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## **FRUIT QUALITY OF HIGHBUSH BLUEBERRY (*VACCINIUM CORYMBOSUM* L.) CV. 'DUKE' DEPENDING ON THE METHOD OF CULTIVATION**

## **JAKOŚĆ OWOCÓW BORÓWKI WYSOKIEJ (*VACCINIUM CORYMBOSUM* L.) ODMIANY 'DUKE' W ZALEŻNOŚCI OD SPOSOBU UPRAWY**

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**Streszczenie.** W Polsce obserwuje się wzrost zainteresowania konsumentów żywnością ekologiczną. Płody rolne z gospodarstw ekologicznych są postrzegane jako produkty zdrowsze i zasobniejsze w substancje bioaktywne. W pracy określono wpływ sposobu uprawy krzewów borówki wysokiej odmiany 'Duke' (konwencjonalnej i ekologicznej) na jakość plonu: wielkość owoców, ich jędrność i skład chemiczny. Badania przeprowadzono w latach 2009–2011 w Pracowni Sadownictwa oraz w gospodarstwie produkcyjnym. Owoce z plantacji ekologicznej były większe, w plonie ogólnym było również więcej owoców dużych (> 12 mm). Owoce te były jednak mniej jędrne oraz charakteryzowały się mniejszą odpornością na uszkodzenia mechaniczne. Jagody zebrane z krzewów uprawianych konwencjonalnie charakteryzowały się większą zawartością ekstraktu i azotanów. Niezależnie od sposobu uprawy owoce małe (< 12 mm) były bardziej jędrne (zarówno w osi wysokości, jak i średnicy) oraz zawierały więcej ekstraktu. Owoce małe oraz zebrane z krzewów uprawianych konwencjonalnie charakteryzowały się również większą zawartością związków polifenolowych, zwłaszcza antocyjanów i flawonów. Owoce małe zawierały również więcej kwasu chlorogenowego.

**Key words:** chemical composition, firmness, fruit size, organic and conventional farming, polyphenols.

**Słowa kluczowe:** jędrność, polifenole, skład chemiczny, uprawa ekologiczna i konwencjonalna, wielkość owoców.

## **INTRODUCTION**

An increase in ecological food is currently observed. The consumer awareness increases, which results in a growing number of ecological farms (Meier-Ploeger 2005). Products from ecological farms are considered to be healthier and more nutritious (Saba and Messina 2003). Apart from the production of healthy food, ecological farming contributes to the protection of landscape and the natural environment (Milivojevic et al. 2012). Agriculture is based on the use of mineral and biological agents; the use of pesticides or synthetic fertilizers is not allowed (Popławski 2009), whereas the use of natural fertilizers, manure, compost and green manure is acceptable. Natural methods of protection from pathogens in the form of plant

extracts are also allowed (Rembiałkowska et al. 2006). Fruits obtained from ecological cultivations contain a larger number of beneficial bioactive compounds – flavonoids, anthocyanins and vitamin C, which have strong antioxidant properties. They are also characterized by a higher content of micro- and macronutrients (Rembiałkowska et al. 2003), organic acids and sugars (Hallmann and Rembiałkowska 2007b).

Conventional farming uses industrial production agents, such as artificial fertilizers and pesticides, which can have a disadvantageous impact on the natural environment (Kuś 1995). Excessive nitrogen fertilization leads to the expansion of green parts of plants (Premuzic et al. 1998). The cultivation is targeted at maximization of profits with low labour intensity and high consumption of industrial products (Kuś and Fotyma 1992). In conventional agriculture, fruits have lower vitamin C content and the fruit skin contains less anthocyanins and flavonoids than in ecological cultivations (Wojdyło et al. 2010).

However, not all plants are equally appropriate for such growing methods. Highbush blueberry cultivars are derived from the genus *Vaccinium*, which grows wild in soils characterized by a low nutrient content. As a result, fertilizing requirements of highbush blueberry are relatively modest, as compared with other fruit crops (Pliszka 2002; Pormale et al. 2009). Furthermore, these plants do not require intensive protection from diseases and pests. Spring frost and birds are the greatest problem, as they may considerably reduce the yield. Bushes are also characterized by specific habitat and soil requirements, different from other cultivated fruit-bearing plants. Acidic peat soils (pH 3.5–4.0) are the best for cultivation, but light sandy soils can also be useful. It is important that soils should have a stable moisture level as roots are sensitive to both the excess and deficiency of water (Moore 1993).

The study determines the method of highbush blueberry cultivation and the size of fruits on their physical parameters and chemical composition, in particular, taking into account polyphenolic compounds.

## MATERIAL AND METHODS

The research was conducted at the Fruit Farming Laboratory and at a certified production farm specializing in the cultivation of highbush blueberry. The plantation consists of an ecological (certified) 40 ha plot situated in the Goleniów Forest in the immediate vicinity of an abandoned peat mine. Natural acidic peat has a thickness of 150–200 cm. A conventional plantation (60 ha), situated in the immediate vicinity of forest was established on sandy soils in which a natural process of acidification occurred as a result of long-term fallow. Plants were planted on raised beds covered with strips of nonwoven fabric for nurseries, which protects them from weeds. The pH of the soil is too high for highbush blueberry requirements on the conventional plantation (Table 1); water used for watering plants by means of a drip-line system placed under the nonwoven fabric was acidified with sulphuric acid to a pH of 4.0–4.2.

The plants were planted with a spacing of 2.0 x 1.2 m, grass grows in the inter-rows. The mineral content in the soil at both plantations ranged from a medium to high level (Table 1). In spring on conventional plantation the nitrogen fertilization was applied at a dose of 45 kg N · ha<sup>-1</sup>.

Table 1. The pH and the content of minerals in the soil  
Tabela 1. Odczyn pH i zawartość substancji mineralnych w glebie

pH	Content – Zawartość [mg · 100 g <sup>-1</sup> ]				
	P	K	Mg	Ca	N-NO <sub>3</sub>
Ecological plantation – Plantacja ekologiczna					
3.4 optimal optymalne	6.1 high* wysoki	5.8 low niski	7.3 high wysoki	32.4	8.4
Conventional plantation – Plantacja konwencjonalna					
5.2 too high za wysokie	3.5 average średnia	15.8 high wysoki	12.1 high wysoki	191	2.3

\* Determined by the needs of fertilization developed by Sadowski et al. (1990) – Określono wg potrzeb nawożenia opracowanych przez Sadowskiego i in. (1990).

The experiment followed a randomised sub-block design (3 blocks, 15 plants in each block). Fruits were harvested manually from all the shrubs covered by the experiment, and prepared the aggregate sample on which measurements. Physical features of fruits (fruit size, firmness, puncture of the skin), soluble solids, titratable acidity and nitrates were measured on fresh berries immediately after the harvest were performed on fresh fruits. Phenolics composition was determined in fruit samples that were kept frozen (−32°C) in polyethylene bags (500 g) until analyzed.

The fruit weight was measured with RADWAG WPX 4500 electronic scales (0.01 g accuracy). Fruit diameter, firmness and puncture resistance of the skin was measured with a FirmTech2 apparatus (BioWorks, USA) of 100 randomly selected berries from three replicate was expressed as a gram-force causing fruit surface to bend 1 mm. Puncture were made using a stamp with a diameter of 3 mm.

Soluble solids content was determined with a digital refractometer PAL-1 (Atago, Japan). Titratable acidity was determined by titration of a water extract of blueberry homogenate with 0.1 N NaOH to an end point of pH 8.1 (measured with an multimeter Elmetron CX-732) according to PN-90/A-75101/04. Nitrates content was measured with a RQflex 10 reflectometer (Merck).

The HPLC analyses of polyphenols were carried out with HPLC apparatus consisting of a Merck-Hitachi L-7455 diode array detector (DAD) and quaternary pump L-119 7100 equipped with D-7000 HSM Multisolvant Delivery System (Merck-Hitachi, Tokyo, Japan). The runs were monitored for phenolic acids at 320 nm, flavonols and luteolin glucoside at 360 nm, and anthocyanin glycosides at 520 nm. Retention times and spectra were compared to that of pure standards and total polyphenols content was expressed as mg per 100 g fruit tissue. Standards of anthocyanidin glycosides were obtained from Polyphenols Laboratories (Norway), while, for phenolic acids, flavonols and from Extrasynthese (France).

In order to determine the significance of differences, a two-factor analysis of variance was carried out, followed by the assessment of the significance of differences at  $P < 0.05$  using the Tukey's test. The statistical analyses were performed using the Statistica software.

## RESULTS AND DISCUSSION

The production of fruits using ecological methods constitutes a smaller burden on the natural environment. It should also have a positive influence on the quality of fruits, their size and chemical composition. On the basis of the tests performed, fruits from the Duke cultivar collected at ecological plantations were larger, the average weight of 100 fruits was 180 g, and the weight of 100 fruits collected from conventionally cultivated fruits was 116 g. In Poland, it is assumed that commercial fruits should have a diameter of 12 mm (Smolarz 2009), which is why they were calibrated. The share of large fruits, which was over 12 mm, was over 63% in the total yield obtained at the ecological plantation, and the average weight of 100 fruits was 262 g. In the total yield obtained at the conventional plantation, there were fewer large fruits (40%), they were also lighter – 100 fruits weighed 171 g (Table 2). Fruits from this cultivar may weigh even up to 343 g (Ochmian 2012), while in the research by Ścibisz and Mitek (2007), one fruit weighed 1.5 g on average.

Table 2. The influence of the cultivation method on the quality of fruits from the 'Duke' cultivar of the highbush blueberry

Tabela 2. Wpływ sposobu uprawy na jakość owoców borówki wysokiej odmiany 'Duke'

		Method growing – Sposób uprawy					
		ecological – ekologiczny			conventional – konwencjonalny		
Fruit size Wielkość owoców		< 12 mm	> 12 mm	mean średnia	< 12 mm	> 12 mm	mean średnia
Participation in the yield of fruit Udział w plonie owoców [%]		36.8	63.2	–	59.5	40.5	–
Weight of 100 fruits Masa 100 owoców [g]		98.3 a*	262 b	180 B**	61.6 a	171 b	116 A
Firmness Jędrność [G mm]	axis diameter oś średnicy	179 b	146 a	162 A	192 b	163 a	177 A
	height wysokość	367 b	335 a	351 A	421 b	384 a	402 B
Puncture axis diameter Siła przebicia osi średnicy [G mm]		91 b	78 a	84.5 A	136 b	104 a	120 B
Soluble solids Ekstrakt [%]		15.1 b	14.4 a	14.8 A	15.9 b	15.3 a	15.6 B
Titratable acidity Kwasowość [g · 100 mL <sup>-1</sup> ]		0.65 a	0.62 a	0.64 A	0.66 a	0.68 a	0.67 A
Nitrates Azotany [mg · 1000 mL <sup>-1</sup> ]		17.3 a	15.7 a	16.5 A	25.4 a	22.7 a	24.1 B

\* Means followed by the same letter do not differ significantly at P = 0.05 according to Tukey multiple range test – Liczby oznaczone tą samą literą nie wykazują istotnych różnic na poziomie  $\alpha = 0,05$ , według wielowymiarowego testu Tukeya.

\*\* The differences between method growing are indicated by capital letter, differences between fruit size are indicated by small letter – Różnice między sposobami uprawy oznaczono dużymi literami, a różnice w wielkości owoców – małymi literami.

Fruits from the conventional cultivation were characterised by a higher resistance to damage – 120 G mm. Higher values, from 153 G mm, were obtained in another experiment, but the fruits were sprayed with calcium fertilisers, which increased their resistance to damage (Ochmian 2012). Fruits collected at the conventional plantation were also firmer, especially along the height axis (402 G mm). This was undoubtedly influenced by the size of fruits, smaller fruits, regardless of the cultivation method, were characterised by greater firmness, both along the height axis and the diameter. Fruits from the Duke cultivar are characterised by lower firmness as compared to fruits from other cultivars (Ochmian et al. 2007), its values were similar to those obtained for fruits from this cultivar in the previous years (Ochmian et al. 2009b).

The research conducted shows differences in the chemical composition of fruits. Fruits from the ecological cultivation had a lower extract content (14.8%), but also the content of harmful nitrates was lower (16.5 mg) than in fruits from the conventional cultivation (Table 2). No differences between small and large fruits were found, on the other hand, in the nitrate content. A similar nitrate content was found in high-bush blueberry fruits cultivated on peat – 17.5 mg · kg<sup>-1</sup> (Ochmian et al. 2009a) and fruits tested by Ostrowska and Ściążko (1996). After the application of the Fruton calcium fertiliser, their content increased to 36.7 mg (Ochmian 2012). In Poland and other countries there is a lack of regulations on permissible nitrate content in fruits (except for bananas). According to the Polish Ministry of Agriculture (DzU 2003) the permissible nitrate content in vegetables meant for feeding babies and young children should not exceed 200 mg NaNO<sub>3</sub> kg<sup>-1</sup>. The fruit acidity was at a similar level, regardless of the cultivation method and the fruit size (from 0.62 to 0.68 g). The fruits from this cultivar were characterised by similar acidity in the research conducted by Duan et al. (2011) as well as by Ścibisz and Mitek (2007). The organic acid content, on the other hand, was slightly higher, as compared to berries from the Spartan cultivar (0.54 g), and it was lower than in fruits from the 'Blueray', 'Jersey' and 'Bluecrop' cultivars; 0.80–0.87 g (Skupień 2006).

Numerous studies confirm the fact that fruits from ecological cultivations contain more polyphenols as well as other antioxidant compounds (Caris-Veyrat et al. 2004, Hallmann and Rembiałkowska 2007a). There are also other contradictory opinions whether ecological foods have a nutritional advantage over their counterparts manufactured in a conventional manner (Riahi et al. 2009). This is reflected by the results obtained which are presented in Table 3. Fruits collected from bushes cultivated in conventional manner were characterised by a higher content of polyphenols (380 mg), as compared with ecological fruits (330 mg). Also, small fruits had a higher content of polyphenols, especially ones from the conventional cultivation, which was 422 mg. These values were higher than those obtained in the research by Wang et al. (2009). Prior et al. (1998) estimated phenolic content at 181–390 mg · 100 g<sup>-1</sup> in blueberry cultivars obtained from different sources.

Polypehols are secondarily synthesized products especially under stress conditions (De Gara et al. 2003, Hodges et al. 2004). At the ecological plantation, the soil and habitat conditions were very similar to natural sites of species from the genus *Vaccinium*. The soil ensured optimal conditions for root development, and the pH at 3.4 was perfect for this species (Table 1). Also, the content of mineral ingredients satisfied the needs of the bushes. The soil at the conventional plantation, on the other hand, was far from optimal, especially the pH was 5.2. This undoubtedly affected the size of the fruits and the stress caused by disadvantageous soil conditions may have influenced the chemical composition.

Table 3. The influence of the cultivation method on the polyphenolic compound content in fruits from the 'Duke' cultivar of the highbush blueberry [ $\text{mg} \cdot 100 \text{g}^{-1}$ ]

Tabela 3. Wpływ sposobu uprawy na zawartość związków polifenolowych w owocach borówki wysokiej odmiany 'Duke' [ $\text{mg} \cdot 100 \text{g}^{-1}$ ]

	Method growing – Sposób uprawy					
	ecological – ekologiczny			conventional – konwencjonalny		
	fruit size – wielkość owoców					
	< 12 mm	> 12 mm	mean średnia	< 12 mm	> 12 mm	mean średnia
Del-gal	30.43 a*	74.81 b	52.62 A**	98.89 b	86.34 a	92.62 B
Del-glu	43.84 b	23.02 a	33.43 B	9.78 a	11.09 a	10.44 A
Del-ara	21.95 b	10.13 a	16.04 A	53.92 a	45.09 a	49.51 B
Cya-gal	11.44 b	4.83 a	8.14 A	16.55 b	10.80 a	13.68 B
Cya-glu	15.69 b	7.98 a	11.84 A	9.30 a	5.07 a	7.19 A
Cya-ara	9.69 b	5.68 a	7.69 B	6.05 b	2.58 a	4.32 A
Pet-gal	4.58 b	2.32 a	3.45 A	16.75 a	15.77 a	16.26 B
Pet-glu	12.41 a	6.27 a	9.34 A	7.91 a	9.18 a	8.55 A
Pet-ara	29.85 b	16.61 a	23.23 B	5.26 b	1.29 a	3.28 A
Peo-gal	18.28 a	31.20 b	24.74 A	41.68 b	32.03 a	36.86 B
Peo-glu	9.37 a	3.78 b	6.58 A	24.39 b	17.87 a	21.13 B
Peo-ara	7.43 b	3.59 a	5.51 B	0.53 a	0.23 a	0.38 A
Mal-gal	10.53 b	5.67 a	8.10 B	1.26 b	0.09 a	0.68 A
Mal-glu	7.70 a	4.76 a	6.23 B	1.51 a	0.85 a	1.18 A
Mal-ara	23.65 a	23.29 a	23.47 B	4.47 a	16.67 b	10.57 A
Chlorogenic acid Kwas chlorogenowy	103.91 b	51.28 a	77.60 A	88.94 b	64.49 a	76.72 A
Que-gal	2.58 b	1.61 a	2.10 A	4.20 a	3.50 a	3.85 B
Que-glu	8.50 b	2.57 a	5.54 A	24.88 b	13.43 a	19.16 B
Que-ram	2.88 b	0.84 a	1.86 A	2.72 b	0.30 a	1.51 A
Keam-rut	3.32 b	1.06 a	2.19 A	3.04 b	1.05 a	2.05 A
Total – Suma	378 b	281 a	330 A	422 b	338 a	380 B

Del-gal – Delphinidin 3-O-galactoside, Del-glu – Delphinidin 3-O-glucoside, Del-ara – Delphinidin 3-O-arabinoside, Cya-gal – Cyanidin 3-O-galactoside, Cya-glu – cyanidin 3-O- glucoside, Cya-ara – Cyanidin 3-O-arabinoside, Pet-gal – Petunidin 3-O-galactoside, Pet-glu – Petunidin 3-O-glukoside, Pet-ara – Petunidin 3-O-arabinoside, Peo-gal – Peonidin 3-O-galactoside, Peo-glu – Peonidin 3-O-glukoside, Peo-ara – Peonidin 3-O-arabinoside, Mal-gal – Malvidin 3-O-galactoside, Mal-glu – Malvidin 3-O-glukoside, Mal-ara – Malvidin 3-O-arabinoside, Que-gal – Quercetin 3-O-galactoside, Que-glu – Quercetin 3- O-glukoside, Que-ram – Quercetin 3-O-ramnoside, Keam-rut – Kaempferol 3-O-rutinoside.

\*,\*\* Explanation see Table 2 – Objasnienia zob. tab. 2.

Anthocyanins constituted the largest group of compounds out of polyphenols determined in the fruits (Fig. 1). Their total content was higher in the conventional fruits (277 mg), as compared with those cultivated ecologically (240 mg). In the research by Wang et al. (2009), the anthocyanin content in the fruits was lower (202 mg), but their percentage content in the total polyphenols was 65%, and it was at a similar level as in small fruits collected at the ecological plantation (67%). On the remaining premises, anthocyanins constituted even up to 79% of the polyphenols determined in large ecological fruits.

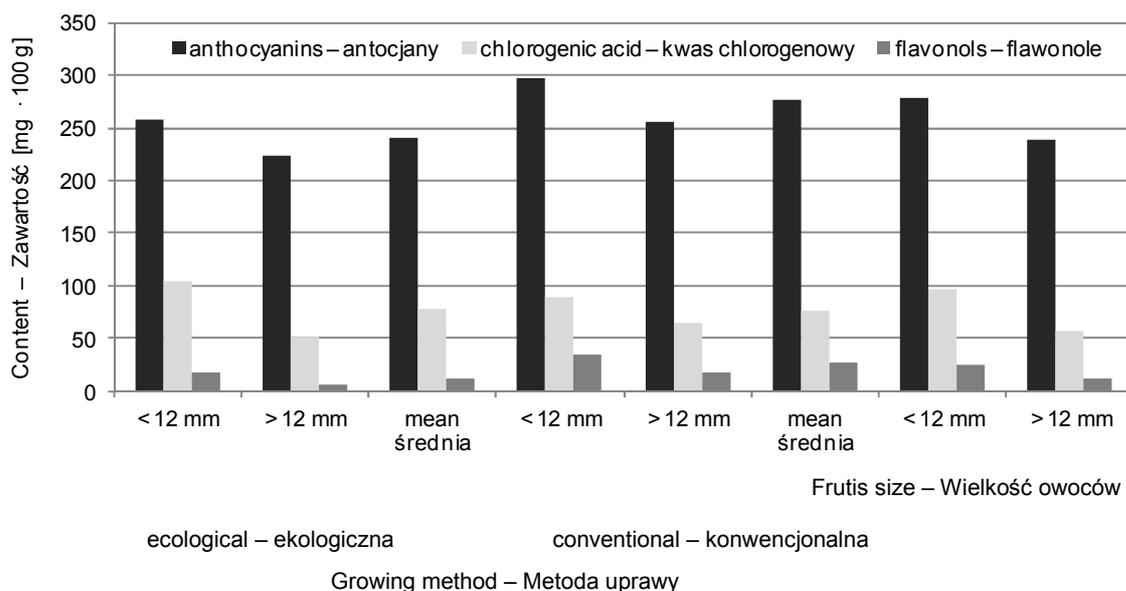


Fig. 1. The content of anthocyanins, flavonols and chlorogenic acid in 'Duke' blueberries depending on growing method and fruit size

Ryc. 1. Zawartość antocyanin, flawonoli i kwasu chlorogenowego w owocach borówki wysokiej odmiany 'Duke' w zależności od metody uprawy oraz wielkość owoców

Considerable differences were observed in the content of the individual polyphenols, depending on the cultivation method and the size of fruits (Table 3). Out of all anthocyanins, delphinidin-3-galactoside compounds constituted the largest group, especially in small fruits from the conventional cultivation (98.9 mg). Small fruits collected from bushes cultivated according to ecological standards were characterised by a content of this compound (30.43 mg), which was three times lower. The content of delphinidin-3-glucoside was higher in these fruits. The average content of delphinidin-3-galactoside in the research by Wang et al. (2008) was 41.74 mg, while the delphinidin-3-glucoside content amounted to 24.64 mg in 100 g of fruits.

Highbush blueberry fruits were also a rich source of chlorogenic acid, especially small ecological fruits (103.9 mg) and conventional ones (88.9 mg). Its content was over twice as low in ecological fruits (51.28 mg). There was no influence in the cultivation method on the chlorogenic acid content in high-bush blueberry fruits. Its content was 76.72 mg · 100 g<sup>-1</sup>

and 77.60 mg · 100 g<sup>-1</sup>. Research by other authors (Ścibisz i Mitek 2007; Wang et al. 2008) indicates the chlorogenic acid content ranging from 40-50 mg in 100 grams of fruits. The content of the aforementioned acid in high-bush blueberry fruits from the Sierra cultivar ranged from 41.71 mg · 100 g<sup>-1</sup> for sawdust cultivation to 70.28 mg · 100 g<sup>-1</sup> for peat cultivation (Ochmian et al. 2009a).

The total flavonol content was higher for the conventional cultivation – 26.57 mg · 100 g<sup>-1</sup> than for the ecological cultivation – 11.69 mg · 100 g<sup>-1</sup>. Small fruits were characterised by a higher content (< 12mm) – 34.84 mg · 100 g<sup>-1</sup> as compared to large fruits (18.28 mg · 100 g<sup>-1</sup>). Highbush blueberries grown organically in New Jersey, Wang et al. (2008) obtained flavonols scope 75.4–197.2 µg · g<sup>-1</sup>, while conventionally cultivated plants had 83.2–119.6 µg · g<sup>-1</sup>.

## CONCLUSIONS

1. Fruits from the 'Duke' cultivar collected at the ecological plantation were characterised by a higher unit weight and a higher share of large fruits (> 12 mm) in the total yield.
2. Small fruits and fruits from the conventional cultivation were firmer and more resistant to mechanical damage.
3. Fruits collected from bushes cultivated in a conventional manner were characterised by a higher content of extract and nitrates. Small fruits were characterised by a higher extract content, regardless of the cultivation method.
4. Small fruits and fruits collected from conventional cultivation bushes were characterised by a higher content of polyphenolic compounds, especially anthocyanins and flavonols. Small fruits also contained more chlorogenic acid.

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**Abstract.** An increase in ecological food is observed in Poland. Produce obtained from ecological farms is perceived as healthier and richer in bioactive substances. The study defines the influence of cultivation methods of high-bush blueberry, 'Duke' cultivar (conventional and ecological) on the yield quality; size of fruits, their firmness and chemical composition. The study was performed in the years 2009–2011 at the Pomiculture Department and a production farm. The fruits collected at an ecological plantation were larger, there were also more large fruits (> 12 mm) in the general yield. These fruits, however, were characterised by lower firmness and lower resistance to mechanical damage. Berries collected from bushes cultivated in a conventional manner were characterised by a higher content of extract and nitrates. Regardless of the cultivation method, small fruits (< 12 mm), were characterised by greater firmness, both along the height axis and the diameter and a higher extract content. Small fruits and fruits collected from conventionally cultivated bushes were also characterised by a higher content of polyphenolic compounds, especially anthocyanins and flavonols. Small fruits also contained more chlorogenic acid.

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