ISSN 1330-9862
(FTB-2060)

original scientific paper

Polyphenol, Anthocyanin and Resveratrol Mass Fractions and Antioxidant Properties of Cranberry Cultivars

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Received: February 20, 2008
Accepted: July 28, 2008

Summary

The study involved an evaluation of five cranberry cultivars grown in Poland: Ben Lear, Pilgrim, Stevens, Early Richard and Bergman. The reference sample comprised wild-grown common cranberry (*Vaccinium oxycoccus*). The mass fractions of total phenolic compounds, anthocyanins and resveratrol (HPLC-DAD), as well as the antioxidant properties ($DPPH^{\cdot}$, $^{\cdot}OH$ and $ABTS^{+}$ radical scavenging capacity) were determined. Statistically significant differences ($p<0.05$) were reported as regards the mass fractions of polyphenols and anthocyanins in the fruit of the analysed cultivars. The highest polyphenol mass fraction was determined in Ben Lear (374.2 mg per 100 g of fresh mass), while Early Richard was the richest source of anthocyanins (77.1 mg per 100 g of fresh mass). The fruit of common cranberry contained the highest quantities of resveratrol (712.3 ng/g of fresh mass), and its mass fraction in the investigated cultivars ranged from 533.4 (cv. Stevens) to 598.2 ng/g of fresh mass (Ben Lear). Common cranberry was also marked by the highest $ABTS^{+}$ scavenging capacity. Stevens and Pilgrim were characterised by a strong capability to scavenge $DPPH^{\cdot}$ and $^{\cdot}OH$ free radicals. Statistically significant differences ($p<0.05$) were observed in respect of the free radical scavenging capacity of most investigated cranberry cultivars.

Key words: cranberry, polyphenols, anthocyanins, resveratrol, antioxidant properties, $DPPH^{\cdot}$, $^{\cdot}OH$ and $ABTS^{+}$ scavenging activity

Introduction

In botanical terms, cranberry is a wild, evergreen dwarf shrub of the family Ericaceae which grows in marshy coniferous forests and bogs. Common cranberry (*Vaccinium oxycoccus*) and the similar looking small cranberry (*Oxycoccos microcarpus*) are evergreen dwarf shrubs with small, narrow leaves and red edible fruit (*1,2*). American cranberry (*Vaccinium macrocarpon*) is a major commercial crop in eastern Canada and north-eastern USA, mainly in Massachusetts, New Jersey, Oregon, Washington and Wisconsin (*3*). Due to its documented medicinal properties, cranberry fruit has become a popular farming product in other countries, including Poland, where until now, cranberry has been farmed commercially only in the central parts of the country (*2*).

Cranberry fruit is a rich source of bioactive components with a broad spectrum of activities. It is particularly known for its antioxidant, anti-inflammatory and antimicrobial properties, which inhibit the growth of pathogenic bacteria such as *Escherichia coli*, *Helicobacter pylori* and other pathogens (*3–5*). For many years, cranberry juice has been a popular folk remedy used espe-

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cially in North America in the treatment of urinary tract infections in women and digestive tract ailments. The anticancer properties of cranberry made it a popular diet component in the prevention of neoplastic diseases (*6,7*).

Flavonoids, anthocyanins, proanthocyanidins, phenolic acids and vitamin C are cranberry fruit compounds which are marked by high biological activity (*8–10*). Recent research points to the particularly high biological activity of resveratrol (*5,11–13*). It is believed that resveratrol is produced by plants in response to stress caused by the environmental pollution, UV radiation, bacterial and fungal infections (*14*). The presence of resveratrol in cranberry and other berry fruits of the genus *Vaccinium*, such as myrtle whortleberry, red whortleberry, and red currant, has been documented in the work of, among others, Häkkinen *et al.* (*15*), Ehala *et al.* (*16*), and Rimando and Barney (*17*). According to Lyons *et al.* (*18*), resveratrol mass fraction in berry fruit is determined largely by the farming region. The above mentioned researchers did not find resveratrol in highbush blueberries in British Columbia. Resveratrol may have a beneficial effect on the circulatory system by inhibiting lipid peroxidation (LDL), preventing blood platelet aggregation and expanding blood vessels (*10,19*). The above mentioned properties are largely the result of antioxidant activity of resveratrol, its ability to neutralise free radicals and induce the production of enzymes responsible for the detoxication of xenobiotics, *e.g.* quinone reductase. Due to a broad spectrum of properties, resveratrol can be classified as a multi-functional compound (*12,14,20*). Synergism may be an important feature of the biological activity of polyphenols: synergistic interactions between wine polyphenols, quercetin and resveratrol were found to decrease the inducible nitric oxide synthesis activity in the cell culture system (*21,22*).

The broad spectrum of biological activities manifested by cranberry fruit prompted the initiation of a study determining whether the Polish climate offers a favourable environment for farming cranberry varieties, which are popularly grown in other geographic regions. The evaluation criteria adopted in this study include the content of total phenolic compounds, anthocyanins and resveratrol as well as the antioxidant properties of cranberry fruit. The reference sample comprised wild cranberry fruit.

Materials and Methods

Fruit

The investigated material comprised five American cranberry cultivars: Pilgrim, Ben Lear, Stevens, Early Richard and Bergman, grown on the Experimental Whortleberry Farm of the Department of Pomology at the Warsaw University of Life Sciences. Common cranberry was harvested in forests of the Olsztyn area. In Poland, cranberry is grown in wet peaty places with high atmospheric moisture, under moderate climate conditions. Farm-grown and wild cranberry fruit were harvested in October 2006. The fruit was frozen and cold-stored (Caravell 0601) at a temperature of –25 °C for 5 to 6 weeks prior to analyses.

Chemicals and standards

Cyanidin-3-glucoside was obtained from Polyphenols Laboratories AS (Sandnes, Norway). The set, comprising a phosphate buffer, chromogen (metmyoglobin, ABTS), substrate (hydrogen peroxide) and the standard (Trolox), was purchased from the Randox Laboratories Ltd., UK. Solvents for HPLC (LabScan, Dublin, Ireland) and redistilled water (Mili-Q, Milipore, El Passo, TX, USA) were employed for the conditioning of the extractive column, analyte elution and the chromatographic analysis (mobile phases). Folin–Ciocalteu reagent and other chemicals were from Merck (Darmstadt, Germany). Trolox, resveratrol, gallic acid standard and DPPH (2,2'--diphenyl-1-picrylhydrazyl) were purchased from Sigma--Aldrich Fluka Co. (St. Louis, MO, USA). All other chemicals were of analytical grade and from ABCHEM (Poland).

Sample preparation

Six samples (10 g) for each of the six cranberry cultivars: wild, Ben Lear, Bergman, Early Richard, Pilgrim and Stevens, were thawed at room temperature (20 °C) for 15 min and were crushed with the use of an Ultra Turrax homogenizer (IKA Works do Brasie Ltd., Germany). The disintegrated samples were extracted three times with 80 mL of methanol solution containing 0.1 % hydrochloric acid. The extract was centrifuged (4000 rpm, 15 min) and the clear supernatant was collected. Methanolic extracts were concentrated under reduced pressure at 40 °C for the determination of total phenolic compounds, total anthocyanins and $DPPH^{\cdot}$, $^{\cdot}OH$ and $ABTS^{+}$ free radical scavenging capacity. The crushed fruit samples were extracted with the use of an ethanol and water mixture (20:80, by volume) to determine resveratrol mass fraction. Extraction took place within 30 min in an ultrasound bath (InterSonic IS-5.5, Beverage Processing Machinery, Olsztyn, Poland) at 60 °C (*23*).

Determination of total phenolic compounds

Folin-Cioccalteu reagent was used to determine total phenolic compounds (*23*). A volume of 1 mL of cranberry extract, diluted 5–6 times with methanol (to obtain absorbance within the range of the prepared calibration curve), was mixed with 0.5 mL of Folin-Ciocalteu reagent previously diluted with distilled water (1:2). A volume of 1.5 mL of 20 % sodium carbonate solution was added to the mixture, shaken thoroughly and diluted to 10 mL by adding distilled water. The mixture was let to stand for 90 min and the blue colour formed was measured at 765 nm with a spectrophotometer (UV/VIS Spectrometer UNICAM, UK). Gallic acid was used as a standard for the calibration curve. The concentrations of gallic acid in the solution from which the curve was prepared were 0, 50, 100, 150, 250 and 500 mg/L. The total mass fraction of phenolic compounds was calculated and expressed as gallic acid equivalent (GAE)/(mg/100 g).

Determination of anthocyanin mass fraction

Total anthocyanins were determined as described by Wrolstad (*24*) and expressed as cyanidin-3-glucoside, which is the dominant anthocyanin in cranberry. The

prepared extract was sampled in the required quantity in 10-mL measuring flasks, and was supplemented with 80 % methanol solution. A volume of 1 mL was sampled, 4 mL of buffer with pH=1 (120 mL of 0.2 mol/L of KCl and 390 mL of 0.2 mol/L of HCl) were added, and absorbance was measured at a wavelength of 502 nm (for cyanidin-3-glucoside) against the buffer with pH=1 as the reagent sample. The resulting value should be within the 0.4–0.6 range. Absorbance was measured also at 700 nm to eliminate disturbances. Successive read-outs were taken at the wavelengths of 502 and 700 nm for the samples prepared in the above manner, but the buffer with pH=1 was replaced with a buffer with pH=4.5 (450 mL of 1 M sodium acetate, 220 mL of 1 M HCl and 330 mL of distilled water). The measurement was compared with the blank sample comprising a buffer with pH=4.5. Anthocyanin mass fraction was expressed as the equivalent of cyanidin-3-glucoside.

Determination of resveratrol

The solid-phase extraction of *trans*-resveratrol was carried out on Bakerbond Speedisk® DVB cartridges (200 mg) purchased from J.T. Baker (Deventer, Holland) and preconditioned by washing with 5 mL of methanol and 5 mL of water. After passing the sample (5 mL) through the cartridge, subsequent washing steps were performed using 5 and 10 mL of deionized water. The cartridge was then dried for 15 min by a constant flow of nitrogen. The absorbed resveratrol was eluted with 2×2.5 mL of methanol.

Analyses were taken using an HP1100 liquid chromatograph (Agilent Technologies, Waldbronn, Germany) equipped with a diode array detector and an autosampler. Detection was carried out by monitoring absorbance signals at 306 nm. The separations were performed on an ACE C18 column (150×4 mm i.d., d_p=3 μm). The analyses were performed at 40 °C and the flow rate was 0.6 mL/min. Solvent A was acetonitrile and solvent B was a mixture of water/acetic acid/acetonitrile (87:3:10 % by volume). Elution employed a linear gradient from 5 to 25 % solvent A for 25 min, then 5 % for 5 min, held for 20 min to wash the column and followed by a return to the initial conditions (5 % solvent A) for 10 min. Aliquots were filtered through a 0.2-μm Nylon Millipore chromatographic filter and injected into the chromatograph. Injection volume was 20 μL (*25*). Identification was achieved by comparing t_r (retention time) and the absorption spectra obtained for the eluted peak with those obtained for the standards.

For quantification, an external standard calibration curve was plotted, ranging from 0.50 to 10.02 mg/L of *trans*-resveratrol. The square regression coefficient of analytical curve was near unit (R^2=0.9999). The limit of detection (LOD) and the limit of quantification (LOQ) were determined based on the detector's signal-to-noise (S/N) ratio. The standard deviation of the S/N was calculated and multiplied by a factor of 3, then this value was added to the average of the S/N to obtain the LOD. LOQ was defined as 10 S/N. At 306 nm, the LOD and LOQ for *trans*-resveratrol were 0.2 and 0.75 μg/mL, respectively, from the current calibration curve.

Determination of DPPH· scavenging activity

The 2,2'-diphenyl-1-picrylhydrazyl radical (DPPH·) scavenging activity was determined by the procedure described by Brand-Williams *et al.* (*26*). A volume of 0.1 mL of the investigated extract was sampled, 3.9 mL of 0.25 % DPPH methanol solution were added, and absorbance was measured at a wavelength of 515 nm with the use of a spectrophotometer (UV/VIS Spectrometer UNICAM, UK) until constant values were obtained. A blank was prepared for each sample using methanol instead of the DPPH solution. The content of the residual DPPH· was calculated for every antioxidant concentration in the investigated samples in line with the following formula:

$$\text{Residual DPPH}^{\cdot}=(A_{sample}/A_{control})\times 100 \qquad /1/$$

A curve illustrating the dependency between the content of the residual DPPH· and the mass of the sample (mg of fruit) was plotted to determine the EC_{50} coefficient. This coefficient determines the mass of fruit (mg) which is required to reduce the initial synthetic concentration of DPPH· in the reaction by 50 %. The obtained results were expressed as μmol of Trolox per g of fruit fresh mass. The calibration curve was constructed by preparing different concentrations of Trolox (0–30 μmol/L), handled as the investigated samples.

Determination of ·OH radical inhibition

The scavenging of the hydroxyl radical (·OH) was measured by the deoxyribose method (*27*). A volume of 0.1 mL of the investigated extract was sampled and the following reagents were added in the indicated order: 0.69 mL of phosphate buffer containing 2.5 mM deoxyribose, 0.1 mL of ammonium iron sulphate containing ethylenediaminetetraacetic acid (EDTA), 0.1 mL of ascorbic acid solution and 10 μL of hydrogen peroxide solution. The mixture was incubated in a water bath for 10 min at 37 °C. After incubation, 1 mL of 2.8 % trichloroacetic acid and 0.5 mL of thiobarbituric acid were added. The mixture was heated in a boiling water bath for 8 min, then it was cooled and absorbance was measured at 532 nm with a spectrophotometer (UV/VIS Spectrometer UNICAM, UK). The reagent sample containing distilled water instead of the investigated extract was prepared simultaneously. The obtained results were expressed as μmol of Trolox per g of fresh mass. The calibration curve was prepared by measuring the absorbance of the samples that had an adequate standard substance concentration. Trolox 2.5 mM was prepared in 5 mM phosphate buffered saline (PBS), pH=7.4, to be used as a stock standard.

Determination of $ABTS^+$ free radical scavenging capacity

The analysis was performed with the use of a set supplied by Randox Laboratories Ltd., UK, comprising a phosphate buffer, chromogen (metmyoglobin, ABTS), substrate (hydrogen peroxide) and the standard (Trolox). Solutions containing 1 mg of total phenolic compounds per mL of methanol were prepared for the determination of total antioxidant activity (TAA). The reagent, standard and proper samples were prepared. A volume

of 0.2 mL of the analysed phenolic compound solution and 1 mL of chromogen were added to the investigated sample and the first absorbance measurement was performed. Then, 0.02 mL of deionised water and 0.02 mL of the standard solution were added to the reagent and to the standard sample, respectively, and absorbance measurement was carried out. After that, 0.2 mL of the substrate were added to all the samples and incubated at 37 °C. Absorbance was measured again at 734 nm and the read-out was taken within 6 min. TAA was expressed as μmol of Trolox per g of fresh mass.

Statistical analyses

All analyses were performed in six replications, rejecting two extreme values (N=4). Tables 1 and 2 contain the mean values. The significance of differences between the mean values was estimated by Duncan's test. Statistical analysis was performed at a significance level of $p<0.05$. The correlation analysis was conducted using Statistica v. 8.0 software.

Table 1. Mass fractions of phenols, anthocyanins and resveratrol in the cranberries

Cranberry cultivars	*w*(phenol)* mg/100 g fresh mass	*w*(anthocyanin)** mg/100 g fresh mass	*w*(resveratrol) ng/g fresh mass
Wild cranberry	(288.5±0.1)^{d}	(43.3±0.1)^{a}	(712.3±0.1)^{a}
Ben Lear	(374.7±0.1)^{b}	(51.9±0.1)^{e}	(598.2±0.3)^{b}
Bergman	(292.1±0.1)^{c}	(73.0±0.2)^{b}	(552.8±0.3)^{c}
Early Richard	(328.8±0.2)^{a}	(77.2±0.2)^{a}	(536.4±0.3)^{d}
Pilgrim	(192.1±0.1)^{e}	(59.9±0.2)^{c}	(537.2±0.2)^{d}
Stevens	(293.9±0.1)^{c}	(54.6±0.2)^{d}	(533.4±0.1)^{e}

Results are mean values±SD (N=6), $p<0.05$; values in the same column followed by the same letter (a–e) are not statistically different ($p<0.05$) as measured by Duncan's test
*expressed as gallic acid equivalent
**expressed as cyanidin-3-glucoside equivalent

Table 2. $DPPH^{\cdot}$, $^{\cdot}OH$ and $ABTS^{+}$ radical scavenging capacity of cranberry fruit

Cranberry cultivars	$DPPH^{\cdot}$ μmol TE per g of fresh mass	$^{\cdot}OH$ μmol TE per g of fresh mass	$ABTS^{+}$ μmol TE per g of fresh mass
Wild cranberry	(36.9±0.1)^{e}	(0.9±0.0)^{c}	(16.4±1.2)^{a}
Ben Lear	(33.9±0.1)^{f}	(0.8±0.1)^{d}	(13.0±1.2)^{d}
Bergman	(42.1±0.1)^{c}	(0.8±0.0)^{d}	(13.1±1.2)^{d}
Early Richard	(41.8±0.2)^{d}	(1.0±0.0)^{b}	(14.6±1.4)^{c}
Pilgrim	(66.0±0.1)^{b}	(0.9±0.1)^{c}	(9.3±1.2)^{e}
Stevens	(68.8±0.1)^{a}	(1.1±0.0)^{a}	(15.4±1.3)^{b}

Results are mean values±SD (N=3), $p<0.05$; values in the same column followed by the same letter (a–f) are not statistically different ($p<0.05$) as measured by Duncan's test
TE – Trolox equivalent
EC_{50} – product mass required to reduce the $DPPH^{\cdot}$ radical by 50 %; the measurement was conducted with the use of a DPPH solution at a concentration equivalent to 0.025 g/L methanol (the studied reaction mixture contained 0.1 mL of the investigated sample and 3.9 mL of DPPH solution)

Results and Discussion

Polyphenol, anthocyanin and resveratrol mass fractions in the fruit of different cranberry cultivars

The mass fractions of total phenolic compounds in the fruit of five investigated American cranberry cultivars ranged from 192.1 in cv. Pilgrim to 374.2 mg per 100 g of fresh mass in cv. Ben Lear expressed as GAE (Table 1). In the reference sample of wild-grown common cranberry fruit, polyphenol mass fraction reached 288.5 mg per 100 g of fresh mass. Statistically significant differences ($p<0.05$) were reported in the mass fraction of polyphenols of all the studied varieties, including common cranberry (Table 1). Wang and Stretch (*28*) recorded smaller quantities of polyphenols in a study investigating several cranberry varieties grown at the Rutgers Blueberry and Cranberry Research Center in Chatsworth (NJ, USA). These authors found that the mass fraction of total phenols expressed as gallic acid equivalent in Pilgrim, Stevens and Ben Lear cultivars was lower, *i.e.* 120.0, 126.0 and 137.5 mg per 100 g of fresh mass, respectively. Polyphenol mass fractions similar to those determined in the present study in different cranberry varieties were reported by Kalt *et al.* (*29*) and Borowska *et al.* (*9*). According to these authors, the average polyphenol mass fraction of cranberry fruit was 2010 mg per 100 g of dry mass.

The investigated cranberry fruit also differed with regard to anthocyanin mass fraction (Table 1). The fruit of the five investigated cultivars was characterised by a higher anthocyanin mass fraction, compared to common cranberry. The highest anthocyanin mass fraction of 77.1 mg per 100 g of fresh mass was reported for Early Richard, while the lowest mass fraction of 52.1 mg per 100 g of fresh mass was observed in Ben Lear. The anthocyanin mass fraction of the reference sample of wild cranberry fruit was lower, 43.4 mg per 100 g of fresh mass. According to Wang and Stretch (*28*), American cranberry fruit is marked by even more pronounced differences in anthocyanin mass fraction, ranging from 19.8 to 65.6 mg per 100 g of fresh mass. The above mentioned authors studied fresh cranberry fruit of Ben Lear, Pilgrim and Stevens cultivars grown at the Rutgers Blueberry and Cranberry Research Center in Chatsworth (NJ, USA) and found that their anthocyanin mass fraction reached 25.0, 20.7 and 22.8 mg per 100 g of fresh mass, respectively. The anthocyanin mass fraction of cranberry fruit studied by Kalt *et al.* (*29*) was determined at 3.1 mg/g of dry mass. The results obtained in this study and the results reported by other authors indicate that in addition to genetic factors, the mass fraction of phenolic compounds in cranberry fruit is also significantly affected by the applied cultivation technology. Similarly to polyphenols, the differences in anthocyanin mass fraction in all the cranberry varieties inves-

tigated in this study, including common cranberry, were statistically significant at $p<0.05$.

Due to the important role played by resveratrol, as postulated by many authors (*11*,*14*,*20*), its mass fraction was determined by HPLC in this study. It was demonstrated that all of the investigated cranberry cultivars contained less resveratrol than wild cranberry fruit, whose resveratrol mass fraction was estimated at 712.3 ng/g of fresh mass.

In the group of the studied cranberry cultivars, high resveratrol mass fraction of 598.2 ng/g of fresh mass was reported for Ben Lear (Table 1, Fig. 1). In terms of dry mass, the above mentioned results are indicative of resveratrol mass fraction of 5.846 and 4.460 μg/g. The differences in the resveratrol mass fraction of the five investigated cranberry cultivars were not statistically significant at $p<0.05$. Statistically significant differences ($p<0.05$) were observed between the analyzed cultivars and the wild cranberry. It should be noted that the investigated cranberry varieties are marked by a high resveratrol mass fraction, which is comparable to that of lingonberry (5.80 mg/g of the dry sample) and grapes (6.50 mg/g of the dry sample), as demonstrated by Rimando *et al.* (*30*).

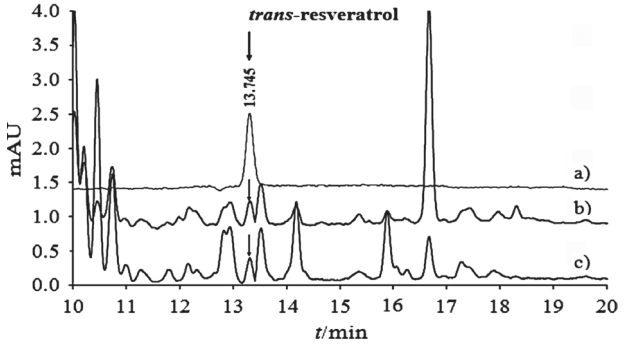

Fig. 1. HPLC-DAD chromatograms recorded at 306 nm of (a) *trans*-resveratrol standard solution at the concentration of 0.5 μg/mL, (b) the solid phase extracted wild cranberry and (c) Bergman cranberry samples. Chromatographic conditions are described in the text

Antioxidant properties of different cranberry cultivars

According to numerous researchers, phenolic compounds, their content and qualitative composition are largely responsible for the antioxidant properties of fruit (*31*,*32*). In the current study, the antioxidant properties of cranberry were determined in view of the $DPPH^{\cdot}$, $^{\cdot}OH$ and $ABTS^{+}$ radical scavenging capacity of fruit methanol extracts (Table 2). The obtained results, expressed in terms of Trolox equivalent (TE), indicate that cranberry fruit, regardless of cultivar, is characterised by the highest $DPPH^{\cdot}$ scavenging capacity within the range of 33.87–68.83 μmol of TE per g of fresh mass. The lowest scavenging capacity, within the range of 0.8–1.04 μmol of TE per g of fresh mass, was reported in respect of OH radicals. High scavenging capacity of all three types of free radicals ($DPPH^{\cdot}$, $^{\cdot}OH$ and $ABTS^{+}$) was attributed to cv. Stevens, followed by cv. Pilgrim. Cultivar Bergman was marked by lower antioxidant activity. The highest $ABTS^{+}$ radical scavenging capacity of 16.42 μmol of TE per g of fresh mass was reported for the comparatively analysed common cranberry. The differences in the antioxidant activity of the investigated samples were statistically significant at $p<0.05$ with two exceptions.

The results of this study were compared with those obtained by Wang and Stretch (*28*), who determined the antioxidant properties of cranberry grown at the Rutgers Blueberry and Cranberry Research Center in Chatsworth (NJ, USA) by the ORAC method. In the group of 10 analysed cranberry varieties, Stevens, Pilgrim and Ben Lear were characterised by mean ORAC values of 9.1, 8.2 and 10.1 μmol of TE per g of fresh mass, respectively. During an earlier research investigating five cranberry cultivars, Wang and Jiao (*4*) had concluded that cv. Ben Lear had an average scavenging capacity of active oxygen species ($O_2^{\cdot -}$, H_2O_2, $^{\cdot}OH$ and O_2). According to Kalt *et al.* (*29*), the $DPPH^{\cdot}$ scavenging capacity of the studied cranberry varieties reached 92.9 TE per g of dry mass. The free radical scavenging capacity of other berry fruit species, such as cowberry, black currant and bilberry, was two- to threefold higher.

Conclusions

The results of the study indicate that the fruit of the analysed cranberry cultivars grown in Poland differs with regard to polyphenol, anthocyanin and resveratrol mass fractions as well as antioxidant properties. In most cases, statistically significant differences at $p<0.05$ were reported. Despite the observed variations, the mass fractions of the analysed bioactive compounds in the investigated cultivars and their antioxidant properties were comparable to the results quoted by other authors. The highest mass fraction of total phenolic compounds, including resveratrol, was recorded in the fruit of Ben Lear, while Early Richard was the richest source of anthocyanins. As regards the antioxidant activity of particular cranberry cultivars, the highest scavenging capacity was observed in respect of $DPPH^{\cdot}$ radicals. Cultivars Stevens and Pilgrim were marked by high scavenging capacity of all the analysed free radicals – $DPPH^{\cdot}$, $^{\cdot}OH$ and $ABTS^{+}$.

Acknowledgements

This work was partly supported by the University of Warmia and Mazury in Olsztyn (Program 528-0709-808).

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