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# Physio-chemical characteristics of strawberry and raspberry: comparison of local and commercial cultivars with their wild relatives in Estonia

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**Abstract.** The chemical composition of old local breeds, wild strawberry (*Fragaria vesca*), and raspberry (*Rubus idaeus*) has not been studied enough as they may not be profitable. We compared local and commercial strawberry (*Fragaria* × *ananassa*) and raspberry (*Rubus idaeus*) cultivars to natural relatives. In the experiment fruit weight, pH, and chemical characteristics were investigated. In contrast with foreign cultivars, local strawberry and raspberry cultivars had smaller berries and a lower anthocyanin content, but a higher total phenolic and ascorbic acid content. The strawberry cultivar 'Edu' had the highest ascorbic acid and 'Regatt-80' the highest total phenolic content. The raspberry cultivar 'Tomo' had the highest quantity of ascorbic acid and 'Alvi' the highest total phenolic content. Due to the higher levels of these bioactive compounds, old local cultivars could be used for the production of nutrient-rich functional food. Wild relatives showed a higher value of dry matter, soluble solids, and anthocyanins than the cultivars. The content of total phenolics in raspberry cultivars and wild raspberries was statistically the same but for strawberries, a higher value was estimated for wild counterparts.

Key words: bioactive compounds, nutrition, Fragaria × ananassa, Fragaria vesca, Rubus idaeus, wild berries.

#### 1. INTRODUCTION

Strawberry (*Fragaria* × *ananassa* Duch.) and raspberry (*Rubus idaeus* L.) are highly appreciated crop species. From 1996 to 2016 the production of strawberries has increased 2.5 times (Faostat, 2017). China and the U.S.A. are countries with the highest strawberry production. During the last two decades, raspberry production has doubled, while the harvested area of both strawberry and raspberry has grown 1.5 times. However, in Estonia the strawberry and raspberry harvested areas have been decreasing since 2006: by 2016 these were 565 ha and 250 ha, respectively.

Wild strawberry (*Fragaria vesca* L.) and raspberry (*Rubus idaeus* L.) species are distributed throughout the Northern Hemisphere. In ancient times, before garden strawberry and raspberry cultivars were developed, people used to pick wild berries. These were consumed fresh or for making jams, desserts, juices, or tea (Määttä-Riihinen et al., 2004; Sõukand and Kalle, 2013). Both plants were also used for medical purposes. Wild strawberry leaves were applied to treat digestive disorder (Jarić et al., 2007), eye problems, or wounds (Wagner et al., 2017) and its roots – diarrhoea (Kanodia and Das, 2009). Contrary to *F. vesca*, wild raspberry was used as a cure for cold, cough, and fever (Sõukand and Kalle, 2013). Tea made from raspberry fruit or leaves had a recreational/panacea effect (Pieroni et al., 2017).

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Currently, it has been proved that eating strawberries or red raspberries inhibits the aging processes in living organisms (Giampieri et al., 2017) and can also prevent coronary heart diseases and cancer (Oomah and Mazza, 1998).

It is well known that minerals, vitamins, and other bioactive compounds are obtained by eating fruit. Additionally, more and more people focus on food nutritional quality and its impact on health (Kelt et al., 1997; Milivojević et al., 2011; Najda et al., 2014). Comparison of physical and chemical characteristics of cultivars and their wild counterparts has shown that, in some cases, economically highly valued commercial cultivars (high yield, large berries) have smaller amounts of beneficial antioxidants. For example, it has been demonstrated that the sugar content of cultivated strawberries was two times lower than that of their wild relatives (Milivoiević et al., 2011). Wild strawberry fruit from natural habitats had higher values of anthocyanins (Najda et al., 2014), total phenolics, and antioxidants than the strawberry cultivars (Milivojević et al., 2011; Mikulic-Petkovsek et al., 2012). Commercial raspberries had a higher sugar content in a Serbian research (Milivojević et al., 2011) but a two times lower content of total phenolics than their wild counterparts (Mikulic-Petkovsek et al., 2012). At the same time, the chemical composition of different strawberry and raspberry genotypes can be highly diverse (Arus et al., 2008; Bobinaite et al., 2012; Ahmed et al., 2014; Gündüz and Özdemir, 2014).

It has been pointed out that biological diversity including agro-biodiversity is seriously threatened in the world and strategies to preserve it are required. Crop wild relatives and landraces (also called local cultivars) are the most threatened among plant genetic resources and deserve to be conserved with priority (Pacicco et al., 2018). Therefore, they are an important source of genes that allow crops to adapt to ever-changing climate conditions and to overcome the constraints caused by biotic and abiotic stresses. Also, they consequently are essential for the production of high-quality food in climate change conditions. European Parliament and Council Regulation No. 1305/2013 article 5 clause 4 declares that it is needed to restore, preserve, and enhance biodiversity, high nature value farming, and European landscapes (Publications Office of the European Union, 2013). The EU regulations have stipulated that small fruit crop growers who cultivate old local cultivars will get some financial support. In Estonia, three local strawberry and six raspberry cultivars have been successfully maintained. The chemical composition of old local Estonian cultivars and wild strawberry and raspberry species is not studied enough because they may not be highly profitable. Thus, the nutritional value of these berries is not well known and no data are available on the main bioactive compounds.

The aim of the present study was to compare the physiochemical characteristics of local and commercial strawberry and raspberry cultivars with their natural relatives

#### 2. MATERIAL AND METHODS

#### 2.1. Plant material

In the experiment, different strawberry and raspberry genotypes were studied: three local ('Edu', 'Helean', and 'Regatt-80') and one commercial ('Sonata') strawberry (*Fragaria* × *ananassa*) cultivars, and five domestic ('Aita', 'Alvi', 'Espe', 'Helkal', 'Siveli', and 'Tomo') and one commercial ('Glen Ample') raspberry (*Rubus idaeus*) cultivars. 'Helkal' is a yellow raspberry cultivar, while others are red raspberries. Foreign cultivars 'Sonata' and 'Glen Ample' are widely known and cultivated in Europe, whereas local cultivars are of Estonian origin and have either limited geographical distribution or are purely endemic in Estonia.

The berries of both species were hand-harvested at full ripeness from the germplasm collection garden (*Calcaric* soil) in the Polli Horticultural Research Centre of the Estonian University of Life Sciences (South Estonia, 58°07′44.5′′N, 25°32′16.8′′E). The fruits of strawberries were picked on 22 and 25 June 2016 (cultivar 'Helean') and raspberries were collected on 11 July 2016.

Wild strawberries were picked on 22–26 July from native populations of the following regions:

- Helme Parish, Valga County (58°00'32.5"N, 25°44'18.5"E) (*Stagni-Mollic Gleysol*),
- Karksi Parish, Viljandi County (58°12′34.0′′N, 25°37′17.6′′E) (*Umbri-Densic Podzol*),
- Haaslava Parish, Tartu County (58°16′58.8′′N, 26°47′22.4′′E) (Albeluvisols on noncalcareous reddish-brown till),
- Kambja Parish, Tartu County (58°13′18.7′′N, 26°46′28.5′′E) (*Umbri-Densic Podzol*).

Wild raspberries were picked on 11–12 July from native populations of the following regions:

- Põdrala Parish, Valga County (58°01'35.2"N, 25°57'38.7"E) (Albeluvisols on noncalcareous reddish-brown till),
- Karksi Parish, Viljandi County (58°12′34.0′′N, 25°37′17.6′′E) (*Umbri-Densic Podzol*),
- Haaslava Parish, Tartu County (58°16′54.1′′N, 26°47′16.9′′E) (*Stagnic Luvisol*),
- Kambja Parish, Tartu County (58°13′18.7′′N, 26°46′28.5′′E) (*Umbri-Densic Podzol*).

In 2016 the active plant growth period started on 6 April (average temperature at this time 5.7 °C) (Estonian Weather Service, 2017). The spring was warmer than

usual. In May the average temperature was 14 °C (normal 11 °C). May was the sunniest and driest month – the precipitation sum was only 20.4 mm (normal 48 mm). However, in June the rainfall was very high – 130.7 mm (normal 87 mm). July was the warmest month of summer with the average temperature of 18.1 °C (normal 18 °C) and the rainfall of 75.2 mm (normal 83 mm).

#### 2.2. Fruit weight and preparation of plant material

Fruit weight was determined by a SAC 51 'Scaltec' scale (maximum 200 g, accuracy 0.01 g). Ten randomly chosen fruits were weighed and the average weight for one fruit was calculated. After that, berries were frozen at –20 °C until the analysis (in October 2016). For preparing laboratory samples, fruits (300 g for strawberry cultivars, in other cases 160 g of fruits) were pureed with a hand blender (Turbo MR 5550 M FP, Braun GmbH, Spain) to get homogeneous pulp.

#### 2.3. Dry matter

The oven drying method was used for the determination of dry matter content. Approximately 10 g of fruit puree was heated under specified conditions in a thermostat (Model 400, Memmert). Dry matter was expressed as grams per 100 g of fruit fresh weight (g 100 g<sup>-1</sup>).

# 2.4. Soluble solids, titratable acids, and the ratio between soluble solids and titratable acids (SS/TA)

Soluble solids (SS) content was measured by a Pocket Pal-1 digital hand-held refractometer (Atago). Transparent juice was separated from pureed pulp by centrifugation (centrifuge 2-16PK, Sigma) for 20 min at 4000 rpm and at 14 °C. The juice samples were first allowed to equilibrate to room temperature (22 °C) before analysing. The refractometer was calibrated with distilled water. Soluble solids content was estimated as grams per 100 g of fruit fresh weight.

Titratable acid (TA) quantity was analysed using a standard acid-base titration (Nielsen, 2017). An aliquot of the sample (40 mL) was titrated with 0.1 M NaOH solution to a phenolphthalein endpoint (pH 8.2). EasyPlus Titration (Mettler Toledo) was used for measuring. The content of titratable acids was expressed as citric acid in grams per 100 g of fruit fresh weight.

For fruit taste investigation, the ratio between soluble solids and titratable acids was calculated. The SS/TA ratio is a quality attribute that indicates fruit tartness or sweetness (the higher the ratio is, the sweeter and less tart is the fruit) (Kelt et al., 1997; Mikulic-Petkovsek et al., 2012).

#### 2.5. Fruit pH

Fruit pH was measured in 50 mL of fruit puree, using a FiveEasy F20-Std-Kit (Mettler Toledo) digital pH-meter with an LE438 sensor (accuracy  $\pm 0.01$ ). According to manufacturer's instructions, the pH meter was calibrated using standard buffers of pH 4.01 and pH 7.00.

#### 2.6. Ascorbic acid

On the basis of previous experiments of Raal et al. (2018) the optimal method for extracting ascorbic acid was worked out: to one part of fresh fruit 20 parts of 1% citric acid solution in water at room temperature was added and the plant material was ground in a mortar for 5 min. After that the solution was filtered through a paper filter and centrifuged at 10 000 g for 10 min. After centrifugation the samples were filtered through a 25 mm diameter 0.45 µm acetate membrane filter and ~1 mL aliquots were dispensed into high-performance liquid chromatograph (HPLC) autosampler 2 mL vials. The content of ascorbic acid was calculated in per cent for absolutely dry drug. The loss on drying was determined by European Pharmacopoeia (European Directorate for the Quality of Medicines & Healthcare, 2010) for fresh strawberry and raspberry fruit. The data are presented as the averaged result of four parallel experiments with standard deviation. Ascorbic acid content was determined by the slightly modified European Pharmacopoeia (2010) HPLC method for the quantification of related substances in ascorbic acid as active pharmaceutical ingredient. The details of the used method were published in an earlier paper by Meos et al. (2017). The column and precolumn used were, respectively, aminocolumn (250 mm × 4.6 mm i.d.) with stationary phase particle size of 5 µm (Phenomenex Luna NH2) and SecurityGuard. Elution (flow rate 1.0 mL min<sup>-1</sup>) was performed with acetonitrile and 0.05 M aqueous solution of potassium-dihydrogenphosphate (75:25), with the column temperature of 45 °C and monitored at 260 nm; 10 μl of each sample was injected into the HPLC.

Commercial reagent *L-ascorbic acid* (Sigma, 255564) was used as an ascorbic acid reference substance. Immediately before the assay, ~0.2 mg mL<sup>-1</sup> reference solution was made with 5% (V/V) acetonitrile aqueous solution. To test the linearity of concentration between the standard and the area of the corresponding peak, the calibration curve was established. On that basis, a linear equation was computed and the concentration of ascorbic acid in the solution was calculated. The correlation coefficient between these parameters within the ascorbic acid concentration range from 0.50 to 0.01 mg mL<sup>-1</sup> was close to 1. Finally, ascorbic acid content was expressed in milligrams per 100 grams (mg 100 g<sup>-1</sup>) of fresh berries.

#### 2.7. Anthocyanins and total phenolics

Anthocyanin content was estimated by the pH differential method (Wrolstad et al., 2005), using a spectrophotometer at 510 nm and at 700 nm in buffers at pH 1.0 (HCl 0.1 N) and pH 4.5 (citrate buffer). The extraction solution contained hydrochloric acid (0.1 M) and ethanol (960 mL L<sup>-1</sup>) at a volume rate of 15:85. The weighed quantity (≈10 g) of fruit puree was mixed with the extraction solution (100 mL) and placed under periodical stirring for 2 h at 22 °C. Hermetically capped flasks with extract were held at 5 °C overnight. Test samples were filtered (pore size ≤1.2 µm) before measurement. The results were expressed in milligrams of pelargonidin-3glycoside (for strawberries) or cyanidin-3-glycoside (for raspberries) per 100 g of fresh weight. Spectrophotometric measurements were performed using a UVmini-1240 spectrometer, Shimadzu, Japan) equipped with 4 mL plastic cuvettes (layer thickness 10 mm).

Total phenolic content was measured according to the Folin–Ciocalteu phenol reagent method (Wrolstad et al., 2005) with modifications. Ethanol–acetone (7:3) solution was used as solvent to extract phenolic compounds. Approximately 5 g of fruit puree was extracted with 50 mL of solvent and left for 2 h at room temperature (22 °C) with periodical stirring. Test flasks were hermetically capped and placed in a fridge at 5 °C for 24 h. In the experiment 0.3 mL of the plant extract was mixed with 7.7 mL of distilled water and 0.5 mL of Folin–Ciocalteu reagent (1:1 solution with water). After 1 min 1.5 mL of sodium carbonate (20 g L<sup>-1</sup>) solution was added. Samples were held for 2 h at 22 °C and absorbance was read at 765 nm. Total phenolic content was expressed in milligrams of gallic acid per 100 g of fresh berries.

#### 2.8. Statistical analysis

The fruit samples were taken from ten plants (cultivars) or clones (wild species). All determinations were performed in triplicate, except for ascorbic acid, which was analysed in quadruplicate. The obtained data were expressed as means (± standard deviation; Table 1, Figs 1, 2). The average influence of cultivars and wild species was calculated and analysed as well. An analysis of variance (ANOVA) followed by the Fisher test was used to statistically assess the equality of means.

#### 3. RESULTS

#### 3.1. Fruit weight and dry matter

The fruit weight of Estonian cultivars 'Edu', 'Helean', and 'Regatt-80' was smaller than that of the commercial cultivar

'Sonata' (Table 1). Nevertheless, the average fruit weight of 'Regatt-80' was the highest among local cultivars. Wild relatives had the smallest fruits, in the range of 0.33–0.55 g.

The commercial raspberry cultivar 'Glen Ample' had the highest fruit weight of 3.29 g, followed by the local cultivars 'Espe' (2.92 g) and 'Helkal' (2.88 g) (Table 1). The fruits of wild raspberries were rather small, in the range of 0.50–0.79 g.

The commercial strawberry cultivar 'Sonata' had one of the lowest dry matter contents (9.8 g 100 g<sup>-1</sup>) among cultivars (Table 1). Estonian cultivars 'Helean' and 'Regatt-80' had a higher, but 'Edu' a lower dry matter content than the commercial cultivar 'Sonata'. The value of dry matter content in wild strawberry was higher in comparison with the cultivars.

The commercial raspberry cultivar 'Glen Ample' had the lowest dry matter content (14.1 g 100 g<sup>-1</sup>; Table 1). The local cultivar 'Alvi' showed the highest value (18.2 g 100 g<sup>-1</sup>). Wild raspberry had a higher level of dry matter compared to the cultivars.

### 3.2. Soluble solids, titratable acids, and the SS/TA ratio

Among strawberry cultivars, the Estonian cultivar 'Regatt-80' had the highest and 'Edu' the lowest value of soluble solids, 11.4 g 100 g<sup>-1</sup> and 7.2 g 100 g<sup>-1</sup>, respectively (Table 1). Strawberry cultivars and natural relatives showed no big difference in the content of soluble solids.

Raspberry cultivars 'Helkal' and 'Aita' had the highest content of soluble solids (11.4 g 100 g<sup>-1</sup>, 11.3 g 100 g<sup>-1</sup>; Table 1). The lowest value of soluble solids among cultivars was recorded for 'Espe'. The average soluble solids content in raspberry cultivars was lower than in their wild counterparts.

The local cultivar 'Regatt-80' differed from other strawberry cultivars in a higher quantity of titratable acids (1.52 g 100 g<sup>-1</sup>), while 'Edu' and 'Sonata' displayed the lowest values of 1.16 g 100 g<sup>-1</sup> and 1.18 g 100 g<sup>-1</sup> (Table 1). Natural relatives of strawberry had a higher content of titratable acids than the cultivars.

The raspberry cultivar 'Glen Ample' had the highest content of titratable acids (2.35 g 100 g<sup>-1</sup>), followed by local cultivars 'Siveli' and 'Tomo', 2.28 g 100 g<sup>-1</sup> and 2.14 g 100 g<sup>-1</sup>, respectively (Table 1). The smallest amount of titratable acids was found for the cultivar 'Espe'. The average titratable acid content of raspberry cultivars was higher than that of wild relatives.

The SS/TA ratio was highest in the strawberry cultivars 'Sonata' (8.2) and 'Helean' (8.0), the lowest in 'Edu' (6.3) (Table 1). The SS/TA ratio was higher in strawberry cultivars than in their wild relatives.

The raspberry cultivar with the highest SS/TA ratio was 'Espe' (6.7; Table 1). 'Glen Ample' and 'Siveli' had

**Table 1.** Physio-chemical characteristics (mean  $\pm$  standard deviation) of strawberry (*Fragaria*  $\times$  *ananassa*) and raspberry (*Rubus idaeus*) cultivars, wild strawberry (*F. vesca*), and wild raspberry (*R. idaeus*)

Cultivar/		Fruit weight	Dry matter	Soluble	Titratable	SS/TA	pH of pureed	Ascorbic acid
Wild relative		(g)		solids	acids		fruits	$(mg\ 100\ g^{-1})$
				(SS)	(TA)			
				(g 100 g <sup>-1</sup> )				
Strawberry								
Cultivar	Edu	$5.08 \pm 0.19^{d}$	$8.3\pm0.1^{\rm g}$	$7.2\pm0.1^{\rm g}$	$1.16 \pm 0.03^{d}$	$6.3 \pm 0.2^{d}$	$3.51\pm0.05^a$	$46.0\pm2.5^a$
	Helean	$5.98 \pm 0.24^{c}$	$10.7\pm0.1^{\rm e}$	$10.3\pm0.1^{c}$	$1.29\pm0.08^c$	$8.0\pm0.1^{\rm a}$	$3.34\pm0.01^{bc}$	$31.5\pm2.3^{\rm c}$
	Regatt-80	$6.16 \pm 0.16^{b}$	$11.4 \pm 0.1^{d}$	$11.4\pm0.1^{a}$	$1.52 \pm 0.03^{b}$	$7.4 \pm 0.1^{b}$	$3.27\pm0.02^{cd}$	$39.5\pm4.0^b$
	Sonata	$12.29\pm0.41^a$	$9.8 \pm 0.1^{\rm f}$	$9.7 \pm 0.1^{e}$	$1.18\pm0.05^{\rm d}$	$8.2\pm0.2^a$	$3.36\pm0.10^b$	$33.3 \pm 1.7^{c}$
Wild	Helme	$0.55\pm0.03^{\rm e}$	$13.1 \pm 0.2^{b}$	$9.9 \pm 0.1^{d}$	$1.30 \pm 0.09^{c}$	$7.6 \pm 0.1^{b}$	$3.29 \pm 0.01^{bc}$	$17.5 \pm 1.5^{d}$
	Karksi	$0.47\pm0.02^{\rm e}$	$12.6\pm0.1^{c}$	$9.3 \pm 0.1^{\rm f}$	$1.37\pm0.15^{c}$	$6.8 \pm 0.1^{c}$	$3.18\pm0.02^{\rm e}$	$10.3 \pm 1.1^{e}$
	Haaslava	$0.33\pm0.04^e$	$13.9\pm0.2^{a}$	$9.8 \pm 0.2^{de}$	$1.71 \pm 0.04^{a}$	$5.8 \pm 0.1^{e}$	$3.22\pm0.01^{de}$	$6.8 \pm 0.1^{e}$
	Kambja	$0.37\pm0.01^{\text{e}}$	$14.1\pm0.1^{\rm a}$	$10.7 \pm 0.1^{b}$	$1.51 \pm 0.05^{b}$	$7.0\pm0.2^{\rm c}$	$3.30\pm0.02^{bc}$	$7.0\pm0.6^{\rm e}$
Average	Cultivar	$7.38 \pm 0.25^{A}$	$7.5 \pm 0.1^{B}$	$9.7 \pm 0.1^{B}$	$1.29 \pm 0.05^{B}$	$7.5 \pm 0.1^{A}$	$3.37 \pm 0.04^{A}$	$37.6 \pm 2.6^{A}$
	Wild	$0.43\pm0.02^{\mathrm{B}}$	$10.1\pm0.2^{\mathrm{A}}$	$9.9\pm0.1^{\rm A}$	$1.47\pm0.08^{\mathrm{A}}$	$6.8\pm0.1^{\rm B}$	$3.25\pm0.01^{\mathrm{B}}$	$10.4\pm0.8^{\mathrm{B}}$
Raspberry								
Cultivar	Aita	$2.55\pm0.30^{bc}$	$15.7\pm0.4^{ef}$	$11.3 \pm 0.1^{\circ}$	$1.82 \pm 0.04^{d}$	$6.1 \pm 0.1^{d}$	$2.92\pm0.01^{\rm f}$	$7.0 \pm 0.8^{\rm g}$
	Alvi	$2.55\pm0.30^{bc}$	$18.2\pm0.3^{bc}$	$10.6 \pm 0.1^{\rm f}$	$1.86 \pm 0.08^{d}$	$5.7 \pm 0.1^{e}$	$3.12\pm0.03^{b}$	$14.3\pm1.3^{\rm e}$
	Espe	$2.92 \pm 0.27^{b}$	$15.3 \pm 0.1^{\rm f}$	$10.3\pm0.1^{\rm g}$	$1.54 \pm 0.03^{g}$	$6.7 \pm 0.1^{c}$	$2.97 \pm 0.01^{d}$	$11.8 \pm 1.0^{f}$
	Helkal	$2.88 \pm 0.60^{b}$	$17.4 \pm 0.2^{d}$	$11.4 \pm 0.1^{c}$	$1.87 \pm 0.04^{d}$	$6.1 \pm 0.1^{d}$	$2.98\pm0.01^d$	$19.5 \pm 2.7^{c}$
	Siveli	$2.33\pm0.46^c$	$16.2\pm0.4^{e}$	$10.6 \pm 0.1^{\rm f}$	$2.28\pm0.05^{b}$	$4.7\pm0.1^{\rm h}$	$2.92\pm0.02^{\rm f}$	$27.5 \pm 1.7^{b}$
	Tomo	$2.34 \pm 0.25^{c}$	$16.1 \pm 0.2^{e}$	$10.7\pm0.1^{ef}$	$2.14 \pm 0.05^{c}$	$5.0 \pm 0.1^{\mathrm{g}}$	$2.94 \pm 0.01^{\text{e}}$	$31.5\pm0.6^a$
	Glen Ample	$3.29\pm0.05^a$	$14.1 \pm 0.4^{\rm g}$	$10.8\pm0.1^{\rm e}$	$2.35\pm0.05^a$	$4.6\pm0.1^{h}$	$2.84\pm0.02^{\rm g}$	$17.3 \pm 1.9^{d}$
Wild	Põdrala	$0.65\pm0.08^d$	$18.8 \pm 0.2^{b}$	$11.6 \pm 0.1^{b}$	$1.43 \pm 0.04^{h}$	$8.2\pm0.3^a$	$3.13 \pm 0.01^{b}$	$4.0\pm0.8^{\rm h}$
	Karksi	$0.79\pm0.01^{d}$	$19.9 \pm 0.1^{\rm a}$	$12.9\pm0.2^a$	$1.66\pm0.05^{\rm f}$	$7.8 \pm 0.2^{b}$	$3.15\pm0.01^{\rm a}$	$4.0\pm0.1^{\rm h}$
	Haaslava	$0.50\pm0.06^{d}$	$15.9\pm0.3^{\rm e}$	$9.6 \pm 0.1^{h}$	$1.81 \pm 0.05^{d}$	$5.3\pm0.1^{\rm f}$	$3.01\pm0.02^c$	$2.5\pm0.6^{\rm h}$
	Kambja	$0.73\pm0.02^{\rm d}$	$17.8 \pm 0.1^{cd}$	$11.0\pm0.1^{\rm d}$	$1.74\pm0.07^{\rm e}$	$6.3\pm0.2^{d}$	$3.02\pm0.02^{c}$	$6.3 \pm 1.2^{g}$
Average	Cultivar	$2.69 \pm 0.32^{A}$	$16.1 \pm 0.3^{B}$	$10.8 \pm 0.1^{B}$	$1.98 \pm 0.05^{A}$	$5.6 \pm 0.1^{B}$	$2.96 \pm 0.01^{B}$	$18.4 \pm 1.4^{A}$
-	Wild	$0.70\pm0.04^{\mathrm{B}}$	$18.1\pm0.2^{\mathrm{A}}$	$11.3 \pm 0.1^{A}$	$1.66 \pm 0.05^{B}$	$6.9 \pm 0.2^{\mathrm{A}}$	$3.08\pm0.01^{\mathrm{A}}$	$4.2\pm0.6^{\rm B}$

Different letters (a–h) indicate significant (p<0.05) difference among all cultivars and wild relatives. Different letters (A, B) indicate significant (p<0.05) difference between average values of cultivars and wild species.

very tart berries: their SS/TA ratios were the lowest in the experiment, 4.6 and 4.7, respectively. Wild raspberries tended to have a higher SS/TA ratio than the cultivars.

#### 3.3. Fruit pH

Only small differences were recorded in the pH of different strawberry genotypes (Table 1). 'Edu' had statistically the highest pH (3.51) among strawberry cultivars. Natural habitats had a lower pH compared to strawberry cultivars.

The differences in pH were small also among raspberry cultivars (Table 1). 'Alvi' had the highest pH (3.12). The average pH of wild raspberries was higher than that of raspberry cultivars.

#### 3.4. Ascorbic acid

The highest ascorbic acid content in strawberry fruits was recorded for the local cultivar 'Edu' (46.0 mg 100 g<sup>-1</sup>), followed by 'Regatt-80' (39.5 mg 100 g<sup>-1</sup>) and 'Sonata'

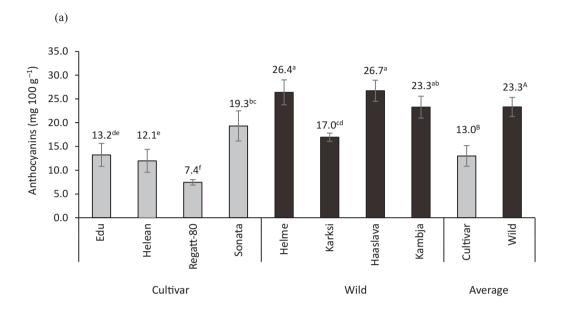
(33.3 mg 100 g<sup>-1</sup>) (Table 1). Wild relatives had a much lower content of ascorbic acid than the cultivars.

Ascorbic acid content varied considerably in different raspberry cultivars (Table 1). The Estonian cultivars 'Tomo', 'Siveli', and 'Helkal' had the highest ascorbic acid contents, 31.5 mg 100 g $^{-1}$ , 27.5 mg 100 g $^{-1}$ , and 19.5 mg 100 g $^{-1}$ , respectively. However, among cultivars, the local cultivar 'Aita' showed the lowest value. The cultivars had a much higher content of ascorbic acid than wild relatives.

#### 3.5. Anthocyanins and total phenolics

The strawberry cultivar with the highest anthocyanin content was 'Sonata' (19.3 mg 100 g<sup>-1</sup>) while 'Regatt-80' had the lowest value (7.4 mg 100 g<sup>-1</sup>) (Fig. 1a). Fruits of wild strawberry had a significantly higher quantity of anthocyanins compared to the cultivars.

Among raspberry cultivars, 'Glen Ample' had the highest anthocyanin content (19.9 mg 100 g<sup>-1</sup>), whereas local cultivars had a slightly lower content (Fig. 1b). Yellow raspberry 'Helkal' was recognized for the lowest anthocyanin



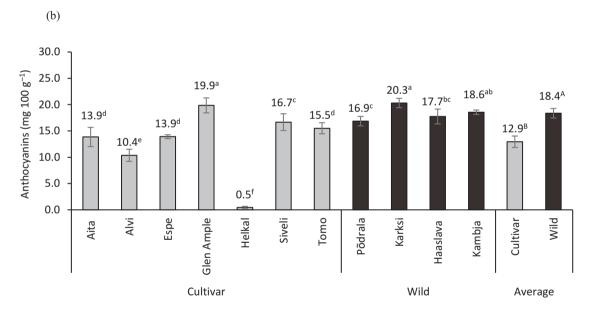


Fig. 1. Anthocyanin content (mean  $\pm$  standard deviation) of strawberry (*Fragaria*  $\times$  *ananassa*) (a) and raspberry (*Rubus idaeus*) (b) cultivars, wild strawberry (*F. vesca*), and wild raspberry (*R. idaeus*). Different letters (a–f) indicate significant (p<0.05) difference among all cultivars and wild relatives. Different letters (A, B) indicate significant (p<0.05) difference between average values of cultivars and wild species.

content. The average anthocyanin content of wild raspberries was 1.4 times higher than that of the cultivars.

The highest total phenolic content in strawberry cultivars was recorded in the local cultivar 'Regatt-80' (553 mg  $100~g^{-1}$ ) and the lowest in 'Sonata' (414 mg  $100~g^{-1}$ ) (Fig. 2a). Wild strawberry had a higher level of total phenolics in comparison with the cultivars.

The Estonian raspberry cultivar 'Alvi' had the highest total phenolic content (Fig. 2b). Yellow raspberry 'Helkal' showed the lowest content of 482 mg 100 g<sup>-1</sup>. Among

raspberry cultivars and wild relatives, no significant differences were observed in the content of total phenolics.

#### 4. DISCUSSION

#### 4.1. Fruit weight and dry matter

The appearance of a product is the most important trait for consumers. More and more attention is paid to fruit weight

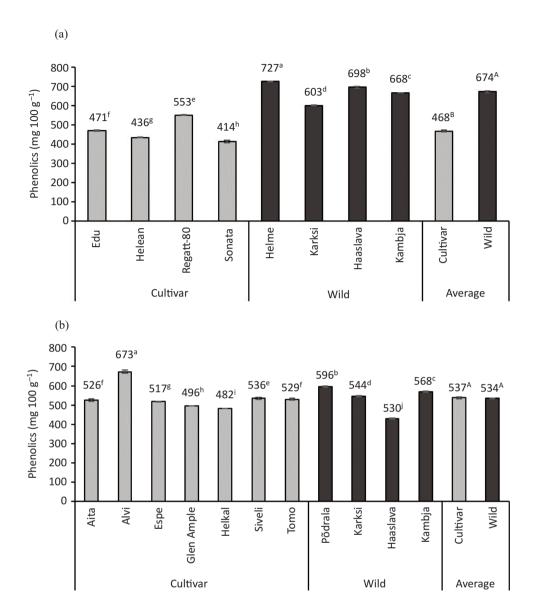


Fig. 2. Total phenolic content (mean  $\pm$  standard deviation) of strawberry (*Fragaria* × *ananassa*) (a) and raspberry (*Rubus idaeus*) (b) cultivars, wild strawberry (*F. vesca*), and wild raspberry (*R. idaeus*). Different letters (a–j) indicate significant (p<0.05) difference among all cultivars and wild relatives. Different letters (A, B) indicate significant (p<0.05) difference between average values of cultivars and wild species.

because customers are eager to buy larger fruit. There was a significant variance in fruit weight among different strawberry genotypes (Table 1). The commercial cultivar 'Sonata' had the highest fruit weight. Typically, the average fruit weight of this cultivar varies from 10.4 to 13.2 g (Polli Horticultural Research Centre, 2014–2015), and in our experiment, it remained within that range as well. The average fruit weight of wild strawberry was 17.1 times lower compared to the cultivars. It has been confirmed by Lithuanian researchers that a significant variation may occur in fruit weight of wild strawberry and there is a high

phenotypic diversity in wild strawberry species (Labokas and Bagdonaitë, 2005). In the present study, this value varied between genotypes almost two times.

An important disadvantage of local cultivars compared to the new ones is a smaller size of fruit, as seen in the present study (Table 1). Local cultivars 'Espe' and 'Helkal' had slightly lower fruit weight in comparison with the commercial cultivar 'Glen Ample'. From previous tests made in Estonia, the fruit weight of two local cultivars 'Helkal' (3.2 g) (Kelt et al., 1997) and 'Aita' (3.1 g) (Arus et al., 2008) has been comparable to

the commercial cultivar 'Glen Ample' in our study. The average fruit weight of wild raspberry was 3.8 times lower than that of the cultivars. In the experimental year, the fruit weight of these strawberry and raspberry cultivars was less than typical. It might have been affected by extremely dry and warm temperatures during the early fruit development stage in May (Estonian Weather Service, 2017). Also in a Canadian study, it has been stated that growing strawberries under plastic mulch could imply a smaller average fruit weight than using a mulching system with plastic mulch with covers (Fan et al., 2012).

The dry matter content of different strawberry genotypes varied greatly (Table 1) and was in the same range as indicated in an earlier study made in Norway (Mazur et al., 2014). The commercial cultivar 'Sonata' had big fruits, but a low content of dry matter. Thus, its fruits mostly consist of water and therefore their nutritional value is relatively low. The average dry matter content of wild strawberry was significantly higher compared to the cultivars. The value of dry matter content in wild strawberry was much lower as mentioned before in an experiment conducted in Poland (Najda et al., 2014).

The average dry matter content in raspberry cultivars in the current study (Table 1) was similar to a study made in Lithuania with different raspberry cultivars, among them 'Siveli' (Bobinaite et al., 2012). Another research (Marjanovic-Balaban et al., 2012), carried out in Bosnia and Herzegovina, provided similar results for wild raspberries. The average dry matter content recorded for raspberry cultivars was significantly lower compared to their wild relatives. Estonian cultivars 'Alvi' and 'Helkal' had a similar dry matter content with wild raspberry. It was found that the commercial cultivar 'Glen Ample' had the lowest dry matter content, and the greatest fruit weight, which again, indicates a lower nutritional value compared to local cultivars.

#### 4.2. Soluble solids, titratable acids, and SS/TA ratio

Soluble solids determine fruit energy value and sweetness because they mainly consist of sugars (Gündüz and Özdemir, 2014). As seen in an earlier research made by Norwegian scientists (Mazur et al., 2014), the content of soluble solids in strawberry cultivars was in a similar range (Table 1). The local cultivar 'Regatt-80' had the highest content of soluble solids. The average soluble solids content of wild strawberry was 1.3 times higher than the average value for the cultivars. It was close to that of two studies made in Turkey and India (Aslantas et al., 2007; Maheshgowda et al., 2016). It has been assured (Gündüz and Özdemir, 2014) that genotype has a significant effect on soluble solids content and individual sugar quantity.

In a study made in Chile (Fredes et al., 2014), a similar soluble solids content was found in raspberry cultivars as

in the current study (Table 1). Local cultivars 'Helkal' and 'Aita' had the highest quantity of soluble solids. In previous tests made in Estonia, 'Helkal' has also had a high level of soluble solids (Arus et al., 2008). The lowest quantity among cultivars was recorded for 'Espe'.

The value of titratable acids for the commercial cultivar 'Sonata' (Table 1) was similar to the result of an experiment made in Estonia, where a foreign cultivar 'Darselect' was tested (Rätsep et al., 2015). A research conducted in India had a significantly lower quantity of titratable acids in seven commercial cultivars (Maheshgowda et al., 2016). A higher average titratable acid content was found in wild strawberry compared to the cultivars (Table 1). It has been ascertained that the content of titratable acids is mostly affected by genotype and growing conditions (Gündüz and Özdemir, 2014).

The highest value of titratable acids was recorded for the commercial cultivar 'Glen Ample' and two local cultivars 'Siveli' and 'Tomo' (Table 1). Even in previous tests in Estonia, the two local cultivars have had a high titratable acid content (Kelt et al., 1997). Raspberry cultivars had a higher content of titratable acids than natural relatives. The titratable acid content for raspberry cultivars was similar to the results obtained from the experiments in Bosnia and Herzegovina (Marjanovic-Balaban et al., 2012).

Strawberry sensory quality depends on the ratio of soluble solids to titratable acid content (Gündüz and Özdemir, 2014). It has been determined that the higher SS/TA ratio of fruit gives a sweeter and less tart taste to the fruit (Kelt et al., 1997; Mikulic-Petkovsek et al., 2012). The SS/TA ratio (Table 1) was similar to the results of the study made in Estonia (Rätsep et al., 2015). However, a Greek study (Zeliou et al., 2018) revealed even higher values for the sugar to acid ratio. 'Sonata' and 'Helean' had the highest value of SS/TA. Cultivars had a higher ratio than natural relatives. Some differences in the SS/TA ratio are due to light intensity and temperature parameters (Maheshgowda et al., 2016). In cooler climate conditions, berries contain less soluble solids and more titratable acids (Remberg et al., 2010; Rätsep et al., 2015).

The SS/TA ratio of raspberry cultivars in the present study (Table 1) was similar to the result of an experiment conducted in Slovenia (Mikulic-Petkovsek et al., 2012). The berries that taste tart generally contain a high level of acids (Mikulic-Petkovsek et al., 2012). The values obtained in the present study concerning the commercial cultivar 'Glen Ample' confirmed this hypothesis. Several studies show that the level of malic acid is higher in fruits of wild berries than in cultivars. For example, it was found in a Serbian study that wild strawberries contain approximately twice and wild raspberries four times more malic acid than cultivars (Milivojević et al., 2011). High malic acid content

also points to the reason why the SS/TA ratio was lower in wild species. In our study, the trend was not similarly regular for both species; it was seen only for strawberries.

#### 4.3. Fruit pH

It is important to control pH to prevent colour and texture loss of food products (Andrés-Bello et al., 2013). A lower pH value prolongs fresh fruit shelf life and inhibits the multiplication of microorganisms. Our study revealed only small variations in the pH of strawberry cultivars (Table 1). The pH of wild strawberry recorded by us was lower than that found in a study made in Turkey (Aslantas et al., 2007).

The pH of raspberries was slightly lower in our study (Table 1) than the results of a study made in Pakistan (Ahmed et al., 2014). A Norwegian study concerning 'Glen Ample' has confirmed that fruit weight correlates negatively with dry matter, soluble solids, titratable acids, and pH (Remberg et al., 2010). In our experiment, only pH had such relationship.

#### 4.4. Ascorbic acid

The local cultivar 'Edu' had the highest quantity of ascorbic acid (Table 1). The ascorbic acid content in our study was lower than in a study carried out in Bosnia and Herzegovina (Marjanovic-Balaban et al., 2012). The average content of ascorbic acid in wild strawberry was 3.6 times lower than in the cultivars (Table 1) but higher than the value obtained in an experiment in Turkey (Aslantas et al., 2007). It has been reported that ascorbic acid content may differ due to genetic variability and acclimatization of genotypes to the growing area (Maheshgowda et al., 2016).

Ascorbic acid content varied considerably in different raspberry cultivars (Table 1) and was compatible with a study carried out in Lithuania (Bobinaite et al., 2012). The highest ascorbic acid content in raspberry fruits was recorded for the cultivar 'Tomo' and the lowest for 'Aita'. Wild raspberry had a considerably (more than four times) lower ascorbic acid content than the cultivars.

#### 4.5. Anthocyanins and total phenolics

The anthocyanin content of strawberries (Fig. 1a) was rather similar to the data obtained in Estonia (Rätsep et al., 2015). In the current study, the cultivar 'Sonata' had a similar content of anthocyanins as described in a study made in Norway with the same cultivar (Aaby et al., 2012). A higher content of anthocyanins was recognized in natural relatives compared to the cultivars. The studies by Aaby et al. (2012) and Gündüz and Özdemir (2014) confirm that anthocyanin content varies in different genotypes. However, pelargonidin derivates are the major anthocyanins in both wild and cultivated strawberry (Määttä-Riihinen et al., 2004; Aaby et al., 2012).

Red raspberries contain a wide spectrum of anthocyanins, mostly cyanidin and pelargonidin glycosides (Määttä-Riihinen et al., 2004). The highest quantity of anthocyanins was found in the commercial cultivar 'Glen Ample' and lowest in the local cultivar 'Helkal' (Fig. 1b). 'Helkal' was expected to have the lowest anthocyanin content because yellow raspberries are known to contain fewer anthocyanins (Määttä-Riihinen et al., 2004; Bobinaite et al., 2012). In our study, total phenolic content was close to the mentioned maximum level in yellow raspberry.

The Estonian raspberry cultivar 'Regatt-80' had the highest level of total phenolics, while the commercial cultivar 'Sonata' had the lowest level (Fig. 2a). A higher total phenolic content in strawberry was reported by a study made in Chile (Fredes et al., 2014). However, a research made in Canada showed a large range in the content of total phenolics in different strawberry genotypes (Xie et al., 2014). Strawberry cultivars had a lower total phenolic content than their natural relatives, as reported by Slovenian researchers (Mikulic-Petkovsek et al., 2012). It has been found (Aaby et al., 2012; Gündüz and Özdemir, 2014) that total phenolic content is significantly different between genotypes. The difference may be due to geographic origin or environmental factors (Gündüz and Özdemir, 2014; Rätsep et al., 2015; Zeliou et al., 2018).

The highest content of total phenolics was estimated for the local cultivar 'Alvi' and the lowest for 'Glen Ample' and yellow raspberry 'Helkal' (Fig. 2b). The Lithuanian researchers Bobinaite et al. (2012) showed that the value in yellow fruited raspberry 'Beglianka' is 391 mg 100 g<sup>-1</sup>. Our results confirmed the results of Bobinate et al. (2012) and Määttää-Riihinen et al. (2004) that yellow raspberry cultivars can also exhibit remarkable levels of total phenolics while being low in anthocyanins. Like in an experiment made in Slovenia (Mikulic-Petkovsek et al., 2012), the average total phenolic contents in raspberry cultivars and their wild counterparts were similar in our study. It was found by Serbian researchers that wild raspberry had a higher total phenolic content (Milivojević et al., 2011), but this trend was not observed in the current study. Wild raspberry showed a pronounced ellagitannin content and conjugated forms of ellagic acid compared to cultivated red raspberries (Määttä-Riihinen et al., 2004).

#### 5. CONCLUSIONS

The current study presents new data on the chemical composition, especially total phenolic and anthocyanin content of old local Estonian strawberry and raspberry cultivars. Wild strawberries had many advantages over the chemical composition of cultivars, like a high level of dry matter, soluble solids, titratable acids, anthocyanins, and total phenolics. Although raspberry cultivars had a high

concentration of some healthy compounds, they contained less dry matter, soluble solids, and anthocyanins than wild raspberry. There was no difference in the total phenolic content compared to the average values of cultivars and wild raspberries.

In contrast with commercial cultivars, all local cultivars had smaller berries, but according to dry matter content, these had higher energy and nutritional value. Moreover, old local cultivars were valuable sources of different antioxidants. Local cultivars had a lower anthocyanin content, but a higher total phenolic and ascorbic acid content compared to common, commercial cultivars. Strawberry cultivars 'Edu', 'Regatt-80' and raspberry cultivars 'Helkal', 'Siveli', and 'Tomo' had a higher ascorbic acid and total phenolic content than the commercial cultivars 'Sonata' and 'Glen Ample' and they should be more appreciated. In order to produce food, richer in bioactive substances, old local cultivars could be more appreciated and used in commercial berry production. These berries can be a good raw material for the production of functional food.

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## Vaarika ja maasika biokeemilised omadused: võrdlus kohalike ja kommertssortide ning looduslike liikide vahel

#### Mailis Vinogradov, Liina Arus, Ain Raal, Andres Meos ja Marge Starast

Üha enam on hakatud tähelepanu pöörama loodusliku elurikkuse hoidmisele, säilitades vanu sorte. Paraku pole selliste aretiste kasvatamine populaarne, sest sageli võivad neil olla väiksem saagikus ja väiksemad viljad. Samas on vanade maasikaja vaarikasortide keemilist koostist suhteliselt vähe uuritud ning bioaktiivsete koostisosade sisaldus pole teada. Käesoleva uuringu eesmärgiks oli võrrelda omavahel Eestis aretatud vanu maasika- (*Fragaria* × *ananassa*) ja vaarikaaretisi (*Rubus idaeus*) nende looduslike liikidega. Katse käigus viidi läbi mõõtmised (vilja mass, pH) ja määrati kuivaine, rakumahla kuivaine, tiitritavate hapete, askorbiinhappe, antotsüaanide ning fenoolide sisaldus. Võrreldes välismaiste sortidega 'Sonata' ja 'Glen Ample', olid kohalikud maasika- ja vaarikasordid väiksema vilja massi ning antotsüaanide sisaldusega, kuid suurema fenoolide ja askorbiinhappe sisaldusega. Eesti maasikasort 'Edu' oli suurima askorbiinhappe ja 'Regatt-80' suurima fenoolide sisaldusega. Kohalik vaarikasort 'Tomo' oli suurima askorbiinhappe ja 'Alvi' suurima fenoolide sisaldusega. Vanade kohalike sortide suuremast bioaktiivsete ainete sisaldusest tulenevalt võiks neid funktsionaalse toidu tootmiseks kasutada. Looduslikel liikidel oli sortidega võrreldes suurem kuivaine, rakumahla kuivaine ja antotsüaanide sisaldus. Kui vaarikate puhul oli fenoolide sisaldus sortidel ja looduslikul vaarikal samaväärne, siis maasika puhul oli näitaja suurem metsmaasikal (*F. vesca*).