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Antioxidant Activity and Phenolic Content of Oregon Caneberries

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Five types of caneberries [evergreen blackberries (*Rubus laciniatus*), marionberries (*Rubus ursinus*), boysenberries (*Rubus ursinus* × *idaeus*), red raspberries (*Rubus idaeus*), and black raspberries (*Rubus occidentalis*)] were analyzed for antioxidant activity by measuring their oxygen radical absorbance capacity (ORAC). In addition, the berries were analyzed for total phenolics, anthocyanins, procyanidins, and ellagic acid content. All of the berries had high ORAC activity ranging from 24 to 77.2 μmol of Trolox equiv/g of fresh berries. Anthocyanin content ranged from 0.65 to 5.89 mg/g, and phenolics ranged from 4.95 to 9.8 mg/g. Black raspberries had the highest ORAC and anthocyanin and phenolic contents. Only red raspberries had detectable amounts of procyanidin oligomers (monomer, dimers, and trimers). All berries had high levels of ellagic acid (47−90 mg/g), but boysenberries had the highest level prior to hydrolysis. The results from this study indicate that these caneberries were high in antioxidant activity and were rich sources of anthocyanins and phenolics.

KEYWORDS: Caneberries; blackberries; raspberries; boysenberries; ORAC; anthocyanins; phenolics; procyanidin; ellagic acid

INTRODUCTION

High intakes of fruits and vegetables have been associated with lower incidences of chronic diseases such as cancer and heart disease (*1*, *2*). In addition to the vitamins and minerals known to be present in fruits and vegetables, phytochemicals such as flavonoids and other phenolics may contribute to this protective effect. Many of these phytochemicals have antioxidant activity and may help protect cells against the oxidative damage caused by free radicals.

Recently there has been an increasing amount of attention given to the health benefits of consuming berries such as blueberries (*3*, *4*). Blueberries contain anthocyanins, the flavonoid pigment responsible for their red to bluish hue. Anthocyanins exhibit antioxidant activity (*5*) and inhibit low-density lipoprotein (LDL) oxidation (*6*). They have also been shown to have vasoprotective and antiinflammatory activity (*7*, *8*). Anthocyanin-rich extracts from European berries such as the bilberry (*Vaccinium myrtillus*) have been sold commercially to treat microcirculation disease and maintain normal vascular permeability (*9*).

Prior et al. have reported that blueberries possess considerably high antioxidant activity that is attributed in part to their anthocyanin content (*3*). Heinonen et al. used two oxidation models, human LDL and lecithin liposomes, to study the antioxidant activity of various berries including red raspberries and black raspberries. They found the berries to possess antioxidant activity, although the degree of activity and relative ranking varied with the model used (*10*). Others have assessed the antioxidant activity of berries including black and red raspberries by measuring the superoxide scavenging activity (*11*). Recently, Wang and Lin (*12*) measured the oxygen radical absorbance capacity (ORAC) of red raspberries, black raspberries, and blackberries and found these berries to have high ORAC levels.

Some berries such as strawberries and black raspberries have been identified as sources of the phenolic compound ellagic acid, which has been demonstrated to have potential cancer chemopreventive activity (*13*). Other berries such as blueberries and cranberries have been identified as sources of procyanidins. Procyanidins are a group of flavonoids composed of flavan-3-ol monomers linked together into polymers of various lengths. Oligomeric procyanidins have been shown to increase plasma antioxidant capacity (*14*) and inhibit atherosclerosis in rabbits (*15*). The ability of blueberries and cranberries to inhibit *Escherichia coli* adhesion and prevent urinary tract infection has been attributed to their procyanidin content (*16*).

The purpose of this study was to study a particular group of berries known as caneberries. Caneberries are a group of berries that grow on leafy canes in temperate regions of the world. The best known commercial caneberries are the red raspberry (*Rubus idaeus*), black raspberry (*Rubus occidentalis*), marionberry (*Rubus ursinus*), evergreen blackberry (*Rubus laciniatus*), and boysenberry (*Rubus ursinus* × *idaeus*). In the United States, caneberries were used centuries ago by Native Americans and settlers for food, as dye for clothing, and for medicinal purposes. In the 19th century cultivated production began in the Pacific Northwest, which is still the leading area of caneberry production (*17*).

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Caneberries are considered to be nutritious, are low in fat, and provide a good source of dietary fiber, vitamin C, and potassium (*17*). To learn more about the antioxidant activity and phytochemical composition of caneberries grown in Oregon, samples were analyzed for ORAC and phenolic, anthocyanin, procyanidin, and ellagic acid contents.

MATERIALS AND METHODS

Chemicals. β-Phycoerythrin, gallic acid, catechin, rutin, isoquercitrin, epicatechin gallate, quercitrin, myricetin, quercetin, and kaempferol were purchased from Sigma Chemical Co. (St. Louis, MO). 6-Hydroxy-2,5,7,8-tetramethyl-2-carboxylic acid (Trolox) was obtained from Aldrich (Milwaukee, WI). 2,2′-Azobis(2-amidinopropane) dihydrochloride (AAPH) was obtained from Wako Chemicals USA, Inc. (Richmond, VA). Methanol and acetonitrile (HPLC grade) were from Fisher Scientific (Springfield, NJ).

Berries. Five species of caneberries were evaluated in this study. These included red raspberries (*R. idaeus*), black raspberries (*R. occidentalis*), marionberries (*R. ursinus*), evergreen blackberries (*R. laciniatus*), and boysenberries (*R. ursinus* × *idaeus*). All berries were grown near Salem, OR. Only berries that were ripe by commercial standards (no green or overripe berries) were harvested and collected from a central processing plant close to the berry farms. Approximately 500 g of each berry was shipped frozen and received overnight. Samples were kept frozen until analysis. At least three determinations were performed for each analysis, and the average was reported.

ORAC Assay. The 500 g sample of each berry was initially ground in a mechanical mill. Then 0.5 g was accurately weighed, and 20 mL of acetone/water (50:50 v/v) extraction solvent was added. The mixture was shaken for 1 h at room temperature on an orbital shaker. On the basis of data from recovery studies we had previously performed, this amount of time was adequate to extract most phenolic compounds. The extracts were centrifuged at 5900 rpm, and the supernatant was ready for analysis. The ORAC assay is based on a procedure of Cao et al. (*18*) and modified for the COBAS FARA II spectrofluorometric centrifugal analyzer (Roche Diagnostic System Inc., Branchburg, NJ) (*19*). Fluorescence is measured at 565 nm with the excitation wavelength at 540 nm. AAPH is used as the source for the peroxyl radical, which is generated as a result of the spontaneous decomposition of AAPH at 37 °C. β-Phycoerythrin is the chosen target protein; its loss of fluorescence is an indication of the extent of damage from its reaction with the peroxyl radical. The protective effect of the antioxidants is measured by assessing the longer fluorescence time/intensity area under the curve of the sample compared to that of the blank in which no antioxidant compounds are present. Trolox, a water soluble analogue of vitamin E, was used as the calibration standard. Fluorescence readings are taken every 2 min for up to 70 min after the addition of AAPH. The ORAC results are calculated on the basis of the calibration curve obtained in each run and reported as micromoles of Trolox equivalents (TE) per gram of fresh weight (FW) or dry matter (DM). Dry matter was determined after lyophilization.

Anthocyanins. *Sample Preparation.* Five grams of the milled whole berries was ground again in a high-speed mill. After grinding, 0.5 g samples were extracted with methanol acidified with 0.1% HCl. The extractions were performed on an orbital shaker operated at 400 rpm at room temperature for 1 h. This has proved to be adequate for complete extraction. The samples were then centrifuged at 5900 rpm for 15 min. The supernatant was recovered and filtered through a 0.45 μm cellulose syringe filter before analysis.

Total Anthocyanin Assay. The total anthocyanins were estimated by a pH differential method (*20*). Absorbance was measured in a COBAS FARA II spectrofluorometric centrifugal analyzer at 510 nm and at 700 nm in buffer at pH 1.00 and pH 4.5, using $A = (A_{510} - A_{700})_{pH1.0} - (A_{510} - A_{700})_{pH4.5}$ with a molar extinction coefficient of cyanidin 3-glucoside of 29600. Results were expressed as milligrams of cyanidin 3-glucoside equivalent per gram of fresh weight or dry matter.

LC-MS Structural Analysis. The structural information of individual anthocyanins was obtained by liquid chromatography coupled with ion trap mass spectrometry (LC-MS^n). The system used for LC-MS analysis was a Finnigan MAT LCQ ion trap mass spectrometer (ThermoFinnigan, San Jose, CA) equipped with an HP 1100 binary system consisting of an autosampler and diode array detector (Hewlett-Packard, Palo Alto, CA) set at 550 nm. The separation of anthocyanin was performed using a Phenomenex (Torrance, CA) 5 μm Phenyl Hexyl column (250 × 4.6 mm). The binary mobile phase consisted of (A) water, acetonitrile, and acetic acid (89:9:2, v/v) and (B) water and acetonitrile (20:80, v/v). The gradient method started at 1 mL/min from 100% A for 25 min and then was linearly changed to 100% B over 15 min. The ionization parameters for the mass spectrometer were optimized using constant infusion of cyanidin 3-glucoside to the ion source. The heated capillary and voltage were maintained at 175 °C and 2 kV, respectively. The full-scan mass spectra from *m*/*z* 100 to 1000 were collected. Tandem mass spectrometry was performed using helium as the collision gas, and the collision energy was set at 25%. All mass spectrometry data were acquired in the positive ionization mode.

Phenolics. *Sample Preparation.* Approximately 15 g of ground berries was weighed into a 50 mL polyethylene centrifuge tube with 20 mL of methanol. The sample was extracted for 1 h at room temperature on an orbital shaker operated at 400 rpm and then centrifuged at 5900 rpm, and the supernatant was immediately analyzed. For ellagic acid (EA), 15 g of the berries was extracted by methanol at 100 °C for 24 h (containing free EA). The extract was then evaporated to dryness and hydrolyzed in 2 N trifluoroacetic acid in methanol at 100 °C for 2 h (containing total EA).

Total Phenolic Assay. Total phenolics content was determined according to the Folin−Ciocalteu procedure on a COBAS FARA II centrifugal analyzer using gallic acid as a standard (*21*). The instrument was operated in the spectrophotometric mode measuring the absorption of 750 nm. The total phenolic content was expressed as gallic acid equivalents in milligrams per 100 g.

LC-MS Structural Analysis. A validated LC-MS method was utilized for structural analysis of phenolic compounds. HPLC conditions were identical to those used for anthocyanin analysis as mentioned above except that the diode array detector was set at 278 nm. The ionization parameters for the mass spectrometer were optimized using constant infusion of rutin to the ion source. The heated capillary and voltage were maintained at 200 °C and 3.5 kV, respectively. The collision energy was set at 30%. All mass spectrometry data were acquired in the positive ionization mode. Catechin, epicatechin, gallate, rutin, isoquercitrin, quercitrin, myricetin, quercetin, kaempferol, chlorogenic acid, benzoic acid, caffeic acid, ellagic acid, ferulic acid, and coumaric acid were used as standards. The identities of individual compounds were confirmed by the retention time and molecular weight obtained by LCQ mass detector.

Procyanidins. *Sample Preparation.* Five grams of each berry was accurately weighed into a 50 mL polyethylene centrifuge tube with 20 mL of extraction solvent containing acetone, water, and acetic acid (70:29.5:0.5 v/v). The mixture of solvent and sample was vortexed, sonicated for 5 min in a water bath at 50 °C, and allowed to extract for 30 min. The mixture was centrifuged at 5900 rpm for 15 min. The resulting supernatant was rotary evaporated at 50 °C under partial vacuum, and the residue was diluted to 5 mL with deionized water. The procyanidin fraction was isolated using SPE columns, which were wet packed with 5 g of Sephadex LH-20 hydrated in 25 mL of water. An aliquot of 5 mL of each of the extracted samples was loaded onto the column. Each column was eluted with 45 mL of 20% methanol/water (v/v) to remove sugars and phenolic acids, followed by 40 mL of 60% methanol/water (v/v) to elute the flavonols and anthocyanins, and finally with 90 mL of 100% methanol for elution of the procyanidins. Other types of samples that have higher concentrations of procyanidins may require more rigorous procedures. However, this procedure was sufficient to elute the amount of procyanidins in these berry samples. The 100% methanol fraction was concentrated by rotary evaporation, and the concentrated material was diluted to a final volume of 5 mL with 100% methanol.

LC-MS Structural Analysis. The LC-MS method was based on that of Adamson and co-workers (*22*). Fluorescence detection was recorded at an excitation wavelength of 276 nm and an emission wavelength of 316 nm. Normal phase separations of the procyanidin oligomers were performed on a Phenomenex (Torrance, CA) 5 μm Luna Silica (3.0 × 250 nm) column at 37 °C with a 5 μL injection volume. The binary

Table 1. Antioxidant Activity (ORAC) and Anthocyanin and Phenolic Contents of Berries

species	ORAC[a,b] (μmol of TE/g)	phenolics[b,c] (mg/g)	anthocyanins[c,d] (mg/g)
evergreen blackberries	27.5 ± 2.6 (175)	4.95 ± 0.13 (30.94)	0.91 ± 0.02 (5.69)
red raspberries	24.0 ± 1.8 (171)	5.17 ± 0.12 (36.93)	0.65 ± 0.03 (4.64)
boysenberries	42.2 ± 2.5 (350)	5.99 ± 0.16 (49.92)	1.31 ± 0.01 (10.92)
marionberries	28.0 ± 2.9 (215)	5.83 ± 0.15 (44.85)	1.55 ± 0.02 (11.92)
black raspberries	77.2 ± 2.5 (453)	9.80 ± 0.10 (57.65)	5.89 ± 0.04 (34.65)

[a] Expressed as micromoles of Trolox equivalents per gram of fresh fruit. [b] Data in parentheses expressed per gram of dry matter. [c] Concentration based upon gallic acid as standard expressed per gram of fresh weight. [d] Concentration based upon cyanidin 3-glucoside as standard expressed per gram of fresh weight.

Table 2. Percent Contribution of Individual Anthocyanins to Total Anthocyanins

anthocyanin	evergreen blackberries (%)	red raspberries (%)	boysen-berries (%)	marion-berries (%)	black raspberries (%)
cyanidin 3-(6′-*p*-coumaryl)glucoside-5-glucoside			56.27[a]		
cyanidin 3,5-diglucoside		89.25			
cyanidin 3-(6′-*p*-coumaryl)sambubioside					22.00
cyanidin 3-(6′-*p*-coumaryl)glucoside				94.86[b]	77.00
cyanidin 3-glucoside	80.43	10.75	43.73		
cyanidin 3-arabinoside	10.19				
cyanidin 3-(6′-malonyl)glucoside	6.06			1.27	
unknown	3.40			3.87	1.00

[a] Coelutes with cyanidin 3,5-diglucoside (minor amount). [b] Coelutes with cyanidin 3-glucoside (minor amount).

mobile phase consisted of (A) dichloromethane, methanol, water, and acetic acid (82:14:2:2, v/v) and (B) methanol, water, and acetic acid (96:2:2, v/v). The gradient method started at 1 mL/min from 0 to 17.6% B in 30 min, followed by 17.6−30.7% B in 15 min, and then 30.7−87.8% in 15 min. In all cases, the columns were re-equilibrated between injections with the equivalent of 25 mL of the initial mobile phase. Catechin standards were prepared and analyzed to establish a response calibration curve from which to calculate the concentration of procyanidins in the samples. The structures of the procyanidins were confirmed by molecular weights obtained by LCQ.

For the LCQ mass spectrometer, an ionization reagent (1.5 M NaOH) was added at a rate of 0.05 mL/min through a tee before the mass detector and was delivered using a secondary HPLC pump. Catechin was used as a calibration standard for tuning the mass detector. The heated capillary and voltage were maintained at 275 °C and 2 kV, respectively. The full-scan mass spectra from *m*/*z* 100 to 2000 were collected. The collision energy was set at 30%. All mass spectrometry data were acquired in the negative ionization mode.

RESULTS AND DISCUSSION

The antioxidant activity as measured by ORAC ranged from 24 μmol of Trolox equivalents (TE)/g in the fresh red raspberries to 77 μmol of TE/g in the fresh black raspberries (171−453 μmol of TE/g of dry matter) (**Table 1**). These values are high when compared to previous values reported for fruits and vegetables (*23*, *24*) and are as high or higher than ORAC values found in blueberries (*3*). The ORAC values of our marionberries and evergreen blackberries (28 μmol of TE/g) were similar to the range of 20.3−24.6 μmol of TE/g found in blackberries by Wang and Lin (*12*), who measured ORAC in blackberries and raspberries grown at the Beltsville Agricultural Research Center in Beltsville, MD. They also reported ORAC values for red raspberries of 15.9−20 μmol of TE/g, which was similar to our value of 24 μmol of TE/g. The ORAC for boysenberries (42 μmol of TE/g) was higher than that seen in either red raspberries or blackberries. We do not know of any other ORAC data on boysenberries. The ORAC of black raspberries was much higher than the levels in the other caneberries. The greatest difference between analyzed and reported values was seen with black raspberries. Wang and Lin (*12*) reported an ORAC value of 28.2 μmol of TE/g, whereas our analysis found a high ORAC of 77 μmol of TE/g. This difference may be due in part to the difference in cultivar. Wang and Lin used black raspberries of the Jewel cultivar, whereas the cultivar of our black raspberries was Munger. In addition, our analysis was performed on the whole berry including seeds, whereas Wang and Lin used only the juice expressed from the berries.

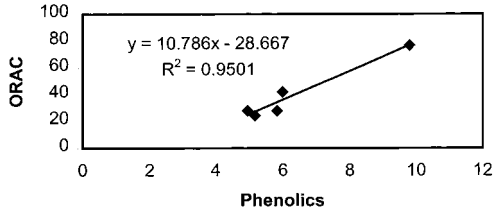

Figure 1. Relationship between ORAC (μmol of TE/g) (*Y*) and phenolics (mg/g) in caneberries.

Total phenolics ranged from 4.95 mg/g in evergreen blackberries to 9.80 mg/g in the black raspberries (30.94−57.65 mg/g of dry matter) (**Table 1**). As illustrated in **Figure 1**, a good linear relationship was found between ORAC and total phenolic content ($r^2 = 0.9501$), implying that the antioxidant activity of caneberries is largely due to the presence of phenolic compounds. The phenolic contents of evergreen blackberries, red raspberries, boysenberries, and marionberries were similar (4.95−5.99 mg/g), whereas black raspberries (9.8 mg/g) had higher levels than the other berries.

Black raspberries also had the highest level of anthocyanins, with 5.89 mg/g (34.65 mg/g of dry matter). The anthocyanin content of the other berries ranged from 0.65 mg/g in red raspberries to 1.55 mg/g in marionberries (4.64−11.92 mg/g of dry matter) (**Table 1**), which was similar to previous reports by Torre and Barritt, who found anthocyanin contents of 0.23−0.59 mg/g for red raspberries and 1.09 mg/g for marionberries (*25*). The relationship between ORAC and anthocyanins was not as strong as with phenolics ($r^2 = 0.932$, data not shown). This observation is in agreement with the findings of Prior and co-workers (*3*).

The anthocyanin contents are presented in **Table 2**. Using LC-MS analysis we are now able to identify various forms of anthocyanins on the basis of molecular weight and fragments

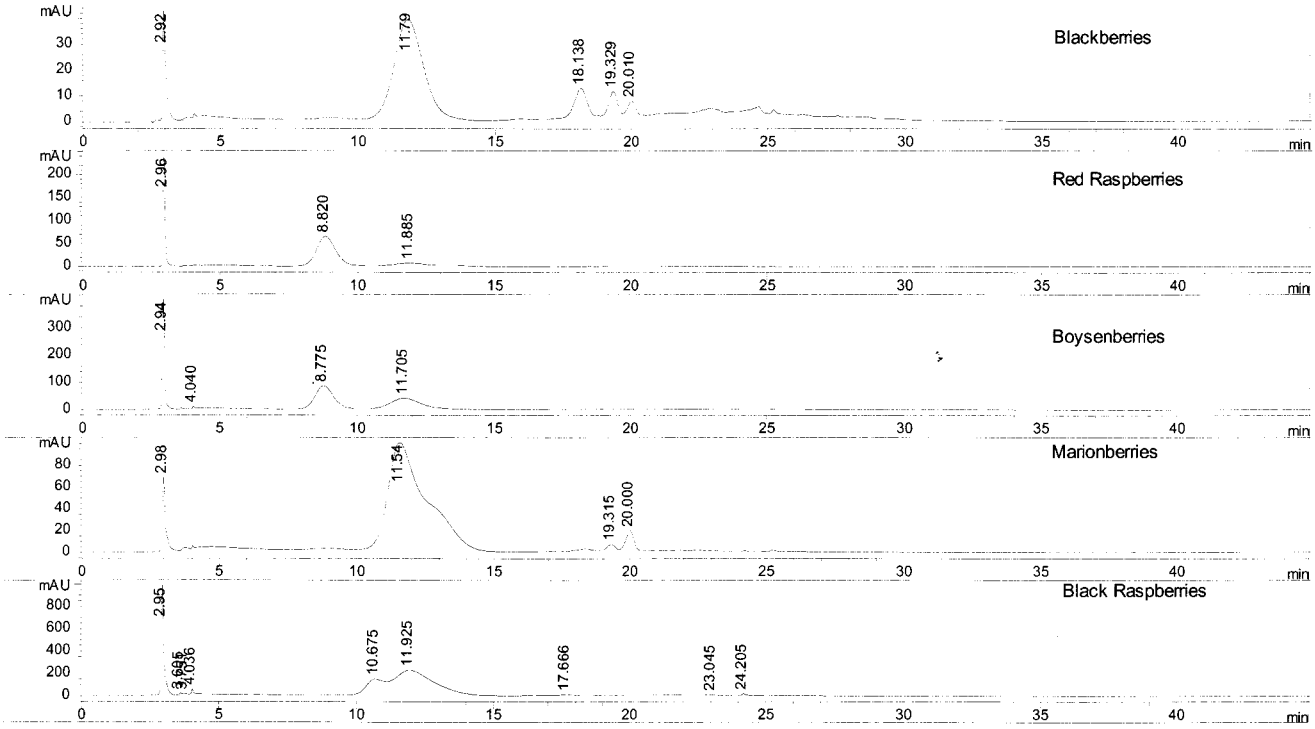

Figure 2. Chromatograms of anthocyanins from blackberries, red raspberries, boysenberries, marionberries, and black raspberries using HPLC coupled with UV detection.

Table 3. Fragmentation Pattern of Anthocyanins Present in Caneberries Analyzed by Ion Trap LC-MS

identification	retention time (min)	molecular ion $[M + 1]^+$	product ion
cyanidin 3-(6′-*p*-coumaryl)glucoside-5-glucoside	8.78	757.1	611.1, 433.0, 287.3
cyanidin 3,5-diglucoside	8.82	611.0	449.2, 287.3
cyanidin 3-(6′-*p*-coumaryl)sambubioside	10.68	727.1	580.7, 287.2
cyanidin 3-(6′-*p*-coumaryl)glucoside	11.92	595.1	449.0, 287.3
cyanidin 3-glucoside	11.79	449.0	287.3
cyanidin 3-arabinoside	18.14	418.7	287.3
cyanidin 3-(6′-malonyl)glucoside	19.33	535.0	491.1, 449.0, 287.3
unknown	20.01	593.1	287.3

of product ions. The HPLC chromatograms of anthocyanin profiles are illustrated in **Figure 2**, and their mass data are shown in **Table 3**. Cyanidin was the only anthocyanidin present in all of the berries, and the *m*/*z* for the aglycon was 287. The anthocyanins in evergreen blackberries and marionberries were predominantly cyanidin 3-glucoside (80.4%) and cyanidin 3-(6′-*p*-coumaryl)glucoside (94.9%). For red raspberries, cyanidin 3,5-diglucoside is the major anthocyanin (89.3%). The primary anthocyanins in boysenberries were cyanidin 3-(6′-*p*-coumaryl)-glucoside-5-glucoside and cyanidin 3-glucoside (56.27 and 43.73%, respectively), whereas cyanidin 3-(6′-*p*-coumaryl)-sambubioside and cyanidin 3-(6′-*p*-coumaryl)glucoside were the primary anthocyanin forms present in black raspberries (22.0 and 77.0%, respectively). Others have identified cyanidin 3-sophoroside as a major anthocyanin in red raspberries and boysenberries (*25*−*27*) and cyanidin 3-rutinoside as a major anthocyanin in black raspberries (*25*, *28*); however, we did not find evidence for these specific compounds based on the mass data.

Although the strong association between total phenolics and ORAC may be due in large part to the presence of anthocyanins, we also chose to study other individual phenolic compounds. Among the various phenolic compounds that were measured, only gallic acid, rutin, isoquercitrin, and ellagic acid were identified and quantified in the samples. Gallic acid was present in evergreen blackberries, marionberries, and boysenberries at 0.02, 0.03, and 0.09 mg/g, respectively. Rutin was present in all berries except boysenberries, at levels of 0.11 mg/g in red raspberries and marionberries, 0.19 mg/g in black raspberries, and 0.24 mg/g in evergreen blackberries. Isoquercitrin was found only in evergreen blackberries at 0.06 mg/g (**Table 4**). In the present study, due to the lack of commercial standards, we could not completely elucidate the profile of phenolics present in caneberries. However, our primary LC-MS data reveal that many phenolic compounds are existing as glycosylated forms. To completely identify each phenolic compound, extensive multi-stage mass study is needed.

Table 4. Phenolic Compounds in Berries

species	gallic acid[a] (mg/g)	rutin[a] (mg/g)	isoquercitrin[a] (mg/g)
evergreen blackberries	0.02 (0.13)	0.24 (1.50)	0.06 (0.38)
red raspberries	nd[b]	0.11 (0.79)	nd
boysenberries	0.09 (0.75)	nd	nd
marionberries	0.03 (0.23)	0.11 (0.82)	nd
black raspberries	nd	0.19 (1.11)	nd

[a] Data in parentheses expressed per gram of dry matter. [b] nd = not detected.

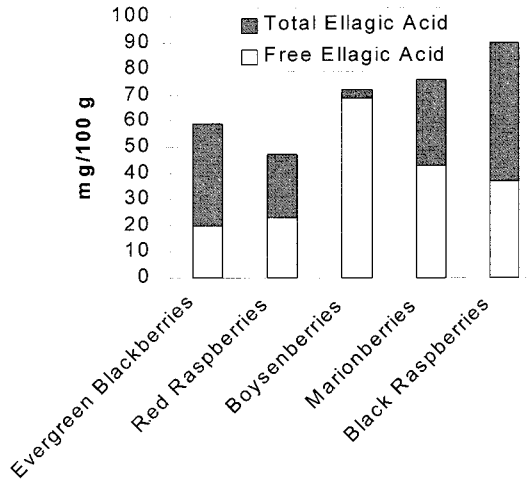

Figure 3. Total and free ellagic acid contents of evergreen blackberries, red raspberries, boysenberries, marionberries, and black raspberries.

The level of total ellagic acid ranged from 47 mg/g in red raspberries to 90 mg/g in black raspberries (**Figure 3**). The free ellagic acid level was ~40−50% of the total ellagic acid present in all berries except boysenberries. Ellagic acid was present primarily as the free form in boysenberries; thus, there was little change in concentration after hydrolysis. There has been a great deal of interest in ellagic acid as a potential anticarcinogen (*29*); however, published data on the ellagic acid content of various foods are limited. Strawberries, raspberries, and thornless blackberries (*Rubus eubatus*) have been reported to be good sources of ellagic acid (*30*−*32*). In addition to these berries, we found that black raspberries, marionberries, evergreen blackberries, and boysenberries were also rich sources.

We also found that boysenberries were unique in that essentially all of the total ellagic acid was present as free ellagic acid. Ellagic acid exists primarily as the high molecular weight form of ellagitannins in food; thus, food samples must be hydrolyzed to yield free ellagic acid so that total levels of ellagic acid can be measured (*33*). This was the situation in the other berries, where the total level of ellagic acid detected was at least twice the level measured prior to hydrolysis.

There is very little information on the absorption and metabolism of ellagitannins in man. In vitro studies with rat intestinal contents have shown that ellagitannnins can be hydrolyzed to ellagic acid at the pH found in the small intestine and cecum but not in the stomach. It is possible that the microflora of the cecum may also participate in the hydrolysis (*34*). Studies with ellagic acid rather than ellagitannins have shown that 10% of the dose given to rats was absorbed and excreted as a metabolite in urine and feces (*35*), whereas mice given higher doses showed absorption rates of ~28% (*36*). Thus, if ellagitannins must be hydrolyzed prior to absorption, boysenberries may be both high in total ellagic acid and available ellagic acid as well.

Table 5. Summary of the Oligomers from Red Raspberries

oligomer	RT (min)	molecular ion $[M-H]^-$	MW	concentration[a] (mg/g)
monomer 1	11.73	289	290	0.02 (0.14)
monomer 2	12.06	433	434	0.02 (0.14)
dimer 1	17.57	561	562	0.03 (0.21)
dimer 2	19.35	577	578	0.03 (0.21)
trimer 1	21.31	833	834	0.01 (0.07)
trimer 2	22.83	849	850	0.01 (0.07)
trimer 3	24.37	865	866	0.01 (0.07)

[a] Based on the wet weight. Data in parentheses expressed per gram of dry matter.

We were able to measure oligomeric procyanidins from monomers to hexamers, but only red raspberries had measurable levels of procyanidins. There was 0.03 mg/g of trimers, 0.06 mg/g of dimers, and 0.04 mg/g of monomers (**Figure 4** and **Table 5**). As far as we know these are the only data on the procyanidin content of caneberries. Because the whole berries were homogenized and analyzed, it is possible that these procyanidins could have been present in the raspberry seeds. Grape seeds are also a good source of procyanidins. Recently, raspberry seeds have been shown to be a good source of unsaturated fatty acids and vitamin E (*37*). Future research will look at seeds separately to see if they are also a good source of procyanidins.

Although specific levels of anthocyanins and phenolics may vary depending on factors such as ripeness and cultivar (*3, 12*), our study found that caneberries grown in Oregon, specifically evergreen blackberries, marionberries, red raspberries, black raspberries, and boysenberries, had high antioxidant activity and

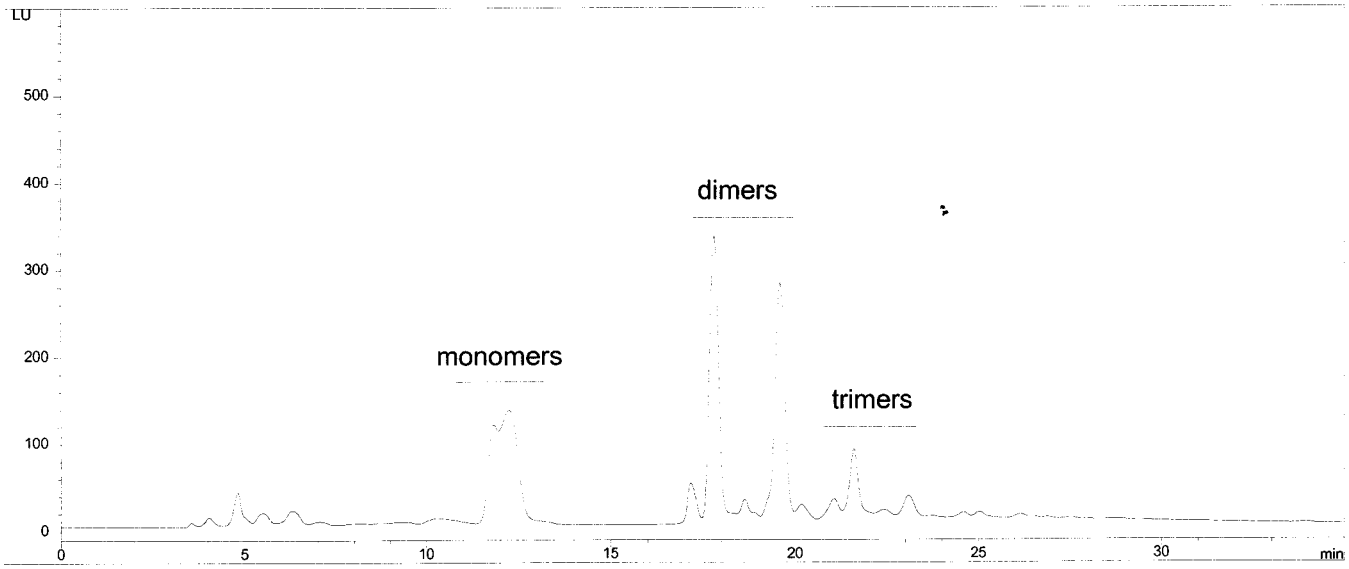

Figure 4. Procyanidin profile of red raspberries using HPLC coupled with fluorometric detection.

were rich in anthocyanins and phenolic compounds. Thus, caneberries can be an important part of a healthy diet.

ABBREVIATIONS USED

ORAC, oxygen radical absorbance capacity; Trolox, 6-hydroxy-2,5,7,8-tetramethyl-2-carboxylic acid; AAPH, 2,2′-azobis-(2-amidinopropane) dihydrochloride; TE, Trolox equivalent.

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