

## PRELIMINARY RESULTS REGARDING THE TOTAL SUGAR CONTENT OF BLUEBERRY GENITORS AND HYBRID PROGENIES

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### Abstract

Blueberry belongs to the *Vaccinium* Genus, *Ericaceae* Family, *Ericales* Order. The highbush blueberry (*Vaccinium corymbosum* L.) is classified as a commercially-grown berry. Blueberries are appreciated for their taste, nutritional value, and both pharmacological properties and biologically active functions. This study objective was to determine the total sugar content in frozen fruits of the highbush blueberry genotypes used as genitors ('Simultan', 'Delicia', 'Duke', 'Azur', 'Northblue', '4/6', '6/38') and 88 hybrid progenies 'Simultan x Duke', 'Simultan x Northblue', 'Azur x 6/38', 'Azur x 4/6', 'Delicia x Duke', 'Delicia x Northblue'. The total sugar content of frozen fruits was analyzed by the phenol-sulfuric acid method. The results showed that the total sugar content of the genitors varied between 2.83 and 3.81 g glucose/100 g. The fruits of 'Simultan' cv. had the highest total sugar content (3.81 g glucose/100 g). The highest total sugar content, 5.486 g glucose/100 g, was found in a 'Simultan × Duke' hybrids.

Keywords: 'Azur', 'Delicia', 'Duke', 'Northblue', 'Simultan', *Vaccinium corymbosum* L.

### 1. INTRODUCTION

Blueberry is one of the major genera in the tribe Vacciniae of the subfamily Vaccinoioadae to the *Vaccinium* Genus, *Ericaceae* Family, *Ericales* Order. The highbush blueberry (*Vaccinium corymbosum* L.) is classified as a commercially-grown berry (Hera et al., 2021). *Vaccinium* sp. is classified into three categories – *Vaccinium angustifolium* (the lowbush), *Vaccinium corymbosum* (the highbush), and *Vaccinium ashei* (rabbit-eye blueberry). *Vaccinium angustifolium* sp., the wild blueberry, is found from the Atlantic provinces westward to Quebec and southward to Michigan and West Virginia (Griffin and Blazich, 2008) and grows to about 15-30 cm in height. The fruits are harvested between July and September and are mainly used raw or processed as frozen or canned fruit. *Vaccinium corymbosum* (highbush blueberries) is mainly grown in Florida and southern Michigan in the US as well as in eastern Canada (Ballington et al., 2001). The highbush blueberry grows to 2-3 m in height. The berries reach a staggered harvest maturity between July and September. *Vaccinium ashei* (hexaploid rabbit-eye blueberry) is native to the Southeastern United States and has

lower chilling requirements compared to *Vaccinium corymbosum*. Rabbit-eye blueberries are also most adaptable, productive, and resilient against biotic stresses (insects and pathogens), especially in a warm climate (Sarkar et al., 2017). The core germplasm collection, maintained as pre-breeding material, consists primarily of *Vaccinium corymbosum*, *V. corymbosum* hybrids, and a small collection of *V. myrtilloides* and *V. angustifolium*. Several varieties released from the program are widely cultivated around the world.

Over time, the interest of Romanian growers led to an increase in highbush blueberry harvested area, production, or consumption. The blueberry harvested area comprises 109270 hectares and the main producer is the United States of America, with 255050 thousand tons (FAOSTAT, 2018, cited by Hera et al., 2021), followed by Europe, with 20718 hectares. Among the European countries with the highest blueberry production can be mentioned Poland - 25301 t, Germany - 12764 t, Netherlands - 10257 t, France - 9127 t, Russian Federation - 3300 t, Italy - 1675 t, Romania – 684 t, and Switzerland – 353 t. It was proved that a higher intake of vegetables and fruits consumed can reduce the risk of developing chronic diseases (Boeing et al., 2012).

Fruits, and especially berries, contain many biochemical compounds (phenolic acids, flavonoids, tannins, vitamin C, vitamin E) with remarkable antioxidant power, resulting in documented antitumor and anti-inflammatory activities (Gündeşli et al., 2019). Moreover, there are several studies regarding the hypoglycemic and hypolipidemic effects of blueberry both, in vitro and in vivo (Huang et al., 2018; Herrera-Balandrano et al., 2021; Li et al., 2022), highlighting their potential to prevent (pre) diabetes (Nunes et al., 2021; Delpino et al., 2022). Most berries such as blueberries, blackberries, and cranberries can be eaten fresh or processed (drinks, jam, jelly) (Scibisz and Mitek, 2009; Howard et al., 2010; Barba et al., 2013). The consumer interest when choosing especially fresh fruits is their taste, which is mainly related to their sugar content.

The objective of this study was to analyze the total sugar content of 95 genitors belonging to *Vaccinium sp.* Germplasm and their progenies hybrids, grown in the Maracineni-Arges area (Southern Romania).

## 2. MATERIALS AND METHODS

### Chemical substances

Concentrated sulfuric acid and phenol were purchased from Redox Bucharest - Sigma Aldrich, Dako, Epp. Romania.

### Plant material

The experiment was carried out at the Research Institute for Fruit Growing, Pitesti (RIFG) in South of Romania at 44°54'12" Northern latitude, and 24°52'18" Eastern longitude, 284 m altitude, in an open field trial in randomized block design with three repetitions plots (5 plants/ genotype/ repetition). The soil type prevailing in the field trial is belonging to the protisoil class, humid aluviosoil type, formed on fluvial deposits with a clayey and sandy granulometric composition. Soil is characterized by acidic reaction (pH 5.85), humus % 2.52, P<sub>2</sub>O<sub>5</sub> 15.2 ppm, also N total % 0.1. There were studied 7 blueberry genotypes with different origins: from Romania ('Simultan', 'Delicia', 'Azur', '4/6', '6/38'), and from America ('Duke', 'Northblue') and 88 hybrid progenies ('Simultan x Duke', 'Simultan x Northblue', 'Azur x 6/38', 'Azur x 4/6', 'Delicia x Duke', 'Delicia x Northblue'), planted at a distance of 1 x 0.50 m. Blueberries were harvested at the full maturity stage, at the beginning of July. All blueberries were stored at -4 °C until to perform the analysis. On the day of the analysis, the fruits were transformed into a homogeneous mass with a vertical mixer.

### **The total sugar content of blueberries**

The total sugar content of blueberries was determined using the UV-Visible spectrophotometric method, an easy and reliable method of sugars measured in vegetable materials. The methodology followed by Dubois et al, 1956, with some modifications was used. This method is based on the fact that simple or complex sugar and its derivatives, including methyl esters with free reducing groups or potentially free, when treated with phenol and concentrated sulfuric acid will generate a yellow-orange color; the reaction is sensible and the yellow-orange color stable. The method is simple, quick, and sensible, and the results are reproducible.

For analysis, 1 g of homogenized vegetal material has been treated with 100 ml distilled water, the samples were kept at 100 °C for 30 minutes and then filtered. 1mL of each filtered aqueous extract was mixed with 5 mL of concentrated sulfuric acid and 1 ml of 5% aqueous solution of phenol. Each mixture was vortexed for 30 seconds for color development. Then, the total sugar content was calculated using the calibration curve of glucose as standard, performed under the same experimental conditions as the samples, using the absorbance values at the maximum absorption, from 490 nm.

#### **Statistical analysis**

Statistical analysis was performed using IBM SPSS 14 program (SPSS Inc., Chicago, IL, USA). All results were statistically evaluated by analysis of variance (ANOVA). Differences between cultivars were highlighted through Duncan's multiple test range ( $p < 0.005$ ). Graphical representations were performed with Microsoft Office Excel 2007.

### **3. RESULTS AND DISCUSSIONS**

The sugars are, along with organic acid, the main taste attribute that orients consumers' preferences towards a certain berry variety, ensuring the financial success of the crop. It is known that sugars are primary products of photosynthesis, and it was previously thought that total content sugar was a fundamental compound correlated with fruit quality and flavor, and also contributes to the nutritional and caloric value of the berries. The quantification of total sugar content (fructose, glucose, and sucrose) was analyzed in the different cultivars and hybrid progenies at harvest maturity.

The study of sugar levels in 11 blueberry cultivars from two regions of China (Weihai and Yingkou) indicated that glucose and fructose were the major components of blueberry soluble sugars, and the correlation analysis showed that glucose, fructose, and sucrose content were positively correlated with total sugar content (Zhang et al., 2020). The authors showed the glucose levels ranged from  $33.79 \pm 0.11$  to  $61.32 \pm 0.79$  mg/g, and the fructose levels ranged from  $22.39 \pm 0.95$  to  $48.02 \pm 0.64$  mg/g, the highest contents of glucose and fructose found in 'Patriot' cultivar of Yingkou. Sucrose levels ranged from  $0.26 \pm 0.00$  to  $4.52 \pm 0.28$  mg/g, with the highest sucrose content found in the 'Jersey' cultivar of Yingkou.

Furthermore, it was reported that the sugar compounds found in berries were 25-33% fructose, 25-27% glucose, and 1-2% sucrose (Okan et al., 2018).

In another study conducted by Ayaz et al. (2001), the fructose and glucose ratios determined in two blueberries (*Vaccinium arctostaphylos* and *V. myrtillus*) native to Turkey were very close.

Regarding the total content of sugar, expressed in g glucose/100 g, it can be observed significant differences in the five genotypes and two hybrids elites (figure 1). The results showed that the total sugar content of the genitors varied between 2.87 and 3.81 g glucose/100 g. The highest content of sugar was obtained on 'Simultan' (3.81 g glucose/100 g) and 'Azur' cvs. (3.39 g glucose/100 g).

Among 'Azur x 4/6' (figure 2) hybrid progenies, the highest total sugar content reached 3.33 g glucose/100 g (2016-3-5) and the lowest was 2.14 g glucose/100 g (2016-3-3, 2016-3-2).

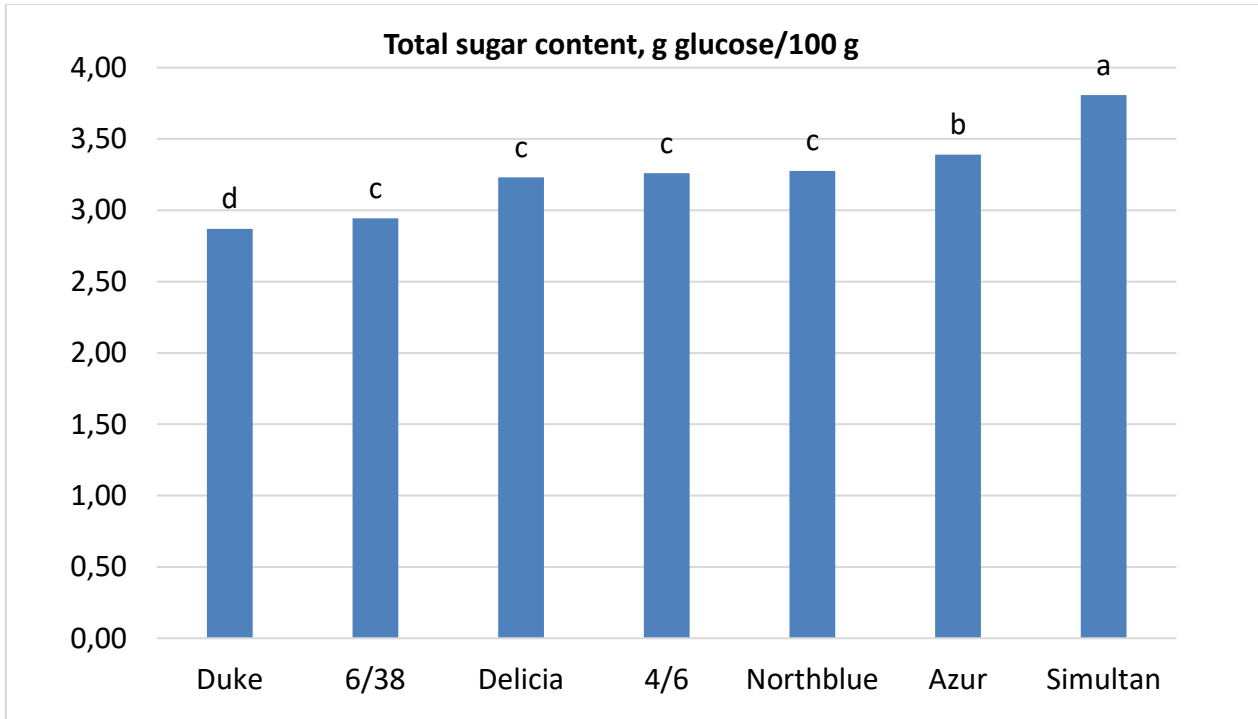


Figure 1. The total sugar content of blueberry varieties

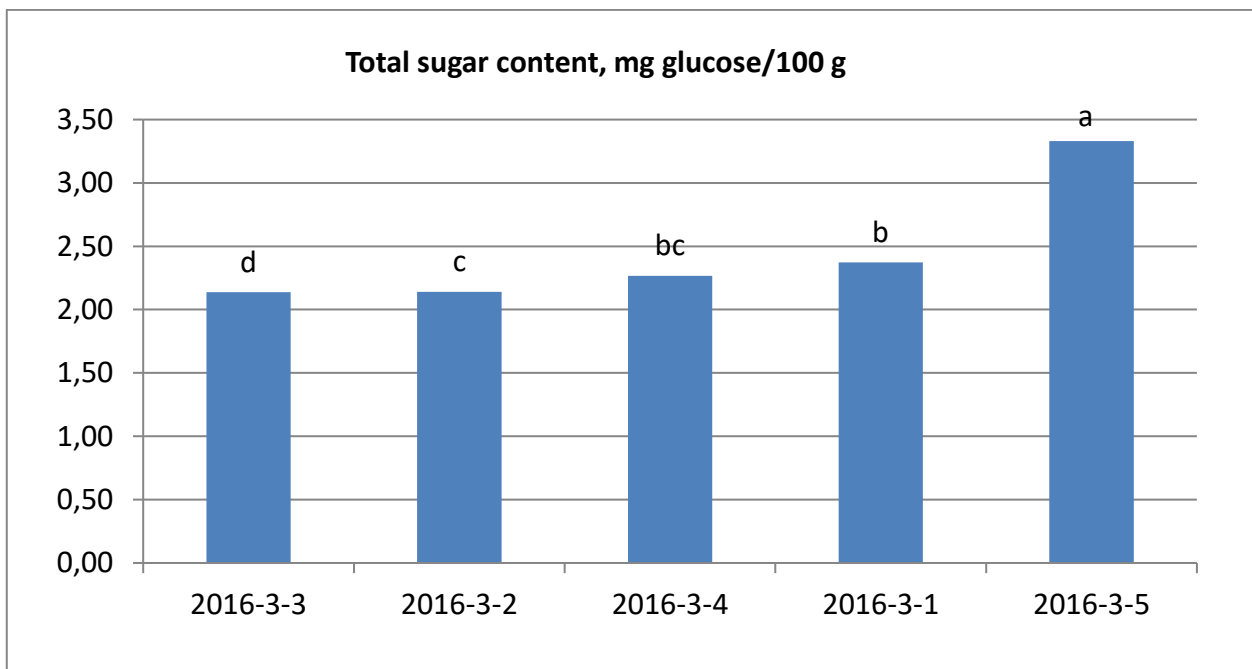


Figure 2. The total sugar content of 'Azur x 4/6' hybrid

For 'Simultan x Northblue' (figure 3) hybrid progenies, total sugar content varied from 3.75 g/100 g (2016-2-8) to 1.26 g glucose/100 g (2016-2-4).

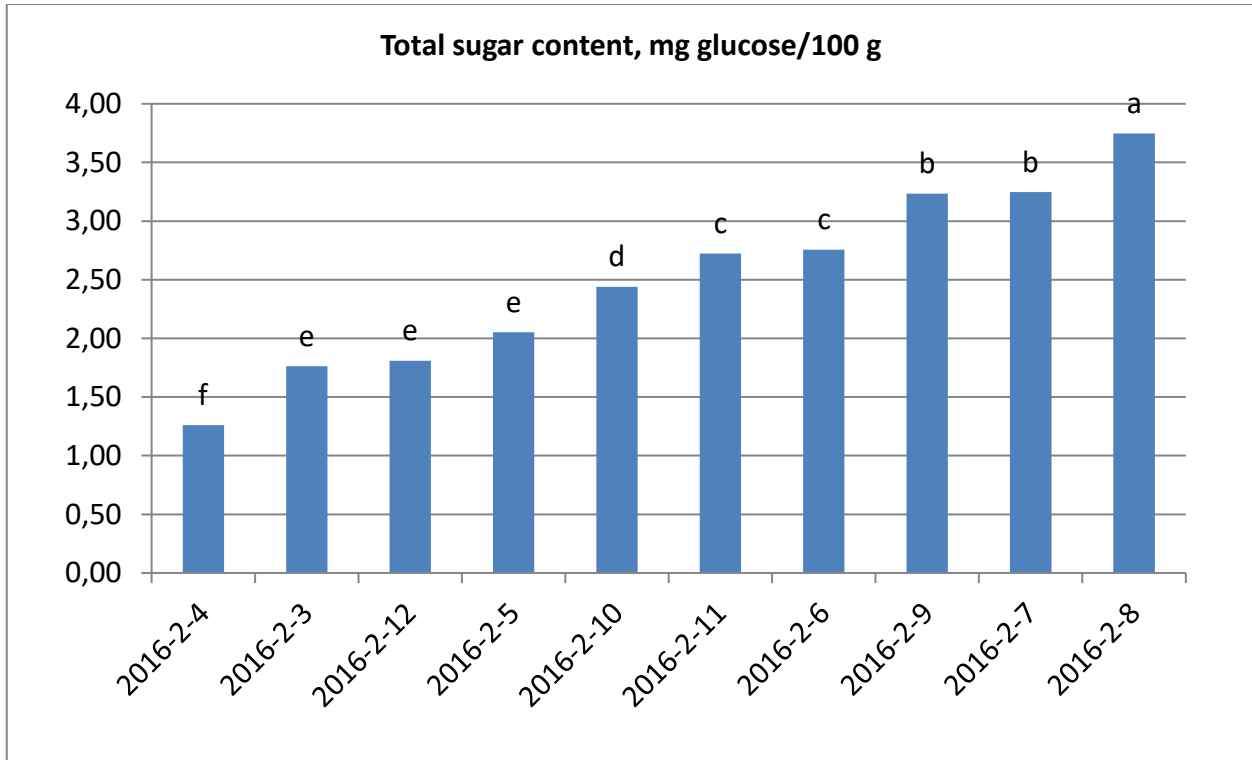


Figure 3. The total sugar content of 'Simultan x Northblue' hybrids

'Azur x 6/38' (figure 4) hybrid progenies has recorded the total sugar content from 3.95 g glucose/100 g (2016-18-11) to 1.80 g glucose/100 g (2016-18-9).

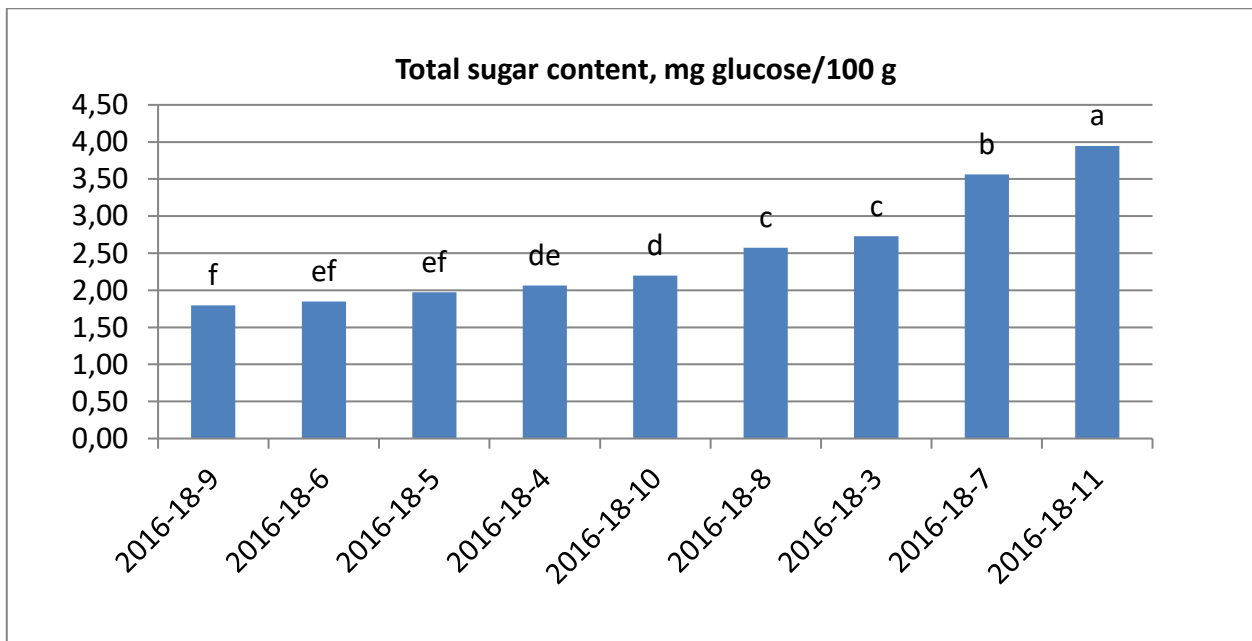
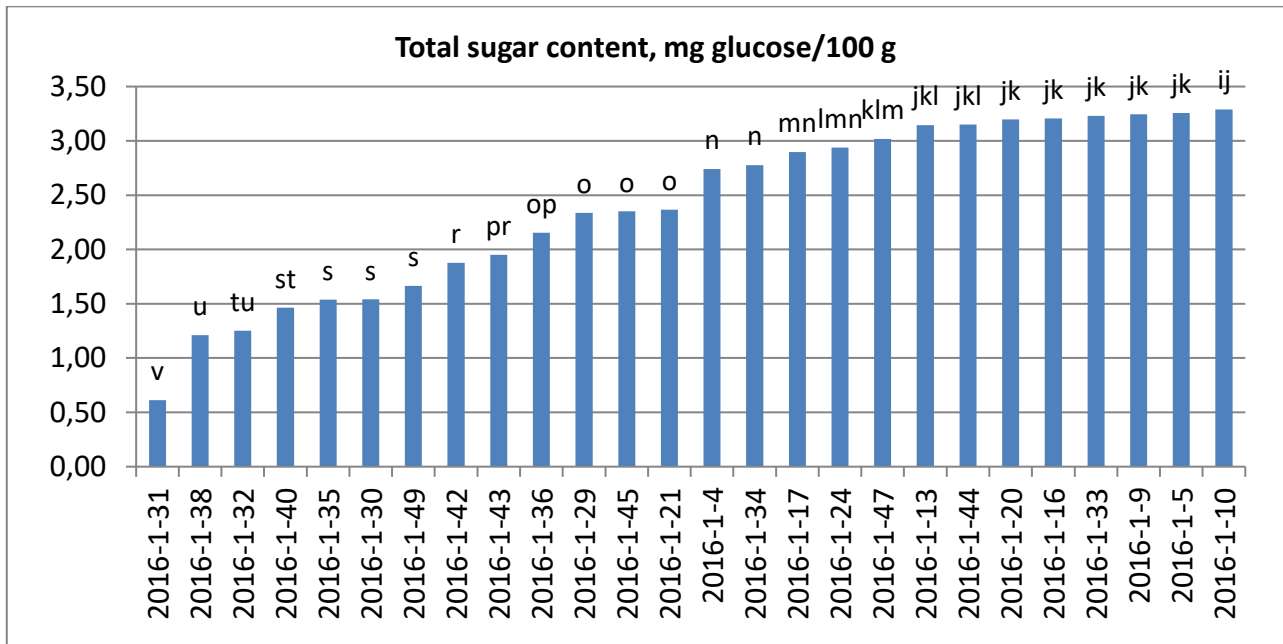
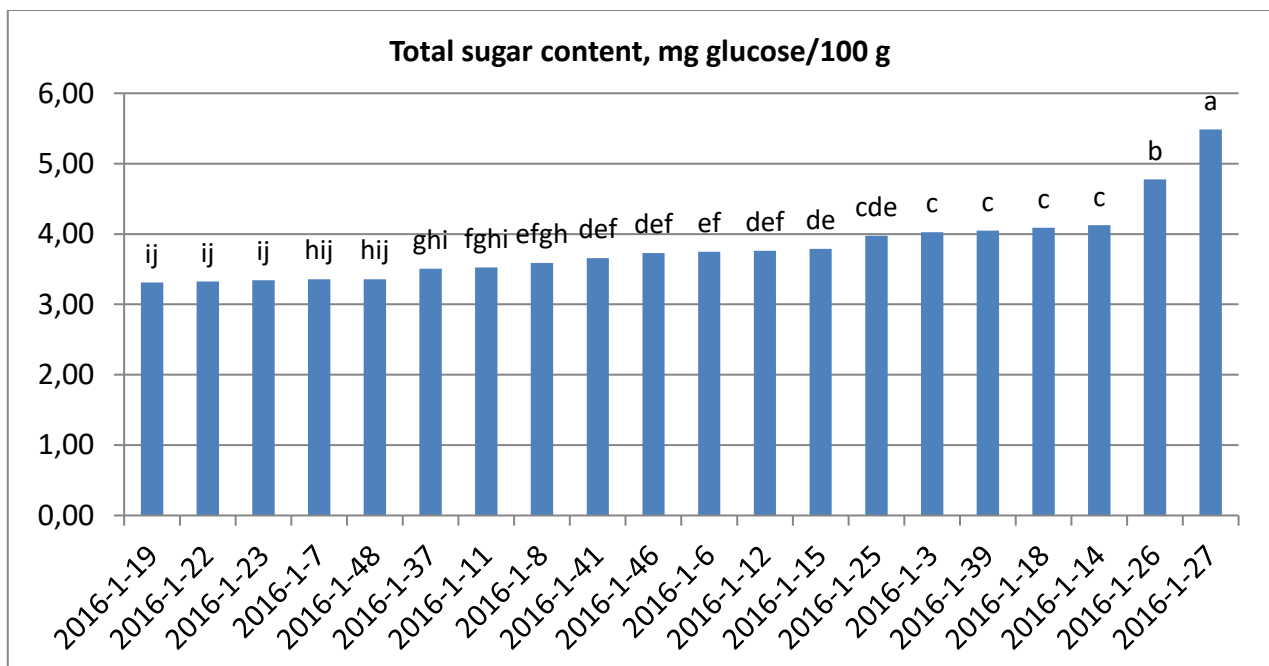


Figure 4. The total sugar content of 'Azur x 6/38' hybrids

Between 'Simultan x Duke' (figure 5) hybrid progenies, only one hybrid showed the highest sugar content (2016-1-27), 5.49 g glucose/100 g, while all others had a lower total sugar content (4.78 g glucose/100 g and 4,12 g glucose/100 g).



a)



b)

Figure 5. The total sugar content of 'Simultan x Duke' hybrids

'Delicia x Northblue' (figure 6) hybrid progenies has obtained 3.62 g glucose/100 g (2016-14-3) and 1.37 g glucose/100 g (2016-14-4). And 'Delicia x Duke' (figure 7) hybrid progenies has recorded the total sugar content from 3.74 g glucose/100 g (2017-17-14) to 1.34 g glucose/100 g (2017-17-11).

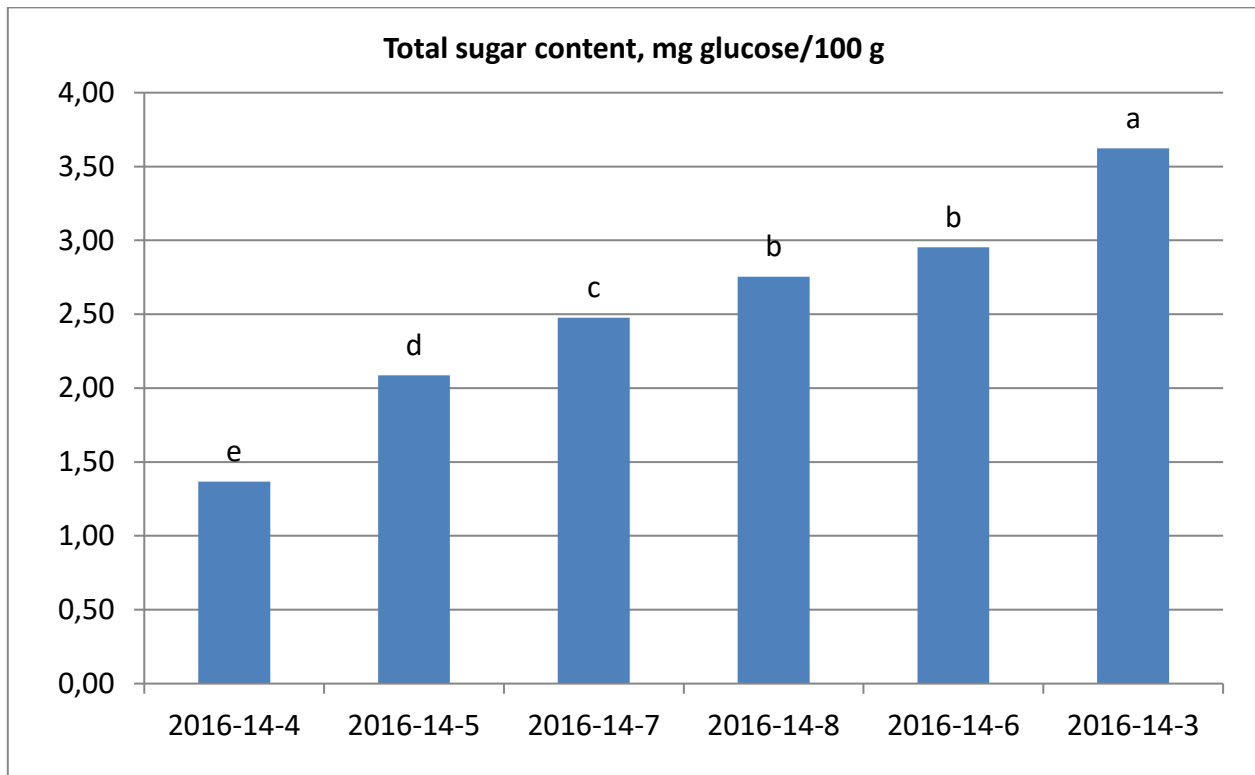


Figure 6. The total sugar content of 'Delicia x Northblue' hybrids

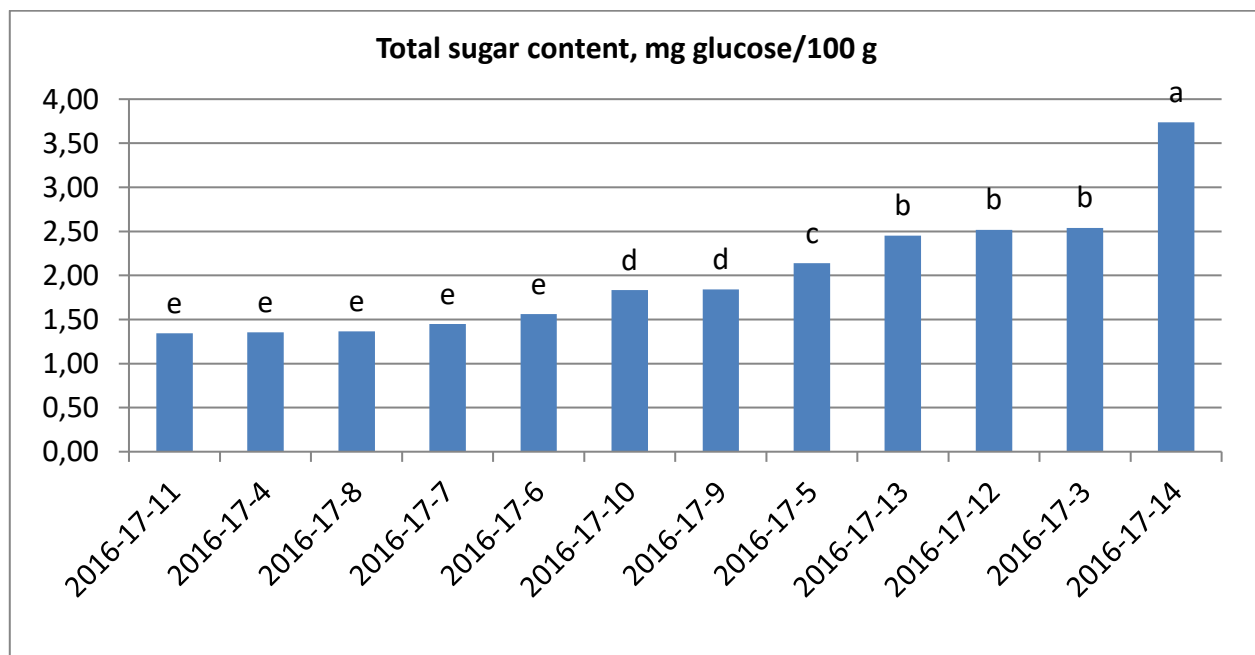


Figure 7. The total sugar content of 'Delicia x Duke' hybrids

The highest total sugar content, 5.49 g glucose/100 g, was found in a 'Simultan × Duke' hybrid, followed by 'Azur x 6/38' hybrid (3.95 g glucose/100 g).

Similar to our study, according to Kalt and McDonald (1996), the glucose + fructose content in three lowbush blueberry cultivars at three stages of fruit maturity, before and after refrigerated storage, ranges from 3.69 g to 11.04 g/100 g fresh weight depending on the cultivar.

Akšić et al. (2019) study high contents of sugars were reported for fruits of three blueberry cultivars grown in an organic and integrated production system, in the village of Pambukovica, municipality Ub, western Serbia. The authors showed that the glucose levels ranged from 26.04±1.06 to 35.51±1.12 mg/g, and the fructose levels ranged from 22.68±1.03 to 30.92±1.29 mg/g in an integrated production system, the highest contents of glucose and fructose found in 'Duke' cultivar. Higher values were obtained in an organic production system. Thus, the glucose levels ranged from 26.42±1.12 mg/g to 46.49±1.53 mg/g, and the fructose levels ranged from 30.52±1.12 mg/g to 43.05±1.01 mg/g, the highest contents of glucose and fructose found in 'Bluecrop' cultivar.

#### 4. CONCLUSIONS

Fruit sugar content represents an important quality trait related to their taste and nutritional value, but also influences aspects such as fermentation capacity (important for vinification, for example) or input costs (added sugar) in the canning industry. Nevertheless, low sugar content berries are the main source of natural antioxidants for diabetics and those who follow calorie-restricted diets. It can vary between cultivars, growing areas, fertilization, irrigation, and soil management practices and also oscillates during the fruit maturation stages. Blueberries are an increasingly popular food, equally consumed fresh, frozen, or processed in various ways (jam, juice, etc.).

In our study, the total sugar content, expressed in mg glucose/100 g, varied significantly among the five genotypes and two hybrids elites. The results showed that the total sugar content of the genitors varied between 2.83 and 3.81 g glucose/100 g. The fruits of 'Simultan' cv. had the highest total sugar content, 3.81 g glucose/100 g. The highest total sugar content, 5.49 g glucose/100 g, was found in a 'Simultan × Duke' hybrids.

The results of our study indicated that blueberries have a total sugar content that covers a wide range of values. This aspect is particularly important in that it meets the requirements of an extremely diverse consumer market and increases the financial success of the blueberry culture.

#### 5. REFERENCES

- Akšić, M.F., Tosti, T., Sredojević, M., Milivojević, J., Meland, M., Natić, M. (2019). Comparison of sugar profile between leaves and fruits of blueberry and strawberry cultivars grown in organic and integrated production system. *Plants* 8(7), 205.
- Ayaz, F.A., Kadioğlu, A., Bertoft, C., Acar, C., Turna, I. (2001). Effect of fruit maturation on sugar and organic acid composition in two blueberries (*V. arctostaphylos* and *V. myrtillus*) native to Turkey. *New Zealand Journal of Crop and Horticultural Science*, 29, 137-141.
- Ballington, J.R. (2001). Collection, Utilization, and Preservation of Genetic Resources in *Vaccinium*. *HortScience*, 36(2), 213-220.
- Barba, F.J., Esteve, M.J., Frigola, A. (2013). Physicochemical and nutritional characteristics of blueberry juice after high pressure processing. *Food Research International*, 50(2), 545-549.
- Boeing, H., Bechthold, A., Bub, A., Ellinger, S., Haller, D., Kroke, A., Leschik-Bonnet, E., Müller, M.J., Oberritter, H., Schulze, M., Stehle, P., Watzl, B. (2012). Critical review: vegetables and fruit in the prevention of chronic diseases. *European Journal of Nutrition*, 51(6), 637-663.
- Delpino, F.M., Figueiredo, L.M., da Silva, T.G., Flores, T.R. (2022). Effects of blueberry and cranberry on type 2 diabetes parameters in individuals with or without diabetes: A systematic review and meta-analysis of randomized clinical trials. *Nutrition, Metabolism and Cardiovascular Diseases*, 32(5), 1093-1109.



- Dubois, M., Giles, K.A., Hamilton, J.K., Rebers, P.A., Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, 28(3), 350-356.
- Herrera-Balandrano, D.D., Chai, Z., Hutabarat, R.P., Beta, T., Feng, J., Ma, K., Li, D., Huang, W. (2021). Hypoglycemic and hypolipidemic effects of blueberry anthocyanins by AMPK activation: In vitro and in vivo studies. *Redox biology*, 46, 102100.
- Howard, L.R., Castrodale, C., Brownmiller, C., Mauromoustakos, A. (2010). Jam processing and storage effects on blueberry polyphenolics and antioxidant capacity. *Journal of agricultural and food chemistry*, 58(7), 4022-4029.
- Gündeşli, M. A., Korkmaz, N., Okatan, V. (2019). Polyphenol content and antioxidant capacity of berries: A review. *International Journal of Agriculture Forestry and Life Sciences.*, 3(2), 350-361.
- Griffin, J.J., Blazich, F.A. (2008). *Vaccinium L. The Woody Plant Seed Manual*. Agric. Handbook No. 727, Washington, DC. US Department of Agriculture, Forest Service, 1154-1159.
- Hera, O., Teodorescu, R., Sturzeanu, M. (2021). Blueberry (*Vaccinium corymbosum*) breeding programme in the main cultivating countries. *Scientific Papers. Series B, Horticulture*. LXV (1), 82-89.
- Huang, W., Yao, L., He, X., Wang, L., Li, M., Yang, Y., Wan, C. (2018). Hypoglycemic activity and constituents analysis of blueberry (*Vaccinium corymbosum*) fruit extracts. *Diabetes, Metabolic Syndrome and Obesity: Targets and Therapy*, 11, 357.
- Kalt, W., McDonald, J.E. (1996). Chemical Composition of Lowbush Blueberry Cultivars. *Journal of the American Society for Horticultural Science*, 121(1), 142-146.
- Li, Z., Tian, J., Cheng, Z., Teng, W., Zhang, W., Bao, Y., Wang, Y., Song, B., Chen, Y., Li, B. (2022). Hypoglycemic bioactivity of anthocyanins: A review on proposed targets and potential signaling pathways. *Critical Reviews in Food Science and Nutrition*, 1-18.
- Nunes, S., Vieira, P., Gomes, P., Viana, S. D., Reis, F. (2021). Blueberry as an attractive functional fruit to prevent (pre) diabetes progression. *Antioxidants*, 10(8), 1162.
- Okan, O. T., Deniz, İ., Yayli, R., Şat, I.G., Öz, M., Serdar, G.H. (2018). Antioxidant activity, sugar content and phenolic profiling of blueberries cultivars: a comprehensive comparison. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca* 46(2), 639-652.
- Sarkar, D., Agustinah, W., Woods, F., Coneva, E., Vinson, E., Shetty, K. (2017). In vitro screening and evaluation of phenolic antioxidant - linked anti-hyperglycemic functions of rabbit-eye blueberry (*Vaccinium ashei*) cultivars. *Journal of Berry Research*, 7(3), 163-177.
- Scibisz, I., Mitek, M. (2009). Effect of processing and storage conditions on phenolic compounds and antioxidant capacity of highbush blueberry jams. *Polish Journal of Food and Nutrition Sciences*, 59(1), 45-52.
- Zhang, J., Nie, J., Li, J., Zhang, H., Li, Y., Farooq, S., Bacha, S.A.S., Wang, J. (2020). Evaluation of sugar and organic acid composition and their levels in highbush blueberries from two regions of China. *Journal of Integrative Agriculture*, 19(9), 2352-2361.