

Antioxidant compounds from four *Opuntia* cactus pear fruit varieties

Joseph O. Kuti*

Texas A&M University, Horticultural Crops & Food Research Laboratory, 214 Howe Agriculture Lab Annex, MSC 228, Avenue B, Kingsville, TX 78363-8202, USA

Received 21 January 2003; received in revised form 27 March 2003; accepted 27 March 2003

Abstract

The antioxidant compounds in extracts from four cactus (*Opuntia* species) fruit varieties were investigated. Conjugated flavonoids (quercetin, kaempferol and isorhamnetin), ascorbic acid, and carotenoids were isolated from the extracts. Quercetin was the most abundant in all varieties [*O. ficus-indica* (green-skinned), *O. lindheimeri* (purple-skinned), *O. streptacantha* (red-skinned), and *O. stricta* var. *stricta* (yellow-skinned)] examined. Kaempferol was found in green-skinned, purple-skinned and red-skinned varieties and isorhamnetin in green-skinned and purple-skinned varieties. The red-skinned fruits contained the most ascorbic acid (815 µg/g fw) and the yellow-skinned fruits the most carotenoids (23.7 µg/g fw). The antioxidant activity of the fruit extracts was stronger in the purple-skinned than other varieties. This result was consistent with total flavonoid content of the purple-skinned fruits. Thus, antioxidant capacity of cactus fruits may be attributed to their flavonoid, ascorbic acid and carotenoid contents. The data indicate that cactus fruits are a rich source of natural antioxidants for foods.

© 2003 Published by Elsevier Ltd.

Keywords: *Opuntia* cactus pear; Antioxidant; Flavonoids; Ascorbic acid; Carotenoids

1. Introduction

Cactus pear, produced by a perennial *Opuntia* cactus, belongs to the Cactaceae family and is well adapted to arid and semiarid climates, where water can be a limitation for cultivation (Benson, 1982). Many species of *Opuntia* cacti produce edible and highly flavoured fruit, known as “cactus pear” (Barbera, Carimi, & Inglese, 1992). Cactus pear fruit is a many-seeded berry with a thick peel, enclosing a delicately flavoured seedy pulp. The flavours of selected cactus pear fruit varieties resemble that of strawberry, watermelon, honeydew melon, fig, banana or citrus (Savio, 1987). Cactus pear fruits are a source of nutrients and vitamins (Sawaya, Khatchadourin, Safi, & Al-Muhammed, 1983; Teles, Stull, Brown, & Whitting, 1984) and are eaten fresh, dried or preserved in jams, syrups or processed into candy-like products (Hoffman, 1980). Their juices are sometimes fermented, using appropriate yeast strains, either into ethanol or wine and other beverages or used in food

flavourings and colourings (Bustos, 1981; Gurrieri, Micheli, Lanza, Tomaselli, Bonomo, & Rizzarelli, 2000; Retamal, Duran, & Fernandez, 1987; Saenz, 1996).

There is increasing evidence that fruits and vegetables may protect against numerous chronic diseases, including cancer, cardio- and cerebro-vascular, ocular, and neurological diseases (Block, Patterson, & Subhar, 1992; Ness & Powles, 1997; Steinmetz & Potter, 1996; Youdim & Joseph, 2001). The protective effect of fruit and vegetables has generally been attributed to their antioxidant constituents, including vitamin C (ascorbic acid), vitamin E (α -tocopherol), carotenoids, glutathione, flavonoids and phenolic acids, as well as other unidentified compounds (Sies & Stahl, 1995). Total antioxidant capacity of many fruits and vegetables has been determined by the oxygen radical absorbance capacity (ORAC) assay, which measures the ability of plant extracts to scavenge peroxy radicals (Cao, Sofic, & Prior, 1996; Wang, Cao, & Prior, 1996).

Polyphenolic flavonoids are metabolic products widely distributed in foods of plant origin and they have numerous biological and pharmacological properties (Cook & Samman, 1996; Hollman, Hertog, & Katan,

* Tel.: +1361-593-3978.

E-mail address: j-kuti@tamuk.edu (J.O. Kuti).

1996) that could potentially afford protection against chronic diseases. Food-derived flavonoids have been shown to possess antioxidant (Gil, Tomas-Barberan, Hess-Pierce, Holcroft, & Kader, 2000), anti-inflammatory (Middleton & Kandaswani, 1992), antimutagenic (Edenharder, Keller, Platt, & Unger, 2001; Namiki, 1990), and anticarcinogenic (Dragsted, Strube, & Larsen, 1993; Yoshida et al., 1990) properties. Flavonoids consist mainly of anthocyanins and anthocyanidins, flavonols, flavones, catechins, and flavanones (Herrman, 1988). Anthocyanins are water-soluble red, blue, and purple pigments in a large number of fruits, vegetables and flowers (Francis, 1989).

Many kinds of flavonoids have been reported in *Opuntia* cactus and types and content vary with variety, tissue type, and maturation (Wallace, 1986). Arcoleo, Ruccia, and Cusmando (1961) first reported the presence of isorhamnetin in flowers of *O. ficus indica*. Other reports have indicated that plants in the Cactaceae family produce flavonol 3-*O*-glycosides (quercetin, kaempferol, and isorhamnetin), dihydroflavonols, flavonones, and flavanonols (Burret, Lebreton, & Voirin, 1982; Meyer & McLaughlin, 1982; Miller & Bohn, 1982; Richardson, 1978; Rosler, Rosler, Mabry, & Kagan, 1966). Nearly all reports on flavonoids found in *Opuntia* cacti have dealt with extraction from the floral tissue (Clark & Parfitt, 1980; Shabbir & Zaman, 1968). Information on flavonoid composition and antioxidant activity of different cactus pear fruits and varieties is scarce. Recently, we reported the presence of phenolic compounds in fruit samples of cactus pears (Kuti, 2000) and Lee and Lim (2000) also reported a possible positive role of *Opuntia* cactus extract, as a natural antioxidant, in scavenging reactive oxidants. The purpose of the present study was to identify antioxidant constituents in cactus pear fruit extracts and to determine antioxidant capacities in different cactus pear fruit varieties using the ORAC assay.

2. Materials and methods

2.1. Fruit sample preparation

Fruits of four *Opuntia* species: *O. ficus-indica* (green-skinned), *O. streptacantha* (red-skinned), *O. stricta* var. *stricta* (yellow-skinned), and *O. lindheimeri* (purple-skinned) cactus pears (Table 1) were harvested from the Texas A&M University-Kingsville cactus pear orchard, when characteristic mature skin colours became manifest (Kuti, 1992). The fruit were hand-picked, separated into peel and pulp tissues, weighed, frozen and stored at -80°C .

2.2. Chemicals

R-phycoerythrin (R-PE) from *Porphyidium cruentum*, 2,3-dihydroxybutane-1,4-dithiol (DTE), L-ascorbic acid

reducing agent 2,3-dihydroxybutane-1,4-dithiol (DTE), β -carotene, and carotenoid standards were purchased from Sigma (St. Louis, MO, USA). 2',2'-Azobis(2-amidinopropane) dihydrochloride (AAPH) was purchased from Wako Chemicals USA Inc. (Richmond, VA, USA). 6-Hydroxy-2,5,7,8-tetramethylchrman-2-carboxylic acid (Trolox) and N, N dimethylformamide (DMF) were obtained from Aldrich (Milwaukee, WI, USA). Quercetin, kaempferol and isorhamnetin standards were obtained from Atomergic Chemetals Corp. (Farmingdale, NJ, USA). Polyphenol (anthocyanin monoglucoside—"kuramanin") was obtained from Polyphenols Laboratories (Sandnes, Norway). All chemicals used in the experiments were reagent- or HPLC-grade and were purchased from VWR (West Chester, PA, USA).

2.3. Extraction and analysis of flavonoids in cactus pear fruits

Flavonoid glycoside extraction and analysis were similar to the methods described by Merken and Beecher (2000), with a slight modification in flavonoid hydrolysis. Flavonoid aglycones were quantitatively analyzed, after acid hydrolysis of flavonoid conjugates, in triplicate, by HPLC using the external standard method. A Water photodiode array detector (Model 996) was used to record UV spectra of flavonols (quercetin, kaempferol and isorhamnetin). Flavonol identifications were based on comparisons to TLC R_f values and HPLC retention times with quercetin, kaempferol and isorhamnetin standards. The peel and pulp samples were analyzed by homogenizing 2 g each in 25 ml of methanol, filtering through miracloth, washing with 50% methanol and adjusting to 60 ml. Flavonol glycosides extracted from the macerated cactus pear tissues with MeOH by a procedure that gave 98% recovery in experiments with spiked samples were hydrolyzed with 1 N hydrochloric acid (HCl) in 50% methanol (MeOH) at 90°C for 30 min. Flavonoid aglycones were quantified at 370 nm using a C_{18} column with a solvent system of MeOH: water (35:65), pH 2.4 with phosphoric acid at 1 ml/min.

2.4. Extraction and determination of ascorbic acid content of cactus pear fruits

For each fruit sample, 5 g of frozen tissues were powdered using a dismembrator. Extraction was carried out in potassium phosphate buffer for ascorbic acid. The homogenate was centrifuged to remove the debris and the supernatant diluted and filtered through Milipore filters of pore-size $0.45\ \mu\text{m}$. The detection of ascorbic acid (AA) by HPLC is based on UV absorption of the reduced form of AA at 264 nm. Since AA is an easily oxidizable compound, it is necessary to add the

Table 1
Fruit characteristics of cactus pear from different *Opuntia* species used in this study

<i>Opuntia</i> spp.	Colour ^a			Peel thickness (cm)	Edible portion (%)
	Peel	Pulp	Juice		
<i>O. ficus-indica</i>	g	l-g	l-g	0.65	58
<i>O. lindheimeri</i>	p	r-p	p	0.30	50
<i>O. streptacantha</i>	r	g-r	r	0.45	60
<i>O. stricta</i> v. <i>stricta</i>	y	y-o	o	0.55	58

^a Different colours of the cactus pear tissues: g = green; l-g = light green; p = purple; r-p = reddish-purple; r = red; g-r = greenish red; y = yellow; y-o = yellowish orange.

reducing agent 2,3-dihydroxybutane-1,4-dithiol (DTE) during preparation. Two aliquots from each homogenized fruit tissue samples were measured, one sample containing ascorbic acid oxidase (20 U/ml dissolved in 50 mM potassium phosphate buffer, pH 6.0), and the other containing no enzyme to make sure that no contaminating compounds were included. Average recovery rates of 89% were obtained and the measured values were therefore corrected for a loss of 11%. A Waters HPLC system equipped with a diode array detector (Model 990) was used. Extracts were chromatographed on a Spherisorb ODS-column with a potassium phosphate buffer as eluent. Samples (5 ml) were injected at a flow rate of 1.0 ml/min. The retention time was 2.26 min and the total development time was 5 min. Ascorbic acid concentration were calculated as $\mu\text{g/g}$ fresh-frozen weight.

2.5. Extraction and determination of carotenoid contents of cactus pear fruits

Approximately 2 g of cactus pear tissue were homogenized with a Waring blender and extracted with 10 ml of hexane/acetone/ethanol (50:25:25, v/v) before being centrifuged for 5 min (IEC, Needham, MA, USA) at 6500 rpm at 5 °C. The top layer of hexane, containing the colour, was recovered and transferred to a 25-ml volumetric flask. The volume of recovered hexane was then adjusted to 25 ml with hexane. Total carotenoid determination was carried out on an aliquot of hexane extract by measuring absorbance at 450 nm in a Genesis-5 Spectronic spectrophotometer (Rochester, NY). Total carotenoids were calculated using an extinction coefficient of β -carotene, $E^{1\%} = 2505$ (Rodriguez-Amaya, 1999).

2.6. Antioxidant capacity of cactus pear fruit constituents

Antioxidant capacity was determined by oxygen radical absorbance capacity (ORAC) assay following procedures previously described by (Cao, Alessio, & Cutler, 1993). This assay measures the ability of antioxidant components in test materials to inhibit the decline in

R-PE fluorescence that is induced by a peroxy radical generator, AAPH. The reaction mixture contained 1.7 ml of 75 mM phosphate buffer (pH 7.0), 100 μl of R-PE (3.4 mg/l), 100 μl of 320 mM AAPH, and 100 μl of sample. Phosphate buffer was used as a blank and Trolox (a water-soluble α -tocopherol analogue) as a standard during each run. The final volume of 2 ml was used in a 10 mm wide fluorometer cuvette. R-PE, phosphate buffer, and samples were pre-incubated at 37 °C for 15 min. The reaction was started by addition of AAPH. Fluorescence was measured and recorded every 5 min at emission of 570 nm and excitation of 540 nm using a Sequoia-Turner Model 450 fluorometer (Englewood, NJ) until the fluorescence of the last reading declined to <5% of the first reading. One blank, one standard, and a maximum of 10 samples were analyzed at the same time. Each sample was repeated at least three times. The ORAC value refers to the net protection area under the quenching curve of R-PE in the presence of an antioxidant. The final results (ORAC value) were calculated and expressed using Trolox equivalents per gramme of fresh-frozen weight.

2.7. Statistical analysis

All data presented were means of three replicates (i.e. 5 fruits each) along with standard errors of means. Correlation coefficients at $P \leq 0.05$ and $P \leq 0.01$ levels between ORAC and flavonoid constituents were performed using Microsoft Excel Data Analysis. Data were further subjected to analysis of variance, and means were compared using Waller and Duncan's Bayes least significance difference (LSD) test. Differences at $P < 0.05$ were considered significant.

3. Results and discussion

3.1. Flavonoid content of cactus pear fruit extracts

Flavonoid contents in the four cactus pear fruit varieties are shown in Table 2. Using extraction and HPLC analysis, as by Merken and Beecher (2000), it was revealed that the predominant flavonoids in the fruits of

Table 2
Flavonoids (flavonols) distribution in fruit tissues of different *Opuntia* cactus pears

<i>Opuntia</i> ^a species	Flavonol content ^b (µg/g fresh weight)		
	Kaempferol	Quercetin	Isorhamnetin
<i>O. ficus-indica</i>	2.2±0.3	43.2±2.5	24.1±1.0
<i>O. lindheimeri</i>	1.1±0.4	90.5±11.5	1.9±0.5
<i>O. streptacantha</i>	3.8±0.5	51.0±4.6	ND ^c
<i>O. stricta var. stricta</i>	ND	9.8±3.0	ND

^a *Opuntia ficus-indica* (green-skinned cactus pear); *Opuntia lindheimeri* (purple-skinned cactus pear); *Opuntia streptacantha* (red-skinned cactus pear); *Opuntia stricta var. stricta* (yellow-skinned cactus pear).

^b Mean for triplicate determination ±SEM.

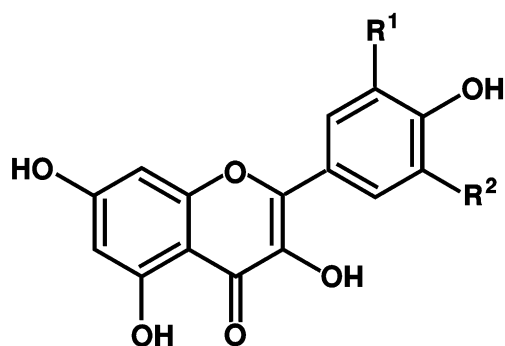
^c ND = not detectable.

Opuntia cactus pears consisted of quercetin, kaempferol and isorhamnetin, respectively (Fig. 1). Individual flavonoids varied greatly among the cactus pear varieties and quercetin was predominant in fruits of all cactus pear varieties examined. While kaempferol was found in fruits of the green-skinned (*O. ficus-indica*), the purple-skinned (*O. lindheimeri*), and red-skinned (*O. streptacantha*) cactus pears, isorhamnetin was found in fruits of the green-skinned and the purple-skinned cactus pears. Kaempferol was not detectable in fruits of yellow-skinned cactus pear (*O. stricta var stricta*) and isorhamnetin was not detectable in red-skinned or yellow-skinned cactus pear fruits. In terms of flavonoid composition, quercetin derivatives accounted for 62.2% (43.2±11.5 µg/g fresh weight), kaempferol derivatives accounted for 3.2% and isorhamnetin accounted for 34.7% of total flavonoids in green-skinned cactus pear fruits. The total flavonoids consisted of 96.8% quercetin (90.5±11.5 µg/g fresh weight) in purple-skinned, 93.1%

quercetin (51.0±4.6 µg/g fresh weight) in red-skinned, and 100% quercetin (9.8±3.0 µg/g fresh weight) in yellow-skinned cactus pear fruits, respectively. Among the cactus pear fruit varieties examined in this study, total flavonoid contents ranged from as low as ~9.8±3.0 µg/g fresh weight in yellow-skinned cactus pear fruits to as high as ~93.5±12.4 µg/g fresh weight in purple-skinned cactus pear fruits. It appears that *Opuntia* cactus pear fruits contain flavonoids common to other fruits and vegetables (Peterson & Dwyer, 1998); however, the flavonoid types and contents, as in other fruits and vegetables, vary with the cultivars (Bilyk & Sapers, 1986; Howard, Pandjaitan, Morelock, & Gil, 2002). The most important classes of phytochemicals in plant foods are phenolics and there are more than 8000 phenolic phytochemicals. The three main classes of dietary phenolics are flavonoids, and phenolic acids (Vinson, Su, Zubik, & Bose, 2001). The flavonoids constitute about one-half of the 8000 or so recognized phenols and are molecules responsible for the colour of fruit and flowers (Cook & Samman, 1996). One of the more interesting findings in this study regarding the phytochemical content of cactus pear fruits, is that they are relatively high in the flavonols (i.e. quercetin). The flavonol quercetin is one of the most commonly consumed flavonoids and has been well studied for its potential health benefits. Quercetin possesses anti-proliferate, anticarcinogenic and antioxidant activities (Kandaswami & Middleton, 1994).

3.2. Ascorbic acid and carotenoid contents of cactus pear fruit extracts

Generally, fruit extracts of all the cactus pear varieties had higher concentrations of ascorbic acid than carotenoids (Table 3). The total ascorbic acid content ranged from 10 to 111 µg/g fresh weight in purple-skinned cactus pear fruits and from 23 to 792 µg/g fresh weight in red-skinned fruits, which are considerably higher than the average vitamin C contents in some common fruits, such as peaches, grapes and apple (Zapata & Dunfour, 1992). The results on total ascorbic acid content in cactus pears agree with previously reported values (Kuti, 1992; Sawaya et al., 1983). Ascorbic acid is an important nutrient antioxidant (Sies & Stahl, 1995). The yellow-skinned cactus pear varieties had the highest total carotenoid concentrations, that ranged from 6.0 to 17.7 µg/g fresh weight and the green-skinned varieties had the lowest carotenoid concentrations, that ranged from 1.2 to 1.7 µg/g fresh weight, respectively. Our data on the total carotenoid concentrations in cactus pear fruits, even though slightly lower than those reported for other fruits, agree with the observation that yellow-coloured fruits usually have higher carotenoids than other coloured fruits (Romariz, 1949; USDA-NCC, 1998). Carotenoids are widely distributed among



Quercetin ($R^1 = \text{OH}$; $R^2 = \text{H}$)

Kaempferol ($R^1 = R^2 = \text{H}$)

Isorhamnetin ($R^1 = \text{OCH}_3$; $R^2 = \text{H}$)

Fig. 1. Chemical structures of flavonoids (quercetin, kaempferol, and isorhamnetin) from cactus pear (*Opuntia* species) fruit.

Table 3
Total ascorbic acid and carotenoid distribution in fruit extracts of different *Opuntia* cactus pears

<i>Opuntia</i> ^a species	Concentration ^b (µg/g fresh weight)	
	Ascorbic acid	Total carotenoids
<i>O. ficus-indica</i>	458	2.9
<i>O. lindheimeri</i>	121	6.7
<i>O. streptacantha</i>	815	14.6
<i>O. stricta</i> var. <i>stricta</i>	437	23.7
LSD (0.05) ^a	148	4.4

^a Means within the column are not significantly different ($P < 0.05$) using Waller and Duncan's Bayes least significance difference (LSD) test.

^b *Opuntia ficus-indica* (green-skinned cactus pear); *Opuntia lindheimeri* (purple-skinned cactus pear); *Opuntia streptacantha* (red-skinned cactus pear); *Opuntia stricta* var. *stricta* (yellow-skinned cactus pear)

coloured fruits and vegetables and contribute to both the appearance and attractiveness of fruit as well as additional nutritional value in the form of dietary antioxidants (Sies & Stahl, 1995). Epidemiological studies have shown that high intakes of carotenoid-rich vegetables and fruits and high blood levels of β -carotene are associated with decreased incidence of some cancers (Slattery, Benson, Curtin, Ma, Schaeffer, & Potter, 2000).

3.3. Total flavonoids and antioxidant capacity of cactus pear fruit constituents

Total flavonoid contents and antioxidant activities of the four varieties of cactus pear fruit are shown in Table 4. The purple-skinned cactus pear varieties had the highest total flavonoids (93.5 ± 12.4 µg/g of fruit), followed by the green-skinned (69.5 ± 3.8 µg/g of fruit),

Table 4
Total flavonoids and antioxidant capacity (ORAC value) in fruit extracts of different *Opuntia* cactus pears

<i>Opuntia</i> species	Total flavonoids (µg/g fresh wt.)	ORAC (µmol of TE/g) ^a
<i>O. ficus-indica</i> (green-skinned)	69.5 ± 3.8^b	26.3 ± 1.8
<i>O. lindheimeri</i> (purple-skinned)	93.5 ± 12.4	49.2 ± 1.7
<i>O. streptacantha</i> (red-skinned)	54.8 ± 5.1	25.2 ± 2.1
<i>O. stricta</i> var. <i>stricta</i> (yellow-skinned)	9.8 ± 3.0	15.8 ± 1.6
LSD (0.05) ^c	6.8	1.1

^a TEAC = the trolox equivalent antioxidant capacity (in units µmol g⁻¹).

^b Mean for triplicate determination \pm standard errors of means of total flavonoids and antioxidant capacities.

^c Means within the column are not significantly different ($P < 0.05$) using Waller and Duncan's Bayes least significance difference (LSD) test.

the red-skinned (54.8 ± 5.1 µg/g of fruit) and the yellow-skinned (9.8 ± 3.0 µg/g of fruit), respectively. Preponderance of the cactus pear fruit flavonoids is in the fruit pulp tissues (data not shown).

The ORAC values measured in terms of Trolox equivalent (TE) among the cactus pear varieties range from as low as 15.8 ± 1.6 µM TE/g of fresh weight in the yellow-skinned fruits to as high as 49.2 ± 1.7 µM TE/g of fresh weight in the purple-skinned fruits (Table 4). The average total TE values were 26.3 ± 1.8 µM TE/g of fresh weight for the green-skinned, 49.2 ± 1.7 µM TE/g of fresh weight for the purple-skinned, 25.2 ± 2.2 µM TE/g of fresh weight for the red-skinned, and 15.8 ± 1.6 µM TE/g of fresh weight for the yellow-skinned cactus pear fruits. Collectively, our data suggest that ORAC and flavonoid contents were significantly different among the varieties of cactus pear fruits. Correlation coefficients between ORAC and total flavonoid contents of the different cactus pear fruits are shown in Table 5. No correlation between ORAC and ascorbic acid or carotenoid content in cactus pear fruits was observed (data not shown). Wang et al. (1996) had previously reported that ORAC is poorly correlated with ascorbic acid content, thus suggesting that other components, such as phenolics (flavonoids) contribute more significantly to the total antioxidant capacity. The correlation coefficient between antioxidant capacity and flavonoid contents ranged from $r = 0.76$ in the yellow-skinned to $r = 0.88$ in the purple-skinned cactus pear varieties. In general, correlation coefficients between ORAC and flavonoid contents were positive and highly significant ($P \leq 0.01$) in the purple-skinned and significant ($P \leq 0.05$) in the green-skinned, the red-skinned and yellow-skinned cactus pear fruits, respectively. This observation agrees with the work of Veliogluo, Mazza, Gao, and Oomah (1998) regarding correlation of total phenolic content with antioxidant capacity in selected fruits, vegetables and grain products. Many natural flavonoids have considerably higher antioxidant potentials than nutrient antioxidants, such as vitamin C (ascorbic acid) and vitamin E and dietary antioxidants, such as

Table 5
Correlation coefficients between antioxidant activity (ORAC value) and total flavonoid content in extracts of different *Opuntia* cactus pears

<i>Opuntia</i> spp.	Correlation coefficient (r) ^a
	ORAC vs. total flavonoids
<i>O. ficus-indica</i>	0.78*
<i>O. lindheimeri</i>	0.88**
<i>O. streptacantha</i>	0.80*
<i>O. stricta</i> v. <i>stricta</i>	0.76*

^a Values are means ($n = 15$).

^b $P \leq 0.05$.

^c $P \leq 0.01$.

Table 6
Relative antioxidant activities of selected reference standards assayed using trolox equivalent (ORAC) assay method

Standard	TE ($\mu\text{mol/l}$)
Trolox C	1.00
Polyphenols (cyanidin 3-glucoside “kuramanin”)	9.37 \pm 0.15
Quercetin	1.08 \pm 0.09
Kaempferol	0.95 \pm 0.01
Ascorbic acid (vitamin C)	0.89 \pm 0.30
Carotenoid (β -carotene)	0.32 \pm 0.02

carotenoids (Vinson, Dabbagh, Serry, & Jang, 1995). While the cactus pear fruit flavonoids content appears to be positively correlated with antioxidant capacity, quercetin, the predominant cactus pear flavonoid, had a good antioxidant index, TE ($\mu\text{mol/l}$), when compared with the index of reference compounds (Table 6). The purple-skinned cactus pears with highest total flavonoids had the highest antioxidant activity, whereas yellow-skinned cactus fruits with lowest total flavonoids had the lowest antioxidant activity.

The high antioxidant capacity in purple-skinned cactus pears, observed in this study, may be due to the high phenolic contents or possibly a combination of individual antioxidants producing synergistic effects. Even though no clear general trend in terms of high antioxidant capacity among cactus pears, as a group, was observed, individual varieties of cactus pears may be important in the selection of cactus pear fruits as a good source of antioxidants that can be used in foods and nutritional supplement formulations.

4. Conclusion

In conclusion, this investigation shows the potential value of *Opuntia* cactus pear fruits as a good source of natural antioxidants and that consumption of cactus pear fruit or its products may contribute substantial amounts of antioxidants to the diet. Based on the available data in this study and the phytochemical contents of cactus pear fruits, there is a high likelihood that cactus pear fruits may provide the types of nutritional and health benefit associated with consumption of fruits and vegetables in general. Further studies on the absorption and effects of cactus pear phytochemicals on antioxidant status in animal models are needed to evaluate their potential health benefit.

Acknowledgements

The author gratefully acknowledges the help of Ms. Hima Bindu Konuru, Research Assistant at the Horticultural Crops & Food Research Lab, for her excellent

technical assistance in HPLC analysis and ORAC assay. This work was financially supported in part by a grant from the USDA-CSRS under the Special Research Grant to the author.

References

- Arcoleo, A., Ruccia, M., & Cusmando, S. (1961). Flavonoid pigments from *Opuntia*. I. Isorhamnetin from flowers *Opuntia ficus-indica*. *Annali Chimica (Rome)*, 51, 81.
- Barbera, G., Carimi, C., & Inglese, P. (1992). Past and present role of the Indian-fig, prickly pear, (*Opuntia ficus-indica* (L.) Miller, Cactaceae), in agriculture of Sicily. *Economic Botany*, 46, 10–20.
- Benson, L. (1982). *Cacti of United States and North America*. Stanford, CA: Stanford University Press.
- Bilyk, A., & Sapers, G. M. (1986). Varietal differences in the quercetin, kaempferol, and myricetin contents of highbush blueberry, cranberry, and thornless blackberry fruits. *Journal of Agricultural & Food Chemistry*, 34, 585–588.
- Block, G., Patterson, B., & Subbar, A. (1992). Fruit, vegetables and cancer prevention: a review of epidemiological evidence. *Nutrition & Cancer*, 18, 1–29.
- Burret, F., Lebreton, P., & Voirin, B. (1982). Les aglycones flavoniques des Cactées: distribution, signification. *Journal Natural Products*, 45, 687–693.
- Bustos, O. E. (1981). Alcohol beverage from Chilean *Opuntia ficus-indica*. *American Journal of Enology & Viticulture*, 32, 228–229.
- Cao, G., Alessio, H., & Cutler, H. (1993). Oxygen-radical absorbance capacity assay for antioxidants. *Free Radical Biology & Medicine*, 14, 303–311.
- Cao, G., Sofic, E., & Prior, R. (1996). Antioxidant activity of tea and common vegetables. *Journal of Agricultural & Food Chemistry*, 44, 3426–3431.
- Clark, W. D., & Parfitt, B. D. (1980). Flower flavonoids of *Opuntia* series *Opuntiae*. *Phytochemistry*, 19, 1856–1857.
- Cook, N. C., & Samman, S. (1996). Flavonoids—chemistry, metabolism, cardioprotective effects, and dietary sources. *Nutritional Biochemistry*, 7, 66–76.
- Dragsted, L. O., Strube, M., & Larsen, J. C. (1993). Cancer-protective factors in fruits and vegetables: biochemical and biological background. *Pharmacological Toxicology*, 72(Suppl. 1), 116–135.
- Edenharder, R., Keller, G., Platt, K. L., & Unger, K. K. (2001). Isolation and characterization of structurally novel antimutagenic flavonoids from spinach (*Spinacia oleracea*). *Journal of Agricultural & Food Chemistry*, 49, 2767–2773.
- Francis, F. J. (1989). Food colorants: anthocyanins. *Critical Review of Food Science & Nutrition*, 28, 273–314.
- Gil, M. I., Tomas-Barberan, F. A., Hess-Pierce, B., Holcroft, D. M., & Kader, A. A. (2000). Antioxidant activity of pomegranate juice and its relationship with phenolic composition and processing. *Journal of Agricultural & Food Chemistry*, 48, 4581–4589.
- Gurrieri, S., Miceli, L., Lanza, C. M., Tomaselli, F., Bonomo, R. P., & Rizzarelli, E. (2000). Chemical characterization of Sicilian prickly pear (*Opuntia ficus-indica*) and perspectives for storage of its juice. *Journal of Agricultural & Food Chemistry*, 48, 5424–5431.
- Herrman, K. (1988). On the occurrence of flavonol and flavone glycosides in vegetables. *Zeitschrift für Lebensmittel-Untersuchung und-Forschung*, 186, 1–5.
- Hoffman, W. (1980). The many uses of prickly pears (*Opuntia* Mill.) in Peru and Mexico. *Plant Resources & Development*, 12, 58–68.
- Hollman, P. C. H., Hertog, M. G. L., & Katan, M. B. (1996). Analysis and health benefits of flavonoids. *Food Chemistry*, 57, 43–46.
- Howard, L. R., Pandjaitan, N., Morelock, T., & Gil, M. I. (2002). Antioxidant capacity and phenol content of spinach as affected by

- genetics and growing season. *Journal of Agricultural & Food Chemistry*, 50, 5891–5896.
- Kandaswami, C., & Middleton Jr., E. (1994). Free radical scavenging and antioxidant activity of plant flavonoids. *Advanced Experimental Biology & Medicine*, 366, 351–376.
- Kuti, J. O. (1992). Growth and compositional changes during development of prickly pear fruit. *Journal of Horticultural Science*, 67, 861–868.
- Kuti, J. O. (2000). Antioxidant activity of *Opuntia* cactus pear. *HortScience*, 35, 433.
- Lee, J. C., & Lim, K. T. (2000). Effects of cactus and ginger extract as dietary antioxidants on reactive oxidant and plasma lipid level. *Food Science & Biotechnology*, 9, 83–88.
- Merken, H. M., & Beecher, G. R. (2000). Measurement of food flavonoids by high-performance liquid chromatography: a review. *Journal of Agricultural & Food Chemistry*, 48, 577–599.
- Meyer, B., & McLaughlin, J. (1982). A note on the phytochemistry of *Opuntia* (Cactaceae). *Cactus & Succulent Journal (USA)*, 54, 226–228.
- Middleton, E., & Kandaswami, C. (1992). Effects of flavonoids on immune and inflammatory cell functions. *Biochemical Pharmacology*, 43, 1167–1179.
- Miller, J. M., & Bohm, B. A. (1982). Flavonol and dihydroflavonol glycosides of *Echinocerus triglochidatus* var. *gurneyi*. *Phytochemistry*, 21, 951–952.
- Namiki, M. (1990). Antioxidant/antimutagens in food. *Critical Review Food Science & Nutrition*, 29, 273–300.
- Ness, A. R., & Powles, J. W. (1997). Fruit and vegetables, and cardiovascular disease: a review. *International Journal of Epidemiology*, 26, 1–13.
- Peterson, J., & Dwyer, J. (1998). Taxonomic classification helps identify flavonoid-containing foods on a semiquantitative food frequency questionnaire. *Journal of American Dietetic Association*, 98, 677–682 685.
- Retamal, N., Duran, J. M., & Fernandez, J. (1987). Ethanol production by fermentation of fruits and cladodes of prickly pear cactus (*Opuntia ficus-indica* (L.) Miller). *Journal of the Science of Food & Agriculture*, 40, 213–218.
- Richardson, P. M. (1978). Flavonols and C-glycosyl flavonoids of the Caryophyllales. *Biochemical & Systematic Ecology*, 6, 283–286.
- Rodriguez-Amaya, D. B. (1999). *A guide to carotenoid analysis*. ILDI Press: Washington DC.
- Romariz, I. (1949). Unusual carotenoids in young leaves. *Chemical Abstracts*, 56, 10578.
- Rosler, H., Rosler, U., Mabry, T., & Kagan, J. (1966). The flavonoid pigments on *Opuntia lindheimeri*. *Phytochemistry*, 5, 189.
- Saenz, C. (1996). Uses of prickly pear fruit. *Intermediate Technology & Food Chain*, 18, 10–11.
- Sawaya, W. N., Khatchadourin, H., Safi, W., & Al-Muhammed, H. M. (1983). Chemical characterization of prickly pear pulp, *Opuntia ficus-indica* and manufacturing of prickly pear jam. *Journal of Food Technology*, 18, 183–193.
- Savio, Y. (1987). Prickly pear cactus. *Cactus & Succulent Journal (USA)*, 59, 113–117.
- Shabbir, M., & Zaman, A. (1968). Chemical investigation of the flowers of *Opuntia elatior*. *Journal of Indian Chemical Society*, 45, 81.
- Sies, H., & Stahl, W. (1995). Vitamins E and C, beta-carotene and carotenoids as antioxidants. *American Journal of Clinical Nutrition*, 62, 1315 s-1321s.
- Slattery, M. L., Benson, J., Curtin, K., Ma, K. N., Schaeffer, D., & Potter, J. D. (2000). Carotenoids and colon cancer. *American Journal of Clinical Nutrition*, 71, 575–582.
- Steinmetz, K. A., & Potter, J. D. (1996). Vegetables, fruit and cancer prevention: a review. *Journal American Dietetic Association*, 97, 1027–1039.
- Teles, F., Stull, J., Brown, W., & Whitting, F. (1984). Amino and organic acids of prickly pear cactus (*Opuntia ficus-indica* L.). *Journal of the Science of Food & Agriculture*, 35, 421–425.
- USDA-NCC. (1998). Carotenoid database for US foods. Available: <http://www.nal.usda.gov/fnic/foodcomp/Data/car98/car98.html>.
- Velioglu, Y. S., Mazza, G., Gao, L., & Oomah, B. D. (1998). Antioxidant activity and total phenolics in selected fruits, vegetables, and grain products. *Journal of Agricultural Food & Chemistry*, 46, 4113–4117.
- Vinson, J. A., Dabbagh, Y. A., Serry, M. M., & Jang, J. (1995). Plant flavonoids, especially tea flavonols, are powerful antioxidants using an in vitro oxidation model for heart disease. *Journal of Agricultural & Food Chemistry*, 43, 2800–2802.
- Vinson, J. A., Su, X., Zubik, L., & Bose, P. (2001). Phenol antioxidants quantity and quality in foods: fruits. *Journal of Agricultural & Food Chemistry*, 49, 5315–5321.
- Wallace, R. S. (1986). Biochemical taxonomy and the Cactaceae. *Cactus & Succulent Journal (USA)*, 58, 35–38.
- Wang, H., Cao, G., & Prior, R. L. (1996). Total antioxidant capacity of fruits. *Journal of Agricultural & Food Chemistry*, 44, 701–705.
- Yoshida, M., Sakai, T., Hosokawa, N., Marui, N., Matsumoto, K., Fugioka, A., Nishino, H., & Aoiike, A. (1990). The effect of quercetin in cell cycle progression and growth of human gastric cancer cells. *Federation of European Biological Science Letter*, 260, 10–13.
- Youdim, K. A., & Joseph, J. A. (2001). A possible emerging role of phytochemicals in improving age-related neurological dysfunctions: multiplicity of effects. *Free Radical Biology & Medicine*, 30, 583–594.
- Zapata, S., & Dufour, J. F. (1992). Ascorbic dehydroascorbic and isoascorbic acid simultaneous determination by reverse phase ion interaction HPLC. *Journal of Food Science*, 57, 506–511.