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RUBUS IDAEUS L. FRUIT NUTRIENTS ARE AFFECTED BY DIFFERENT GROWING TECHNOLOGIES

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SUMMARY

The biochemical compounds of red raspberry (*Rubus idaeus* L.) fruits cultivated with conventional growing technology and on a nutrient substrate were studied during 2019–2020 at the Federal Horticultural Research Center for Breeding, Agrotechnology and Nursery, Moscow, Russia. The antioxidant activity, phenolic compounds, and ash constituents of the fruits and the metabolites of the alcoholic extract of the raspberries were determined. The effect of growing technologies, i.e., conventional vs. nutrient substrate, on the accumulation of macro- and microelements in raspberry fruits was established. In red raspberries grown on nutrient substrate, the antioxidant activity decreased by 25 times (aqueous extract) and 1.5 times (alcoholic extract). The K and Na contents and Se contents of red raspberries grown on nutrient substrate were 1.5 and 3 times higher than those of raspberries of grown with conventional technology. Raspberries grown with conventional technology contained 2 times more Ca, Ni, and Mn and 7.4 times more Fe than raspberries grown on nutrient substrate. The total amount of elements in raspberries grown through soilless cultivation was 5.5% higher than that in berries grown conventionally. A total of 48 compounds were identified in the alcoholic extracts, and only 29 substances were found in berries grown on a nutrient substrate. Sugar and citric acid constituted the largest share of red raspberry components. Fructose and turanose disaccharide synthesis in raspberries grown on nutrient substrate was 20% higher than that in conventionally grown raspberries. A total of 48 organic compounds with different biological activities were identified. They included five substances with antimicrobial activity, three phenolic substances, eight organic acids, four sugar acids, nine amino acids, and 19 sugars and their derivatives. At the same time, 42 compounds were found in raspberries grown with traditional technology, and 21 compounds were identified in raspberry fruits grown on nutrient substrate. Three fatty acids, namely, α-linoleic acid (polyunsaturated omega-6 fatty acid), palmitic acid, and stearic acid (saturated fatty acid), along with cinnamic acid, shikimic acid, and chrysin were found in berries grown conventionally.

Keywords: *Rubus idaeus* L., conventional growing, nutrient substrate, nutrients, antioxidant activity, ash constituents of fruits, metabolites, bioactive compounds

Key findings: The qualitative and quantitative compositions of the nutrients differed in raspberry fruits grown with conventional and nutrient substrate technologies.

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INTRODUCTION

Modern raspberry breeding must shift its focus from traits related to agronomic indicators to those related to the sensory qualities (Jennings *et al*., 2016) and potential health benefits (M*et al*., 2016) of fruits and the growth of new cultivars on a nutrient substrate. Analytical chemistry has revealed significant advances in environmental, biochemical, and genetic factors underlying the accumulation of certain compounds in fruits (Moyer *et al*., 2002; Prichko *et al*., 2021). In addition, the information regarding the mechanisms of action of specific phytochemicals in relation to human health is becoming the scientific basis for the creation of new raspberry cultivars (Beekwider *et al*., 2005; Krause-Baranowska *et al*., 2014). Raspberries have long been positioned as a rich source of biologically active compounds that have a positive effect on human health. Raspberry fruits are used for the treatment and prevention of many diseases (Stoner, 2009; Eremeeva *et al*., 2019). The therapeutic and preventative properties of raspberries are related to their biochemical composition. In raspberries, the most important primary metabolites are soluble dry substances, organic acids, sugars, fiber, fats, and proteins, which affect nutritional value (Cekig, 2010; Mazur *et al*., 2014). Biologically active substances in red raspberries are represented by vitamins, polyphenolic compounds, and macro- and microelements (Xiao *et al*., 2017; Zhbanova, 2018; Stojanov *et al*., 2019). These phytonutrients have anti-inflammatory and antioxidant effects. The nutrients and bioactive components contained by raspberries play important protective roles in human health (Freeman et al., 2016).

The quantitative and qualitative compositions of raspberry metabolites depend on genotype, fruit maturity, natural and climatic conditions, and growing technology (Kula *et al*., 2016; Palonen *et al*., 2017; Akimov *et al*., 2020; Will *et al*., 2020). In 2018, global raspberry production reached 870 209 tons in an area of 124 971 ha; Russia was the leading raspberry producer, supplying 19% of the global total (FAOSTAT, 2020). The demand for raspberries has risen sharply in Europe and North America mainly due to the freshness, organoleptic features, nutritional value, and health claims of these fruits (Overview Global Berry Market, 2020).

The conventional soil-based cultivation of perennial raspberries (*Rubus idaeus* L.) is declining in Germany, Switzerland, and in other parts of northwestern Europe because of root diseases and issues related to fruit quality. In these countries, the raspberry production system has changed, and cultivation with nutrient substrates, pots, and polytunnels are preferred over traditional cultivation. Such structures enable regulating microclimatic conditions and limiting the spread of diseases and pests (Marchi *et al*., 2019; Linnemannstöns, 2020; Tan *et al*., 2020).

In Russia, the current raspberry growing technology is not widely used but has great future prospects. At the same time, raspberries are already being successfully grown in high tunnels in the Leningrad and Tula regions, and their construction in Bryansk, Voronezh, Kaluga, Moscow, and the south of Russia is being planned. In this regard, the question regarding the nutritional value of fruits cultivated on a nutrient substrate base arises. Qiu *et al*. (2016) reported that the soluble solid contents of raspberries grown conventionally or on a nutrient substrate do not differ. However, Svensson (2016) reported that conventionally grown raspberries have the best fruit quality.

In consideration of such contradictory information, the nutrient contents of raspberries grown through conventional technology and on a nutrient substrate were comparatively analyzed. This work might be the first study to provide a basis for planning further breeding efforts to create raspberry cultivars that are suitable for growing on substrates. The main hypothesis of this research is that the metabolic profiles of red raspberry fruits may differ in accordance with growing technologies. The objective of this research was to measure the antioxidant activity, total phenolic compounds, ash constituents, bioactive compounds, and metabolomic profiles of red raspberry fruits grown with conventional and nutrient substrate technologies.

MATERIALS AND METHODS

Experimental conditions

Raspberries were grown with conventional technology during 2019–2020 at the Federal Horticultural Research Center for Breeding, Agrotechnology, and Nursery (FHRCBAN) on the site of the Kokino genetic collection (53°15'N, 34°12′E). The soil was gray forest, medium loamy, and well cultivated. The arable layer had a depth of 26 cm and a humus content of 3.2%, P_2O_5 content of 35 mg 100 g^{-1} of soil, and K_2O content of 13.5 mg 100 g^{-1} of soil. The soil solution was weakly acidic (pH 6.1). Bare-rooted plants were planted with a 3.0 m × 0.5 m scheme. Then, the line of shoots was established with a width of 40 cm. The soil of the between-row spacing was maintained as black fallow. Fruiting was carried out on annual shoots. After harvest and the end of leaf fall in November, the shoots were mowed to the soil level and discarded.

The experimental subjects were the fruits of the primocane raspberry selection 44-154-1 (‘Penguin’ × ‘Bryanskoe Divo’) (Figure 1). This selection is vigorous and produces 4–5 upright, strongly branching shoots and 150–200 raspberries. The fruits are large (average weight of 5.0–5.5 g, maximum weight of 7.8 g), conical, crimson, firm, and well separated from the receptacle with a taste of 4.0–4.2 points on a ranking scale of 1–5. The harvesting period under the open-field condition lasted from the end of July to the end of September.

Figure 1. Fruiting of selection 44-154-1 under conventional growth (left) and on a nutrient substrate (right).

Raspberries were also grown during 2019–20 on nutrient substrate obtained from Tula Berry Company LLC, Tula, Russia (53°71′N, 36°24′E). This technology involves cultivating raspberry plants in 10 L pots filled with a substrate based on coconut fibers. The containers with plants were placed under a film canopy without side walls. The pots were installed with a layout of 3.0 m × 1.0 m on plastic pallets to prevent the root system from coming into contact with soil. Mineral nutrients and water were provided with a drip irrigation tape in accordance with the protocol developed by FHRCBAN. For chemical analysis, berries were collected at the optimal ripeness stage at the end of August. Three replicates, each consisting of 30 fruits for a total of 90 fruits per sample, were selected. Thereafter, the fruits from each replicate were pooled to obtain a mixed sample for biochemical analysis. All the analyses were performed in triplicate.

Chemicals

All the chemical substances chosen for analysis were of analytical grade and were procured from Sigma Aldrich (USA) and Merck KgaA (Germany).

Sample preparation

From the middle probe, 300 g samples were prepared and extracted with double distilled water (for the determination of antioxidant activity and phenol composition) and methanol (pure) by using a high-speed homogenizer (10 000 rpm, 1 min, UltraTurrax T25 Basic, IKA). After centrifugation at 4000 × *g* (Sigma, Germany) for 10 min, the supernatant was used for measurement. Extraction and measurements were carried out with a three-fold repetition.

Data recorded

The parameters determined were the ash constituents, antioxidant activity, total phenolic compounds, and metabolomic profiles of the fruit extracts of raspberries grown conventionally and on a nutrient substrate.

Antioxidant activity determination

Antioxidant activity was measured in accordance with the method of Brand-Williams *et al*. (1995) by using the compound 2,2-diphenyl-1-pikrylhydrazyl (DPPH ˙). A Thermo Helios ɣ spectrophotometer (Thermo Fisher Scientific, made in England) was used. Samples that had been homogenized in distilled water were placed on a Lab-PU-01 shaker (Russia) for 8 h and then filtered. Antioxidant activity was measured 10 min after interaction between the extract and reagent at the wavelength of 515 nm. Antioxidant activity values were calculated by using the following formula:

$$\text{Inhibiting DPPH} = (AC - AAt) = AC/100(\%)$$

where,
AC = DPPH solution absorption
AAt = absorption in the presence of the antioxidant

Analysis of total phenolic compounds

The amount of total phenolics content (TPC) was determined with Folin–Ciocalteu reagent in accordance with the method described by Velioglu *et al*. (1998). A standard curve with gallic acid was used. Different concentrations of gallic acid were prepared in water, and absorbance was recorded at 750 nm. A total of 100 µL of diluted sample (1:10) was dissolved in 500 µL of Folin–Ciocalteu reagent and 1000 µL of distilled water. The solutions were mixed and incubated at room temperature for 1 min. After 1 min, 1500 µL of 20% sodium carbonate (Na_2CO_3) solution was added. The final mixture was shaken and then incubated for 2 h in the dark at room temperature. Absorbance was measured at 750 nm by using a Helios Y UV–vis spectrophotometer, and the results were expressed in mg of gallic acid calculated on the basis of the wet weight of the fruits.

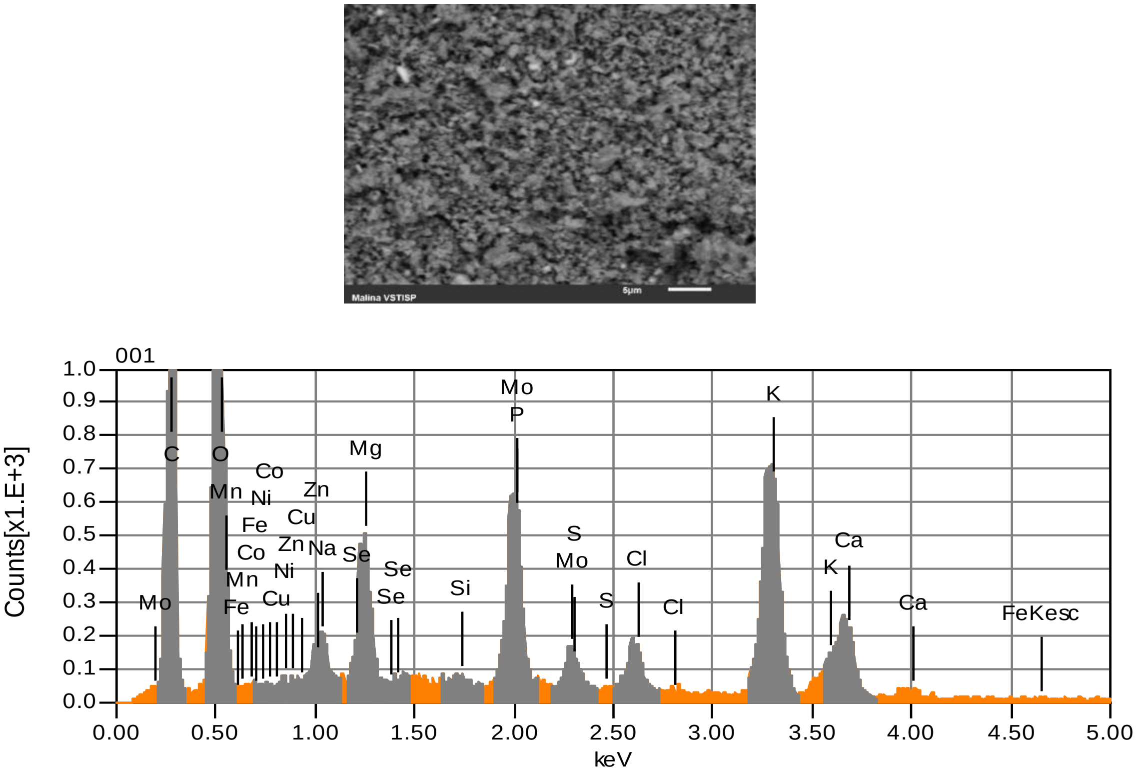

Figure 2. EDS-analysis results report: studied area and spectrogram.

Energy dispersive spectrometry analysis

The chemical composition of the basic ash constituents (Na, Mg, P, S, Si, K, Cl, Mn, Co, Fe, Ca, Cu, Zn, Ni, Se, and Mo) was determined via energy dispersive spectrometry (EDS) on the analytical raster electron microscope JEOL JSM 6090 LA. The microscope had a resolution of 4 nm and was operated at an accelerating voltage of 20 kV (secondary electron image). Zooming was performed from 10× to 10 000×. The working distance during elemental analysis was 10 mm. The energy-dispersive spectrometer enabled quantitative X-ray microanalysis with the desired area of analysis at a point or area and to obtain elemental distribution maps. X-ray microanalysis data were presented in accordance with standard protocols and included the images of the microstructures of the sample under study, the table of the weight data and atomic correlation, spectra, and histograms. An example of the obtained spectra is shown in Figure 2. The concentration of the desired elements can be determined on the basis of spectral line intensity. The fractional accuracy of the chemical analysis was determined as follows: at element concentrations of 1% to 5%, the accuracy was less than 10%; at the element concentrations of 5% to 10%, the accuracy was less than 5%; and at the element concentrations of more than 10%, the accuracy was less than 2%. A total of 50 ash areas of each sample were studied. The local analysis was 3 mm, and the scanned area was not less than 12 μm.

Processing of leaf probes for EDS analysis

Raw plant material, namely, primocane raspberries, with an average mass of 10 g were dried in a drying oven at 50 °C–60 °C. The dried samples were mineralized in a muffle furnace Naberterm (Germany) at

450 °C. The obtained ash was dispersed ultrasonically at 18 kHz for 15 min. An even layer of a dispersant was applied on the object table covered with carbonic scotch.

Metabolic analysis by gas chromatography–mass spectrometry

Metabolite analysis was performed via gas chromatography–mass-spectrometry (GCMS) on a JMS-Q1050GC chromatograph. A DB-5HT capillary column (Agilent, USA, length 30 m, inner diameter of 0.25 mm, film thickness of 0.52 μm, and helium as the gas carrier) was used. Substances were identified on the basis of withholding values and mass spectra from the NIST-5 National Institute of Standards and Technology library, USA. The scanning range was 33–900 m/z. The probability of substance determination was within 75%–98%. The temperature gradient during the analysis was 40 °C–280 ° C. The oven temperature was increased from 40 °C to 130° C at the rate of 1 °C min^{-1}, from 130 °C to 200° C at 2° C min^{-1}, and from 200 °C to 280° C at 4° C min^{-1} and held at 280 °C for 40 min. The temperature of the ion source was 200 °C. The gas flow (helium) in the column was 2.0 mL min^{-1} in split-flow injection mode. A total of 1–2 mL of the evaporated extract injected as the sample.

Processing of leaf probes for GCMS analysis

Raw plant material was homogenized with an IKA homogenizer (Germany). Then, 0.5 g of the sample was extracted with 15 mL of pure ethanol and centrifuged with Sigma 3-18KHS (Germany). A total of 200 μL of the centrifuged material was evaporated to dryness under helium flow. Derivation was performed by using BSTFA (N,O bis-trifluoracetamide trimethylsilyl) for trimethylsilylation in accordance with the method described by Lebedev (2003). BSTFA was used for silylation within 30 min at 100 °C.

Statistical analysis

All analyses were performed in triplicate. The results were expressed as mean values (n = 3) with standard deviation (SD). MS Excel software was used for statistical analysis (Microsoft Excel, v. 2016).

RESULTS AND DISCUSSION

Antioxidant activity and phenolic compounds

The capacity of the raspberry fruit extracts to trap DPPH^+ free radicals was used as a measure of total antioxidant capacity and total phenolic content (Table 1). The antioxidant activities in the alcohol and aqueous extracts of conventionally grown raspberries differed slightly. However, for raspberries grown on a nutrient substrate, the antioxidant activity of the alcohol extract was 18 times higher than that of the aqueous extract. The antioxidant activity of the aqueous extract of the conventionally grown raspberries was 25 times higher than that of the raspberries grown on a nutrient substrate. The antioxidant activity of the alcohol extract of the conventionally grown raspberries was 1.5 times higher than that of the raspberries grown on a nutrient substrate. Hassimotto *et al*. (2008) stated that the values of antioxidant activity (DPPH) can be classified as high (>70% inhibition), intermediate (40%–70% inhibition), and low (<40% inhibition). In accordance with this classification, red raspberries were found to be a good source of antioxidants. The antioxidant activity of the red raspberries in this study corresponded to the range found in other studies (Cek and Ozgen, 2010; Jin *et al*., 2012).

Considerable attention has been paid to the study of the phenolic composition of raspberry fruits (Moreno-Medina *et al*., 2018; Anjosa *et al*., 2020). Phenolics are responsible for the antioxidant potential of raspberry plants

Table 1. Influence of the growing methods on the antioxidant activity (expressed in %) of aqueous (AAA) and methanolic (AAM) extracts and the total content of polyphenols (TPC, expressed in mg equivalent of gallic acid [mg/100 g TW]) in the fruits of red raspberry (44-154-1).

Growing methods	Determined indicators		
	AAA	AAM	TPC
Conventional growing	53.32 ± 1.23	58.71 ± 1.21	1110 ± 2.28
Nutrient substrate	2.12 ± 0.39	38.91 ± 1.11	730 ± 1.28

Mean of three determinations ± standard deviation.

Table 2. Ash constituents of raspberries and mass % in ashes.

Elements	Cultivation methods	
	Conventional cultivation	Nutrient substrate
K	15.404 ± 2.201	23.723 ± 2.012
P	5.826 ± 0.902	5.644 ± 0.812
Mg	3.006 ± 0.412	3.398 ± 0.384
Mo	3.306 ± 1.011	3.918 ± 0.912
Ca	4.862 ± 0.811	2.564 ± 0.452
Co	3.602 ± 0.611	3.291 ± 0.512
Ni	2.042 ± 0.321	1.756 ± 0.402
Zn	1.148 ± 0.312	1.226 ± 0.372
Na	0.644 ± 0.122	1.051 ± 0.108
Cl	1.894 ± 0.611	0.988 ± 0.571
S	0,568 ± 0.084	0,713 ± 0.082
Mn	0,571 ± 1.012	0,325 ± 0.754
Cu	0,336 ± 0.045	0,368 ± 0.067
Se	0,083 ± 0.054	0.257 ± 0.075
Fe	0.377 ± 0.042	0.051 ± 0.054
∑	43.87	49.39

Mean of three determinations ± standard deviation.

(Zhang *et al*., 2015). Conventionally grown berries accumulated 1110 mg 100 g^{-1} phenols, whereas phenol accumulation in raspberries grown on nutrient substrate was 35% lower (730 mg 100 g^{-1}). The obtained results correspond with the data of Bobinaitė *et al*. (2016), who reported that the total content of phenols varied from 200 mg 100 g^{-1} to 500 mg 100 g^{-1} depending on the studied raspberry cultivars. In accordance with the polyphenol classification proposed by Vasco *et al*. (2008), red raspberries can be categorized as a crop with average phenol content.

The correlation coefficients for the antioxidant activities of the aqueous and alcohol extracts of raspberry fruits with phenol content were high (r = 1). Other studies also reported similar data and found a linear correlation between the total antioxidant capacity and the phenol content in raspberry fruits, i.e., r = 0.911 and r = 0.965 (Wang and Lin, 2000; Deighton *et al*., 2000).

Ash constituent

Raspberries are known to be rich in minerals (Pereira *et al*., 2018). Fifteen chemical elements were consistently identified in the studied raspberry samples regardless of growing technology (Table 2). The contents of the elements in conventionally grown red raspberries decreased in the following order: K > P > C a> Mo> Co ≈ Mg > Ni > Cl > Zn > Na > Mn ≈ S > Fe > Cu > Se. The contents of the elements in raspberries grown on nutrient substrate decreased in the order of K > P > Mo > Mg > Co > Ca > Ni > Zn > Na > Cl > S > Cu > Mn > Se > Fe. Among the macronutrients, K had the highest concentration. The same phenomenon has also been observed in other berry crops, i.e., actinidia, blackberry, strawberry, and blueberry (Pereira *et al*., 2018; Kozak *et al*., 2021). However, raspberries grown on nutrient substrate contained 1.5 times more K and Na and 3 times more Se than conventionally grown raspberries. The higher content of minerals in this variant was apparently associated with regular amendment with macro- and microelements. At the same time, conventionally grown fruits contained 2 times more Ca, Ni, and Mn and 7.4 times more Fe than fruits grown on nutrient substrate. Total element content was 5.5% higher in raspberries grown on nutrient substrate than in conventionally grown raspberries.

GCMS analysis

Bobinaitė *et al*. (2016) and Will *et al*. (2020) revealed that the fruits of the same raspberry cultivars grown at different latitudes did not have significant differences in terms of primary and secondary metabolite contents. In this study, we assumed that geographical location did not play a significant role in the accumulation of biochemical substances. In total, 48 main compounds were identified, and the peak heights of these compounds were not lower than 0.05% on the instrument scale.

A total of 42 and 21 compounds were found in conventionally grown raspberries and in raspberries grown on nutrient substrate, respectively. Fruit flavor and taste were highly influenced by sugar content. In red raspberry fruits, sugars constituted the largest share: D-glucose (90%–100%), D-psicopyranose (70%), D-(–)-fructofuranose (44%–65%), ethyl a-D-glucopyranoside (32%–36%), disaccharide D-(+)-turanose (14-34%), and citric acid (70-91%) (Figure 3).

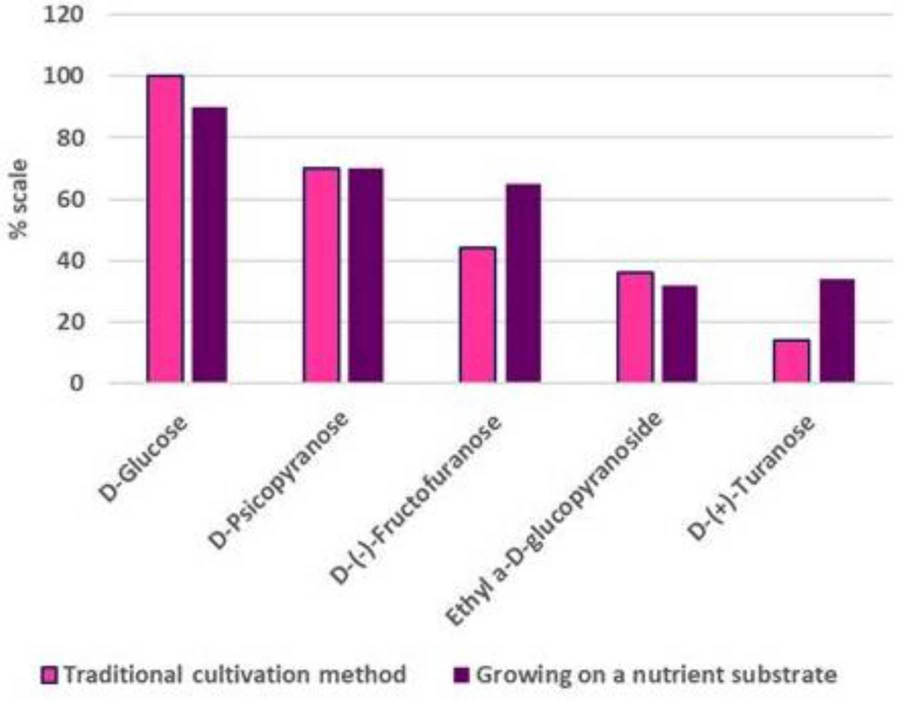

Figure 3. Comparative composition of the basic sugars of the 44-154-1 red raspberry.

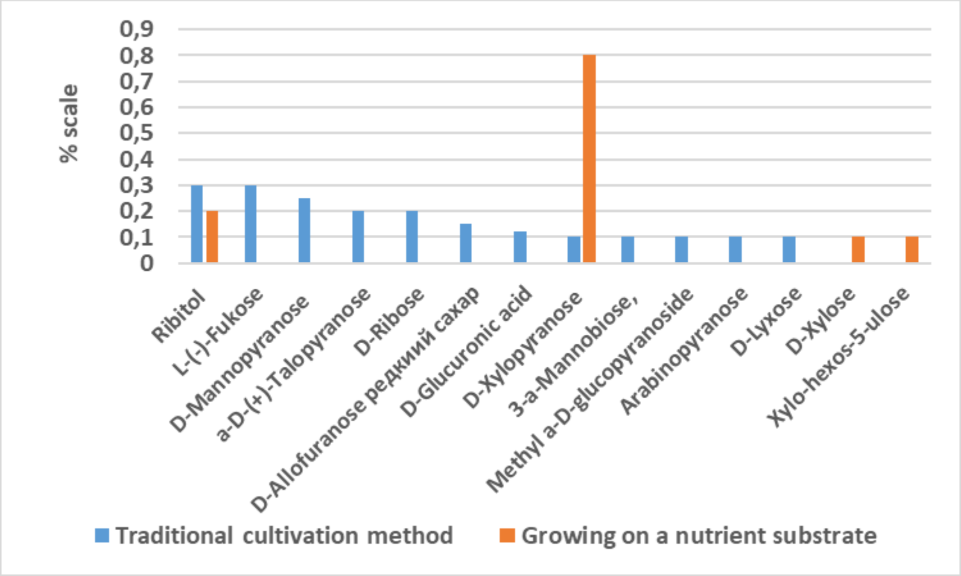

Figure 4. Comparative composition of basic sugars and their derivatives of the 44-154-1 red raspberry.

Glucose content was lower by 10%, whereas fructose and turanose disaccharide contents were higher by 20% in red raspberry fruits grown on a nutrient substrate than in conventionally grown raspberries. In addition to the main sugars, 14 other carbohydrates and their derivatives were identified in red raspberry fruits at the proportion of 0.1%–0.3%. Previous studies have shown that red raspberries contain glucose, fructose, sucrose, sorbitol, mannitol, and myo-inositol (Muir *et al*., 2009; Megías-Pérez *et al*., 2014; Lee, 2015).

Twelve carbohydrates and their derivatives were synthesized in conventionally grown red raspberry fruits, i.e., ribitol L-(−)-fucose, D-mannopyranose, D-(+)-talopyranose, D-ribose, D-allofuranose, D-glucuronic acid, D-xylopyranose, 3-a-mannobiose, methyl-a-D-glucopyranoside, arabinopyranose, and D-lixose. However, when grown on nutrient substrate, only four substances, i.e., ribitol, D-xylopyranose, D-xylose, and xylohexos-5-ulose, were synthesized in raspberry fruits, and the latter two were found only in berries grown on a nutrient substrate. D-xylopyranose content was 8 times higher in red raspberries grown on a nutrient substrate (Figure 4). A large group of substances consisting of organic acids participate in the physiological processes of plants and exhibit antioxidant properties. They are important components in human nutrition (Table 3). A total of 20 organic compounds were found, among which five, namely, 4-amibobutanoic acid, benzoic acid, lactic acid, malic acid, and the sugar alcohol glycerol, had an antibacterial effect. Citric acid dominated. Previous studies have confirmed that red raspberries have high contents of sugars and citric acid (Kafkas *et al*., 2008; Cekig, 2010; De-Souza *et al*., 2014; Schulz and Chim, 2019). Succinic and fumaric acids perform an important biological role in the human body.

Notably, 18 out of 20 organic acids were observed in raspberry fruits grown with conventional technology, and only 10 were found in berries grown on a nutrient substrate. At the same time, the share of glycerol content in berries grown on a nutrient substrate was 45 times lower than that in raspberries grown with conventional technology. In this work, some phenolic compounds, namely, cinnamic (caffeic) and shikimic acids, as

Table 3. Organic compounds detected in the methanolic extract of raspberry fruits and % scale.

No.	Substances	Peak height, % of scale		Biological characteristic
		Conventional cultivation	Nutrient substrate	
1	4-Amibobutanoic acid	0.11	–	Antimicrobial[1]
2	Benzoic acid	0.08	0.08	Antimicrobial[2]
3	Lactic acid	0.05	0.05	Antimicrobial[3]
4	Glycerol	9.0	0.2	Antimicrobial[4]
5	Malonic acid	0.1	0.1	Organic acid
6	Erythro-pentonic acid	0.1	–	Organic acid
7	Clyceric acid	0.1	–	Sugar acid
8	Fumaric acid	0.1	–	Organic acid (biological role)[5]
9	DL-malic acid	0.2	0.25	Antimicrobial[6]
10	Erythronic acid	0.2	–	Sugar acid
11	Succinic acid	0.1	0.1	Organic acid (biological role)[7,8]
12	Oxalic acid	0.1	–	Organic acid
13	b-Hydroxypyruvic acid	–	0.08	Organic acid
14	Citric acid	97	70	Organic acid
15	Propanedioic acid	0.1	0.1	Organic acid
16	Arabinoic acid	–	0.08	Sugar acid
17	Glutaric acid	0.2	-	Sugar acid
18	Cinnamic (Caffeic) acid	0.1	-	phenolic compound, antioxidant
19	Shikimic acid	0.1	-	phenolic compound, antioxidant
20	Chrysin	0.12	–	flavonol, antioxidant (biological role)[9]

Note: – not found; [1] – Meeta et al., 2014 ; [2] – Park et al., 2001; [3] – Wang et al., 2015; [4] – Fluhr et al., 2008; [5] – Golda et al., 2012; [6] – Raybaudi-Massilia et al., 2009; [7] – Mills and O'Neill, 2014; [8] – Terasaki et al., 2018; [9] – Khoo, Chua , Balaram, 2010.

well as chrysin, were found only in raspberries grown with conventional technology. Cinnamic (caffeic) acid possesses antioxidant and anti-inflammatory properties and has shown potential therapeutic benefits in experimental diabetes and hyperlipidemia (Alam *et al*., 2016). Shikimic acid is widespread in various plants and has potential biological properties. Moreover, it has pharmacological relevance because it also acts as an intermediate in the manufacture of many drugs mainly due to its antiviral effects (Singh *et al*,, 2020). Chrysin is a natural flavonoid that is currently under investigation due to its important biological anticancer properties (Khoo *et al*., 2010).

The proportion of amino acids in red raspberries did not exceed 0.1% of the norm (Table 4). Six amino acids were identified in red raspberries grown with conventional technology: aspartic acid, glutamic acid, proline, serine, tyramine, and threonine. However, only two amino acids (glutamic acid and tyramine) were found in red raspberries grown on a nutrient substrate. The amino acids isoleucine, leucine, valine, alanine, citrulline, arginine, glutaric acid, proline, glutamine, asparagine, aspartic acid, histamine, glycine, and tyrosine have been found in the fruits of many plants (Whang *et al*., 2007; Kim *et al*., 2010).

Fatty acids are concentrated mainly in raspberry seeds and are of great interest in the cosmetic and pharmaceutical industries (Moreno-Medina *et al*., 2018). Three fatty acids, i.e., α-linoleic acid (polyunsaturated omega-6 fatty acid), palmitic acid, and stearic acid (saturated fatty acid), were found only in conventionally grown red raspberries (Table 4). Previous studies have reported the same fatty acids in raspberry fruits, (Parry *et al*., 2005; Caidan *et al*., 2013). The results of this study were also consistent with the conclusions of other researchers, who stated that raspberries that are rich in basic phytochemicals have

Table 4. Amino and fatty acids detected in the methanolic extract of red raspberry fruits and % scale.

No.	Amino acids	Peak height, % of scale		Biological characteristic
		Conventional cultivation	Nutrient substrate	
1	Aspartic acid	0.1	–	Amino acid
2	Glutamic acid	0.08	0.08	Amino acid
3	Proline	0.1	–	Amino acid
4	Serine	0.1	–	Amino acid
5	Tyramine	–	0.05	Amino acid
6	Threonine	0.1	–	Amino acid
	Fatty acids	-	-	-
7	α-Linoleic acid	0.1	–	Polyunsaturated omega-6 fatty acid
8	Palmitic acid	0.1	–	Saturated fatty acid
9	Stearic acid	0.1	–	Saturated fatty acid

high antioxidant activity (Andrianjaka-Camps *et al*., 2016; Palonen and Weber, 2019).

CONCLUSIONS

In addition to genotypes, growing technology determined the synthesis of primary and secondary metabolites in red raspberry fruits. Raspberries grown with conventional technology had superior phytonutrient composition and quantitative content and higher antioxidant activity than raspberries grown on nutrient media. Although the total amounts of minerals in both variants were approximately the same, significant differences were observed for individual elements. The present approach can be of great importance in the biochemical screening of red raspberry fruits and the identification of the most valuable genotypes in terms of biochemical composition.

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