntia humifusa*), is increasing due to its nutritional and physiological value (Choi and Shin, 2011; Jung et al., 2011). However, there are few reports on its physical properties and antioxidant activity even though many studies on the physiological function of other *Opuntia* spp. have been reported (Choi and Shin, 2011; Feugang et al., 2006; Jung et al., 2011; Stintzing and Carle, 2005).

The mineral and dietary fiber contents of *Opuntia* spp. cladodes vary with species, maturity, and cultivation area climate. Regardless of differences between the species, it has been reported that *Opuntia* spp. cladodes contain higher calcium (Ca) and dietary fiber contents relative to vegetables, fruits, and nuts (Herández-Urbiola et al., 2011; Ramírez-Moreno

et al., 2011). The *Opuntia* spp. cladodes are also widely known for their strong viscous materials and hydrophilic polysaccharides of large molecular weight due to their great capacity to absorb and retain water (Stintzing and Carle, 2005; Trachtenberg and Mayer, 1981). The soluble dietary fiber and Ca contents of *Opuntia* spp. cladodes are considered to be very important elements for their physical properties because the main cell wall polysaccharide consists of low methoxyl pectin (Cárdenas et al., 2008). As a result, the *Opuntia* spp. cladodes exhibit very strong physical properties such as water holding capacity, swelling, and viscosity compared to other plants that are based on starchy polysaccharides and insoluble dietary fiber that exhibit weak physical properties in hydration treatments (Ayadi et al., 2009; Majdoub et al., 2010). Therefore, *Opuntia* spp. cladodes could be considered as a potential source of industrial hydrocolloids such as pectin and gums, which are used as food additives to modify the appearance, taste, texture, and stability of processed soups, breads, ice cream, and drinks (Dogan et al., 2002; Sáenz et al., 2004). Therefore, previous studies on the hydrophilic polysaccharides of *Opuntia* spp. cladodes have suggested it as a potential source for natural food additives such as thickening and gelling agents (Majdoub et al., 2010). Overall, it is important to consider *Opuntia* spp. as a source of a variety of important factors for addition into health functional foods (Herández- Urbiola et al., 2011; Ramírez-Moreno et al., 2004).

Therefore, the main objectives of this study were to investigate the physicochemical properties of *Opuntia humifusa* cladodes (KCC) grown in Korea and to examine the antioxidant activities of an 80% ethanol extract and its fractions to evaluate the possibility for the application of KCC in new health functional foods.

## Materials and Methods

### Plant Materials and Reagents

The Korean native cactus (*Opuntia humifusa*), which was harvested between November and December 2008, was purchased from a farm located in the surrounding area of Iksan, South Korea. The cladodes were removed manually from the cactus and freeze-dried after removing thorns and seeds. The freeze-dried cladodes were ground using a roll mill (single-type stainless roller, Shinpoong Eng. Ltd., Gwangju, Gyeonggi, Korea), sifted using a 100 mesh sieve, and then kept in a plastic container at 4°C until use.

Ethanol and water were used for HPLC analysis (J.T. Baker Chemical, NJ, USA). 2,2-Diphenyl-1-picrylhydrazyl (DPPH), Folin-Ciocalteau reagent, butylated hydroxytoluene (BHT), gallic acid, protocatechuic acid, chlorogenic acid, *p*-hydroxybenzoic acid, syringic acid, isovanillic acid, *p*-coumaric acid, ferulic acid, *trans-m*-coumaric acid, salicylic acid, *trans*-cinnamic acid and rutin, taxifolin, myricetin, luteolin, quercetin, kaempferol, and isorhamnetin were purchased from Sigma-Aldrich Co. (St. Louis, MO, USA), and other reagents were of analytical grade.

### Extraction and Preparation of Cladodes for Analysis

The freeze-dried cladodes (1 kg) were added to 10 L of ethanol/water (8/2, v/v) and stirred for 12 h at room temperature. After centrifuging at 10,000 × g for 15 min, the supernatant was filtered through filter paper (Whatman No. 4), and the filtrate was pooled followed by freeze-drying after evaporating at 40°C. Distilled water (1.2 L) was added to the freeze-dried 80% ethanol extract (120 g) and stirred for 6 h, then fractionated sequentially with hexane (1.2 L, 1 h), chloroform (1.2 L, 1 h), ethyl acetate (1.2 L, 1 h), and butanol (1.2 L, 1 h) three times. Each fraction was freeze-dried after centrifuging, filtering, and evaporating using the same conditions as above. The yields of 80% ethanol, hexane, chloroform, ethyl acetate, butanol, and water were 231 $g \cdot kg^{-1}$, 68 $g \cdot kg^{-1}$, 12 $g \cdot kg^{-1}$, 9 $g \cdot kg^{-1}$, 105 $g \cdot kg^{-1}$, and 749 $g \cdot kg^{-1}$, respectively.

The 80% ethanol extract and its fraction were then dissolved in ethanol/water (8/2, v/v) at a concentration of 0-2 $mg \cdot mL^{-1}$ to analyze DPPH radical scavenging ability, total phenolics, total flavonoids, and HPLC-DAD chromatography.

### Proximate Composition, Dietary Fiber, and Mineral Analysis

Moisture, crude lipid, crude protein, and crude ash content were analyzed according to AOAC official methods (1996). Dietary fiber content was determined using the method of Prosky et al. (1998). A dietary fiber assay kit (Sigma Chemical Co., St. Louis, MO, USA) was used to measure soluble dietary fiber (SDF), insoluble dietary fiber (IDF), and total dietary fiber (TDF) after enzymatic removal of starch and protein. Mineral content was measured according to the KFDA (Korea Food & Drug Administration) methods (2005). Each sample (0.1 g) was dispersed in 600 $mL \cdot L^{-1}$ nitric acid (10 mL) to wet the ash, followed by the addition of 600 $mL \cdot L^{-1}$ perchloric acid (1 mL). Then, distilled water was added to generate a total of 100 mL. Na, Mg, Zn, Cu, Fe, and Ca were measured using AAS (Solaar M5, Thermo Elemental Ltd., Cambridge, England), while P was measured by spectrophotometry (UV-1601, Shimadzu Co., Kyoto, Japan) at 650 nm.

### Physical Properties Analysis

Water holding capacity (WHC) was measured using the procedure described by Chen et al. (1998). Each sample (0.1 g) was placed in a 50 mL centrifuge tube to which 25 mL of

distilled water was added and mixed using a glass rod. They were stirred under gentle mechanical stirring for 20 min at 30°C. The supernatant was removed carefully, and the tube was inverted for 30 min on filter paper to remove residual distilled water. Moisture content of the precipitate was determined by drying at 105°C in a forced-air oven overnight.

Oil holding capacity (OHC) was determined similarly to WHC except that a commercial vegetable oil was used instead of water. Sample (0.1 g, $W_1$) and vegetable oil (25 mL) were placed in a 50 mL centrifuge tube and mixed using a glass rod. They were stirred under gentle mechanical stirring for 20 h at 30°C before centrifugation for 20 min at 15,000 × g. The supernatant was removed carefully, and the tube was inverted for 1 h on filter paper to remove residual oil. The residue ($W_2$) was weighed again, and the difference in weight between $W_1$ and $W_2$ was calculated as the OHC.

Swelling was measured using the procedure described by Robertson et al. (2000). Sample (1 g) was placed in a glass cylinder in which 20 mL of distilled water was added and remained standing for 20 h. Results are expressed as mL of swollen sample per g of dry sample.

Viscosity was measured using a Haake viscometer (VT550, Karlsruhe, Germany) with a plate-type rotor PQ1 (diameter 2.8 cm, gap 1 mm) and a temperature controller (DC 30, Karlsruhe, Germany). The applied shear rate was 100 $s^{-1}$ at 25°C for 60 s. Distilled water was added gradually to the cladodes powder to make a suspension (100 $g \cdot L^{-1}$) under gentle mechanical stirring at 25°C for 2 h. The suspensions were allowed to equilibrate at 25°C for 4 h before measurements were taken. Suspensions (50 $g \cdot L^{-1}$) of carboxylmethyl cellulose, xanthan gum, and pectin were used for comparison.

### Determination of Total Phenolic and Flavonoid Content

Total phenolic content was measured using the method of Dewanto et al. (2002). Folin-Ciocalteu's phenol reagent (0.2 mL) and 50 $g \cdot L^{-1}$ $Na_2CO_3$ (3 mL) were added to each sample (0.1 mL). The mixture was shaken and allowed to stand for 1 h in the dark at room temperature. Absorbance was measured at 725 nm using a spectrophotometer (UV-1650PC, Shimadzu Co., Kyoto, Japan). Gallic acid was used as a standard, and total phenolic content is expressed in gallic acid equivalents. Total flavonoid content was measured using the method of Jia et al. (1999). Sample (500 μL) was mixed with 600 $mL \cdot L^{-1}$ $NaNO_2$ (75 μL), then the mixture was shaken and allowed to stand for 5 min at room temperature. The mixture was then sequentially mixed with 100 $g \cdot L^{-1}$ $AlCl_3$ (150 μL), 1 $mol \cdot L^{-1}$ NaOH (500 μL), and distilled water (275 μL). Absorbance was measured at 510 nm using a spectrophotometer (UV-1650PC, Shimadzu Co., Kyoto, Japan). Catechin was used as a standard, and total flavonoid content is expressed in catechin equivalents.

### Antioxidant Activity Analysis

DPPH radical scavenging ability was measured using the method of Brand-Williams et al. (1995). Sample (0.2 mL) was mixed with 2.8 mL DPPH solution (60 $\mu mol \cdot L^{-1}$), and the mixture was allowed to stand for 30 min in the dark at room temperature. Absorbance was measured at 515 nm using a spectrophotometer, and BHT was used as a comparison. Scavenging ability was calculated using the following formula: scavenging ability (%) = [(Absorbance 515 nm of control – Absorbance 515 nm of sample) / Absorbance 515 nm of control] × 100.

### Analysis of Phenolic Compounds by HPLC-DAD Chromatography

Phenolic acid and flavonoid contents of the ethyl acetate fraction were measured using an HPLC analysis system (Sycam, Gilching, Germany) according to the methods described in Jin et al. (1999). A sunfire C18 column (25 cm × 4.6 mm, Waters Co., MA, USA) with a C18 guard column (2 cm × 4.6 mm) was used at 25°C. Separated phenolic acids and flavonoids were detected at two wavelengths (280 and 325 nm) with a photodiode array detector (PDA, S3210, Sycam, Gilching, Germany). Phenolic acids were detected at 280 nm, and flavonoids were detected at 325 nm. The mobile phase consisted of 10 $\mu L \cdot L^{-1}$ formic acid in 100 $mL \cdot L^{-1}$ acetonitrile (solvent A) and 10 $\mu L \cdot L^{-1}$ formic acid in 900 $mL \cdot L^{-1}$ acetonitrile (solvent B). Flow rate was kept at 1 $mL \cdot min^{-1}$ for a total running time of 45 min. The gradient program was as follows: 100% A at 0-2 min, 100% A to 90% A at 2-6 min, 90% A to 37% A at 6-31 min, 37% A to 50% A at 31-41 min, 50% A to 100% A at 41-45 min, 100% A at 45-50 min.

### Statistical Analysis

All data are expressed as the mean ± standard deviation (SD) for at least triplicate analyses of the same sample. Data were analyzed by ANOVA using the SAS statistical analysis system (SAS Institute Inc., Cary, NC, USA). Differences among samples were analyzed using Duncan's multiple range tests ($P < 0.05$).

## Results and Discussion

### Chemical Characteristics of Korean Cactus Cladodes (KCC)

The chemical composition and dietary fiber content of KCC are shown in Table 1. The KCC contained water (55.5 $g \cdot kg^{-1}$ DW), crude protein (47.3 $g \cdot kg^{-1}$ DW), crude fat (12.5 $g \cdot kg^{-1}$ DW), and crude ash (201.2 $g \cdot kg^{-1}$ DW). These values

**Table 1.** Proximate composition, minerals, and dietary fiber of Korean cactus cladodes.

| Component | | Content |
|---|---|---|
| Moisture (g·kg$^{-1}$ DW) | | 55.5 ± 2.4 B$^{z}$ |
| Crude protein (g·100g$^{-1}$ DW) | | 47.3 ± 1.0 C |
| Crude fat (g·100g$^{-1}$ DW) | | 12.5 ± 0.2 D |
| Crude ash (g·100g$^{-1}$ DW) | | 201.2 ± 3.1 A |
| Mineral (mg·100g$^{-1}$ DW) | Ca | 1,967.8 ± 5.8 A |
| | Mg | 1,411.2 ± 1.9 B |
| | K | 1,269.6 ± 7.4 C |
| | P | 1,110.0 ± 20.9 D |
| | Na | 282.8 ± 3.8 E |
| | Zn | 20.4 ± 0.0 F |
| | Fe | 16.8 ± 0.3 G |
| | Cu | 2.2 ± 0.0 H |
| Dietary fiber (g·kg$^{-1}$ DW) | Soluble dietary fiber | 233.2 ± 18.4 C |
| | Insoluble dietary fiber | 270.5 ± 16.3 B |
| | Total dietary fiber | 503.3 ± 34.6 A |

$^{z}$Values are mean ± SD (n = 3). Different upper case letters (A-H) in the same column are significantly different by Duncan's multiple test ($P < 0.05$).

**Table 2.** Physical properties of Korean cactus cladodes.

| Component | Content |
|---|---|
| Water holding capacity (g water/g DW) | 20.6 ± 0.4 A$^{z}$ |
| Oil holding capacity (g oil/g DW) | 2.6 ± 0.7 C |
| Swelling power (mL·g$^{-1}$ DW) | 16.8 ± 0.3 B |

$^{z}$Values are the mean ± SD (n = 3). Different upper case letters (A-C) in the same column are significantly different by Duncan's multiple test ($P < 0.05$).

are slightly different compared to the previous reports for KCC (Hahm et al., 2011; Jung et al., 2011; Lee et al., 1997). The difference in content might be attributed to the species, cultivation area, and part of cactus studied (Lee et al., 1997). Interestingly, we observed that crude ash was the most abundant component, which was the same as previous reports (Hahm et al., 2011; Jung et al., 2011).

The KCC was consisted of large amounts of minerals, with the primary elements being Ca (1,967.8 mg·100 g$^{-1}$ DW), Mg (1,411.2 mg·100 g$^{-1}$ DW), K (1,269.6 mg·100 g$^{-1}$ DW) and P (1,110 mg·100 g$^{-1}$ DW), and small amounts of other minerals (Na, Zn, Fe, Cu) were also detected. In the previous investigation by Yoon et al. (2009), similar findings were reported in which the most abundant mineral in the *Opuntia ficus indica* stem was Ca followed by Mg and P. In particular, the Ca content in KCC was found to be much higher than in other plants such as spinach (1,151.2 mg·100 g$^{-1}$ DW), lettuce (703.1 mg·100 g$^{-1}$ DW), cabbage (511.5 mg·100 g$^{-1}$ DW), and broccoli (439.3 mg·100 g$^{-1}$ DW) (USDA, 2012). The results point toward the fact that KCC could be used as a potential source of Ca for the elderly, children, perimenopausal women, and pregnant women (Ramírez-Moreno et al., 2011).

Soluble dietary fiber (SDF), insoluble dietary fiber (IDF), and total dietary fiber (TDF) contents of KCC were 233.2 g·kg$^{-1}$ DW, 270.5 g·kg$^{-1}$ DW, and 503.3 g·kg$^{-1}$ DW, respectively. The results showed higher SDF content than those of *Opuntia ficus indica* reported by previous researchers, in which the SDF, IDF and TDF contents of *Opuntia ficus indica* cladodes were 57-130 g·kg$^{-1}$ DW, 304-441 g·kg$^{-1}$ DW, and 418-512 g·kg$^{-1}$ DW, respectively (Ayadi et al., 2009; Ramírez-Moreno et al., 2011). In addition, Valente et al. (2010) reported that the TDF of *Opuntia monocanta* cladodes was 185 g·kg$^{-1}$ DW. Taken together, these results suggest that KCC could be a potential candidate to prevent chronic diseases such as obesity, diabetes, and gastrointestinal disorders due to its high dietary fiber content (Bach-Knudsen, 2001). On the other hand, KCC could also be considered a great source of natural Ca and TDF, suggesting a good source for manufacturing functional processed food products.

## Physical Characteristics of KCC

The physical properties of KCC are shown in Table 2. The water holding capacity (WHC) and swelling power of KCC were 20.6 g water/g DW and 16.8 mL·g$^{-1}$ DW, respectively,

**Table 3.** Total phenolics and flavonoids contents of 80% ethanol extract and fractions from Korean cactus cladodes [μg·mg$^{-1}$ extract (or fraction)].

| Extraction solvents | | Total phenolics[z] | Total flavonoids[y] |
|---|---|---|---|
| 80% Ethanol extract | | 23.0 ± 1.5 a | 23.5 ± 2.3 a |
| Fractions | Hexane | 46.4 ± 1.0 bC[x] | 56.3 ± 0.5 aB |
| | Chloroform | 14.0 ± 1.5 bD | 57.6 ± 4.7 aB |
| | Ethyl acetate | 112.8 ± 1.3 aA | 89.7 ± 3.7 bA |
| | Butanol | 54.0 ± 2.9 aB | 37.9 ± 1.5 bC |
| | Water | 15.3 ± 2.9 aD | 5.2 ± 0.1 bD |

[z]Total phenolic content is expressed as μg gallic acid equivalent per mg of extract (or fraction).
[y]Total flavonoid content is expressed as μg catechin equivalent per mg of extract (or fraction).
[x]Values are the mean ± SD (n = 3). Different lower case letters (a-b) in the same row are significantly different by Duncan's multiple test ($P < 0.05$). Different upper case letters (A-D) in the same columns are significantly different by Duncan's multiple test ($P < 0.05$).

**Table 4.** Parameters of calibaration curve for phenolic standards and contents for phenolic compounds of ethyl acetate fraction from Korean cactus cladodes (g·kg$^{-1}$ fraction).

| Phenolic compounds | | Standard | | | | | | Ethyl acetate fraction |
|---|---|---|---|---|---|---|---|---|
| | | No. | Retention time (min) | Linearity range (μg·mL$^{-1}$) | Correlation Coefficient ($R^2$) | LOD (μg·mL$^{-1}$) | LOQ (μg·mL$^{-1}$) | Content |
| Phenolic acids | Gallic acid | 1 | 4.52 | 0.2-4 | 0.9952 | 0.00081 | 0.00271 | 0.15 ± 0.05 FG[z] |
| | Protocatechuic acid | 2 | 6.73 | 0.2-4 | 0.9999 | 0.00104 | 0.00347 | 2.60 ± 0.00 B |
| | Chlorogenic acid | 3 | 7.91 | 0.2-4 | 0.9999 | 0.00095 | 0.00317 | 0.37 ± 0.02 E |
| | *p*-hydroxybenzoic acid | 4 | 9.10 | 0.2-4 | 1.0000 | 0.00590 | 0.01967 | 4.38 ± 0.23 A |
| | Syringic acid | 5 | 10.13 | 0.2-4 | 0.9999 | 0.00072 | 0.00239 | 0.66 ± 0.02 D |
| | Isovanilic acid | 6 | 10.39 | 0.2-4 | 0.9994 | 0.00094 | 0.00314 | 0.71 ± 0.14 D |
| | *p*-coumaric acid | 7 | 12.76 | 0.2-4 | 0.9949 | 0.00237 | 0.00790 | 0.61 ± 0.00 D |
| | Ferulic acid | 8 | 13.70 | 0.2-4 | 0.9989 | 0.00037 | 0.00123 | 2.22 ± 0.09 C |
| | *trans-m*-coumaric acid | 9 | 14.68 | 0.2-4 | 1.0000 | 0.00053 | 0.00178 | 0.20 ± 0.00 F |
| | Salicylic acid | 10 | 20.27 | 0.2-4 | 1.0000 | 0.00112 | 0.00375 | 0.22 ± 0.10 EF |
| | *trans*-cinamic acid | 11 | 21.01 | 0.2-4 | 0.9994 | 0.00022 | 0.00073 | 0.01 ± 0.00 G |
| | Total | | | | | | | 12.13 ± 0.07 |
| Flavonoids | Rutin | 12 | 11.00 | 0.2-4 | 0.9991 | 0.00208 | 0.00695 | 0.61 ± 0.02 E |
| | Taxifolin | 13 | 14.13 | 0.2-4 | 0.9998 | 0.00126 | 0.00419 | 13.97 ± 0.65 A |
| | Myricetin | 14 | 15.71 | 0.2-4 | 0.9985 | 0.00123 | 0.00409 | 1.35 ± 0.04 D |
| | Luteolin | 15 | 18.95 | 0.2-4 | 0.9990 | 0.00219 | 0.00729 | 0.24 ± 0.00 F |
| | Quercetin | 16 | 19.43 | 0.2-4 | 0.9990 | 0.00036 | 0.00119 | 1.89 ± 0.02 C |
| | Kampferol | 17 | 22.55 | 0.2-4 | 1.0000 | 0.00106 | 0.00353 | 0.22 ± 0.04 F |
| | Isorhamnetin | 18 | 22.85 | 0.2-4 | 1.0000 | 0.00098 | 0.00327 | 3.59 ± 0.02 B |
| | Total | | | | | | | 21.86 ± 0.64 |

[z]Values are the mean ± SD (n = 3). Different upper case letters in the same column are significantly different by Duncan's multiple test ($P < 0.05$).

resulting in significantly much higher values (11.1 g water/g DW and 7.4 mL·g$^{-1}$ DW, respectively) compared to previous studies on *Opuntia* spp. cladodes (Ayadi et al., 2009; Rosado and Díaz, 1995). It has been suggested that WHC and swelling power might possibly be affected by the amount of hydrophilic polysaccharide such as soluble dietary fiber, which is contained in the cell wall polysaccharides (CWP) of plants (Sáenz et al., 2004). Therefore, high amount of soluble dietary fiber in KCC could be attributed to the increase in WHC and swelling power through associating with large amounts of water (Cárdenas et al., 2008). According to Rosado and Díaz (1995), the high WHC of CWP in *Opuntia* spp. cladodes retards the absorption of glucose and cholesterol in the gastrointestinal tract. Such physiological effects of

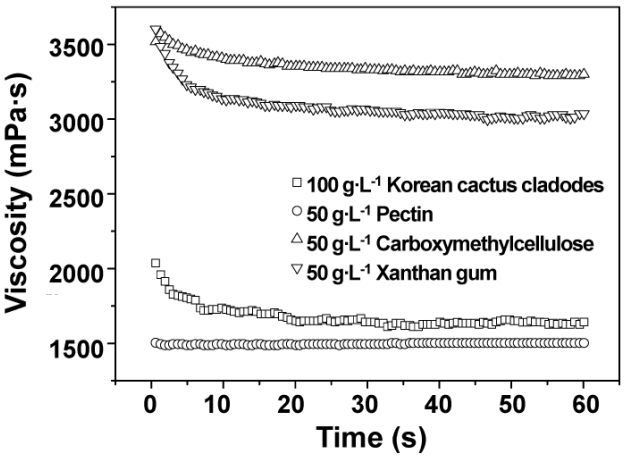


**Fig. 1.** Viscosity of 100 g·L$^{-1}$ Korean cactus cladodes suspension at 25°C and 100 s$^{-1}$.

CWP might result from high WHC and swelling as well as viscosity (Chau et al., 2004; Robertson et al., 2000). In fact, KCC, which has a higher value of SDF and WHC than those previously reported for *Opuntia* spp. cladodes, is expected to provide similar physiological properties.

The oil holding capacity (OHC) of KCC was 2.6 g oil/g DW, which was a higher value (1.3 g oil/g DW) than that previously reported for *Opuntia* spp. cladodes (Ramírez-Moreno et al., 2011). The OHC, representing the ability to associate with oil, could possibly be affected by both hydrophobic materials and IDF (Borderías et al., 2005). In particular, IDF was believed to be more effective. Therefore, the high oil absorption of KCC might be due to its high percentage of IDF. In addition, this value is also higher than those of previous reports for dietary fibers from agricultural byproducts obtained from apple, grapefruit, lemon, and orange (Figuerola et al., 2005). Furthermore, IDF prevents the absorption of fat in the body, resulting in beneficial health effects in the large intestine by increasing growth of lactic acid bacteria through production of short chain fatty acids and decrease in pH (Bach-Knudsen, 2001).

Viscosity of the KCC suspension was measured with a constant shear rate (100 s$^{-1}$) for 60 s, as shown in Fig. 1. The viscosities of 100 g·L$^{-1}$ KCC suspension, 50 g·L$^{-1}$ carboxylmethyl cellulose suspension, and 50 g·L$^{-1}$ xanthan gum suspension showed a non-Newtonian flow behavior that showed shear thixotropy and time dependency, while the 50 g·L$^{-1}$ pectin suspension showed a Newtonian flow behavior with a constant viscosity for 60 s. The apparent viscosity of 100 g·L$^{-1}$ KCC suspension after 60 s was 1,642.3 mPa·s, which was lower than that of the 50 g·L$^{-1}$ carboxylmethyl cellulose suspension (3300.0 mPa·s) and the 50 g·L$^{-1}$ xanthan gum suspension (3037.0 mPa·s), whereas it was higher than that of the 50 g·L$^{-1}$ pectin suspension (1503.0 mPa·s). The results showed a similar pattern with a previous report by Medina-Torres et al. (2000), who stated that the mucilage from *Opuntia ficus indica* cladodes showed a pseudoplastic flow behavior (shear-thinning, non-Newtonian flow behavior) as the shear stress decreased with increasing shear rate.

The KCC showed changed viscosity due to hydration below 60°C because of large amounts of pectin, which has the ability to form gel. The hydrophilic polysaccharides such as carboxylmethyl cellulose, xanthan gum, and pectin swell when they form hydrogen bonds with water. Because of this phenomenon, the viscosity is increased (Cárdenas et al., 2008). In addition, under the same shear rate and temperature, the viscosity could be increased due to higher hydrophilic polysaccharide concentrations, greater molecular weight, and more branched chains (Lopes da Silva and Rao, 1995). On the other hand, the viscous polysaccharides of cactus cladodes are composed of low methoxyl pectin (Cárdenas et al., 2008; Majdoub et al., 2010). Indeed, gel could be obtained by crosslinking through ion binding between the carboxyl groups of pectin and divalent cations such as Ca and Mg (Lopes da Silva and Rao, 1995).

Hydrophilic polysaccharides have been used in multipurpose commercial food additives such as bulking agents, gelling agents, emulsifiers, stabilizers, and thickeners because of their characteristic properties including high water holding capacity, great swelling power, and high viscosity at low temperature (Dogan et al., 2002). Thus, with these physical characteristics of the hydrophilic polysaccharides in KCC, it is important to consider exploiting KCC as a functional additive for improving the functional properties of foods.

## DPPH Radical Scavenging Ability and Phenolic Acid Composition

The 80% ethanol extract from KCC was analyzed to measure DPPH radical scavenging ability and total phenolic and flavonoid contents. The extract was sequentially fractionated depending on the polarity of the solvent with hexane, chloroform, ethyl acetate, butanol, and water. The total phenolic and flavonoid contents of all fractions are shown in Table 2. The 80% ethanol extract exhibited 23.0 g·kg$^{-1}$ extract of total phenolic content, resulting in higher content than that of previous report which showed 13.4 g·kg$^{-1}$ for total phenolic contents of 80% ethanolic extract of *Cheonnyuncho* (Kim et al., 2011). Among the fractions, the highest total phenolic content was observed in the ethyl acetate fraction (112.8 g·kg$^{-1}$ fraction). In regard of total flavonoid content, the 80% ethanol extract showed 23.5 g·kg$^{-1}$ extract. The ethyl acetate fraction showed the highest value (89.7 g·kg$^{-1}$ fraction), followed by the chloroform (57.6 g·kg$^{-1}$ fraction) and hexane (56.3 g·kg$^{-1}$ fraction) fractions.

The antioxidant activities in the 80% KCC ethanol extract

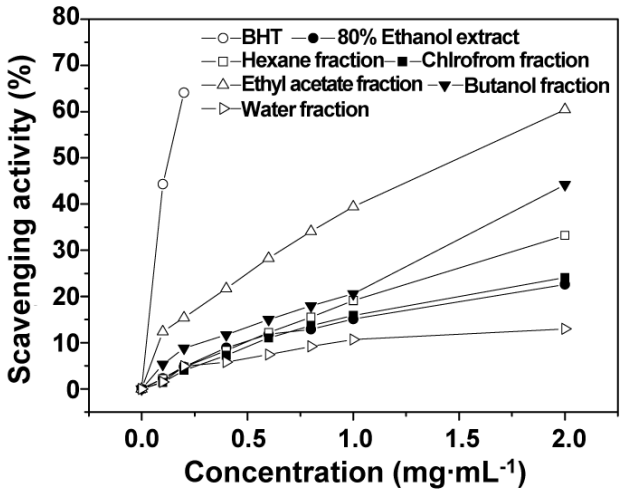


**Fig. 2.** Scavenging radical activity of 80% ethanol extract and fractions from Korean cactus cladodes. The concentration is expressed as mg·mL$^{-1}$ of mg extract (or fraction) per mL of 80% ethanol solvent.

as well as in its fractions were determined using the DPPH radical assay (Fig. 2). When the concentrations of 80% ethanol extract and its fractions were increased, their antioxidant activity showed a tendency to increase. Among the fractions used, the ethyl acetate fraction exhibited greatest activity for the DPPH radical assay compared to the rest of the other fractions. Based on our findings, we strongly suggest that the antioxidant activity of the ethyl acetate fraction might play an important role in stabilizing the reactive free radicals via the electron donor. DPPH radical scavenging ability of 80% KCC ethanol extract exhibited 15.1%, resulting in slightly lower than that of previous report which showed 19.2% for 80% ethanolic extract of *Cheonnyuncho* at the same concentration (1 mg·mL$^{-1}$) (Kim et al., 2011). In addition, DPPH radical scavenging ability of various solvent fractions showed a similar trend to previous report, in which DPPH radical scavenging ability of fractions from methanol extract of *Opuntia humifusa* stems showed 35.5% for butanol fraction, 15.4% for water fraction, and 7.2% for hexane fraction at the same concentration (1 mg·mL$^{-1}$) (Jung et al., 2012). Thus, the high amount of total phenolics and flavonoids might have an influence on KCC antioxidant activity. Several studies with *Opuntia* spp. have pointed out the correlation between phenolic compounds and antioxidant activity due to the hydroxyl groups that are contained in phenolic acids (Ayadi et al., 2009; Guevara-Figueroa et al., 2010). This phenomenon is important for improving human health due to the antiobesity, antidiabetes, anticancer, antimicrobial, and antioxidant properties (Feugang et al., 2006; Stintzing and Carle, 2005).

As discussed previously, phenolic compounds are important plant constituents for oxidant scavenging ability due to their hydroxyl groups. Moreover, it is suggested that polyphenolic compounds have beneficial health effects for humans. The quantitative spectrum of phenolic acids and flavonoids in the ethyl acetate fraction from KCC were detected. In general, 18 different types of phenolic compounds (11 phenolic acids and 7 flavonoids) were detected. The parameters of calibration curve for phenolic standards and contents of phenolic compounds in the ethyl acetate fraction from KCC are shown in Table 4.

The values of linearity ranges, limit of detection (LOD), and limit of quantization (LOQ) were evaluated using standards of phenolic acids and flavonoids. Linearity of each standard was evaluated within the range of 0.2-4 μg·mL$^{-1}$, resulting at least 0.99 of correlation coefficients ($R^2$). In regarding of LOD values for phenolic acids, it was ranged from 0.00022 to 0.00590 for *trans*-cinamic acid and *p*-hydroxybenzoic acid, respectively, whereas flavonoids from 0.00036 to 0.00219 for quercetin and luteolin, respectively. On the other hand, LOQ values were ranged from 0.00073 to 0.01967 μg·mL$^{-1}$ for phenolic acids and 0.00119-0.00729 μg·mL$^{-1}$ for flavnoids, respectively.

In terms of the ethyl acetate fraction from KCC, the major phenolic acids were *p*-hydroxybenzoic acid (4.38 g·kg$^{-1}$ fraction), protocatechuic acid (2.60 g·kg$^{-1}$ fraction), and ferulic acid (2.22 g·kg$^{-1}$ fraction), whereas the major flavonoids were taxifolin (13.97 g·kg$^{-1}$ fraction), isorhamnetin (3.59 g·kg$^{-1}$ fraction), quercetin (1.89 g·kg$^{-1}$ fraction), and myricetin (1.35 g·kg$^{-1}$ fraction). The results showed similar pattern with the previous reports, in which phenolic acids contained gallic acid, protocatechuic acid, *p*-hydrobenzoic acid, *p*-coumaric acid, ferulic acid, wheares flavonoids contained rutin, taxifolin, myricetin, quercetin, kampferol, and isorhamnetin in *Opuntia ficus indica* cladodes (Guevara-Figueroa et al., 2010; Stintzing and Carle, 2005). In addition, Lee and Lee (2010) identified toxifolin showed a similar antioxidant activity in BHA as a major flavonoid in ethyl acetate fraction of KCC.

In current study, we conclude that KCC exhibited unique physicochemical properties and high content of phenolic compounds with high antioxidant activities. Considering our findings, we propose that KCC could be used for manufacturing functional food products.

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