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The effect of genotype and rootstock on polyphenol composition of selected apple cultivars in Estonia

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Abstract

The aim of the two year study, 2006 and 2007, was to find out the concentration of the main polyphenols and the estimated content of total polyphenols in the peel of six apple (*Malus* x *domestica* Borkh.) cultivars: 'Talvenauding', 'Krista', 'Liivi Kuldrenett', 'Lobo', 'Cortland' and 'Antei', grown in Estonia. Additionally, the effect of rootstocks B.396, M.26 and 'Antonovka' seedling on the content of polyphenols of 'Talvenauding' apples was studied. High-performance liquid chromatography (HPLC) was used to quantify the polyphenols: catechin, chlorogenic acid, phloridzin, quercetin, quercetin galactoside and quercitrin, all identified by tandem mass spectrometry (MS/MS).

The content of polyphenols was affected by the genotype, rootstock and also by the weather conditions of the particular year. The largest variation of concentrations between the cultivars occurred in quercetin and quercetin galactoside. 'Krista' and 'Lobo' had a 23-fold difference in quercetin concentration being 117.6 mg 100 g^{-1} FW and 5.8 mg 100 g^{-1} FW, respectively. Polyphenols in the apple peel occurred in the following decreasing order: phloridzin > quercitrin > catechin > chlorogenic acid > quercetin galactoside > quercetin and this order was not affected by the genotype. The cultivars with higher concentration of catechin and flavonols had also higher estimated content of total polyphenols. Among the studied rootstocks, the concentration of polyphenols in 'Talvenauding' apples was higher when vegetative rootstocks were used compared to the seedling rootstock.

Keywords: *Malus* x *domestica*, catechin, chlorogenic acid, phloridzin, flavonols, HPLC.

Introduction

Over the past two decades studies *in vitro* and *in vivo* have shown the beneficial effect of fruits and vegetables on human health (Wang et al., 1997; Joshipura et al., 1999). Much of the protective effect has been attributed to phytochemicals, which are non-nutrient plant compounds such as carotenoids, flavonoids, isoflavonoids and phenolic acids (Kampa et al., 2004). Apples are a rich source of phytochemicals, and epidemiological studies have linked the consumption of apples with reduced risk of some cancers, cardiovascular disease, asthma and type II diabetes (Knekt et al., 2002).

Hydroxycinnamic acid derivatives (chlorogenic acid), flavonols (quercetin glycosides), flavanols (catechin, epicatechin, procyanidins) and dihydrochalcones (phloridizin) are the major phenolics in apples (Lee et al., 2003). The distribution of phenolic compounds varies considerably among different cultivars and also within different tissues (Khanizadeh et al., 2008). Anthocyanins and quercetin glycosides exist exclusively in the apple peel whereas epicatechin, procyanidins, phloridzin and chlorogenic acid are found in both peel and flesh of apples (Hagen et al., 2007). Since the apple peel contains more antioxidant compounds, especially quercetin, the peel may have higher antioxidant activity and higher bioactivity than the flesh (Eberhardt et al., 2000).

According to the reported data the important factors influencing the content of polyphenols in the apple are: cultivar properties, fruit maturity, weather conditions of the harvesting season, processing, agricultural conditions, crop load, development of infection, fruit position within the canopy and geographic location (Awad et al., 2000; Van der Sluis et al., 2001). The effect of rootstock is less discussed. Scalzo et al. (2005) have found that the rootstocks in apricots and peaches play an important role in determining the total amount of phenolic compounds. On the other hand, considering factors influencing apple quality, the list also includes rootstocks. From the health point of view the concentration of polyphenols is considered as an important constituent of apple quality. Hence the influence of rootstock should also be taken into account as an important factor influencing the concentration of polyphenols.

Phenolic compounds are induced in plants by various biotic and abiotic stresses (Dixon, Paiva, 1995). Cold treatments and drought stress cause increases in levels of (-)-epicatechin and quercetin-3-galactoside in *Crataegus laevigata* and *C. monogyna*. These types of treatments also enhance the antioxidant capacity of the shoot extracts, and may be the primary function of these cold-inducible flavonoids (Kirakosyan et al., 2003). Marais et al. (2001) have found that a fluctuating temperature resulted in better colour and higher anthocyanin concentrations for apple fruits harvested from different areas.

For many years, the important goals in apple research in Nordic climate have been finding out cultivars and rootstocks with good winter hardiness and appealing appearance. However, yield quality in respect to the health-beneficial compounds is becoming increasingly important every year. Data about phenolic content in Nordic apple cultivars is almost non-existent and information about the effect of rootstock on polyphenol content is also absent. It could be assumed that the content of polyphenols could be increased by choosing the suitable rootstock and cultivar combination, but there is no data available weather the influence is consistent in different years.

The aim of the two year study, 2006 and 2007, was to find out the concentration of the main polyphenols and the estimated content of total polyphenols in the peel of six apple (*Malus* x *domestica* Borkh.) cultivars: 'Talvenauding', 'Krista', 'Liivi Kuldrenett', 'Lobo', 'Cortland' and 'Antei', grown in Estonia. Additionally, the effect of rootstocks B.396, M.26 and 'Antonovka' seedling on the content of polyphenols in the peel of 'Talvenauding' apples was studied.

Materials and methods

Plant material and agricultural practices. The experiment was carried out in 2006 and 2007 at the Estonian University of Life Sciences' Rõhu Research Center (58°21′ N, 26°31′ E) located in South Estonia. The apple trees for cultivar testing were grafted onto 'Antonovka' seedlings and planted in 1986 with a distance of 4 m between the trees and 8 m between the rows.

The experiment's cultivars (and country of origin) were: 'Talvenauding' (Estonia), 'Krista' (Estonia), 'Liivi Kuldrenett' (Baltic), 'Lobo' (Canada), 'Cortland' (USA) and 'Antei' (Belarus).

The trees for rootstock testing were planted in 2001 with 2 m between the trees and 4 m between the rows. 'Talvenauding' was grafted onto semi-dwarfing rootstock M.26 (England), dwarfing rootstock B.396 (Russia) and vigorous rootstock 'Antonovka' (Russia) seedling.

The ground between the rows was grassed and the rows were treated with herbicides. Soil analysis of the vegetative rootstock experiment orchard showed: pH_{KCl} 7.0, P – 175 mg kg^{-1}, K – 197 mg kg^{-1}, Ca – 2980 mg kg^{-1} and humus – 4.7%. Soil analyses for the cultivar testing orchard showed: pH_{KCl} 6.3, P – 216 mg kg^{-1}, K – 236 mg kg^{-1}, Ca – 2830 mg kg^{-1} and humus 5.9%. Since the soil analyses showed no nutrient deficiencies and the trees were growing well, no mineral fertilizers were used at the experimental plantation in either of the experimental years. For plant protection, the apple trees were sprayed four times during the vegetation period to prevent apple scab using copper oxide chloride at the end of April, cypprodinil in the middle of May, difenoconazole at the end of June and ditianon in the middle of July.

Weather conditions. According to agro climatic regions of Estonia, and also taking into account several studies of microclimate, Kask (2000) has divided Estonia into different regions based on their suitability for fruit production. Rõhu is situated in the Tartu region, where cold winters with very low temperatures (sometimes below −37ºC, in certain areas −39ºC) are typical. A vegetation period free of night frosts is short (130 days) and night frosts may be severe. Snow falls quite late and severe frosts in November without snow cover (below −20ºC or even more on the ground) may damage fruit crops.

In the current experiment, the weather during May, June and July in 2006 was fairly dry as only 49% of the average precipitation for these three months fell (Fig. 1). Only in August 2006, the precipitation was more than the average for this period. Conversely in 2007, the precipitation for May was double the average and the driest month was August. In 2006, the mean air temperature was above the average, being especially high in July: 18.5ºC, (average 16.7ºC) and September 13.1ºC (average 10.4ºC). The mean air temperature in 2007 was close to the average except for May 13.6ºC (average 11.0ºC) and August 17.7ºC (average 15.6ºC).

The duration of sunlight hours in 2006 was longer in July and September and shorter in August than the average, but in 2007 was longer in June and August and shorter in July and September compared to the average.

Polyphenol analysis. In both years 'Krista' and 'Liivi Kuldrenett' apples were harvested in mid-September, 'Antei', 'Talvenauding', 'Cortland' and 'Lobo' in the third ten-day period of September. Samples of 50 first quality fruits per cultivar were picked from the outer periphery of the canopy. Fruits were stored at between 2 and 5ºC and 90… 95% relative humidity in a normal atmosphere storehouse for three months. The analyses of polyphenols were performed on the ripe apples at the end of November.

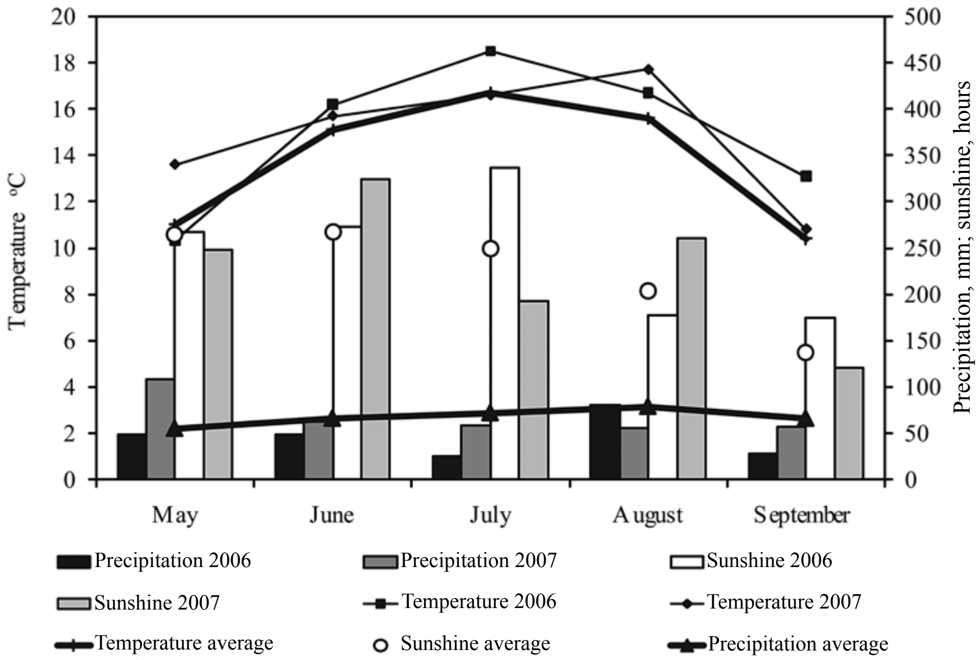


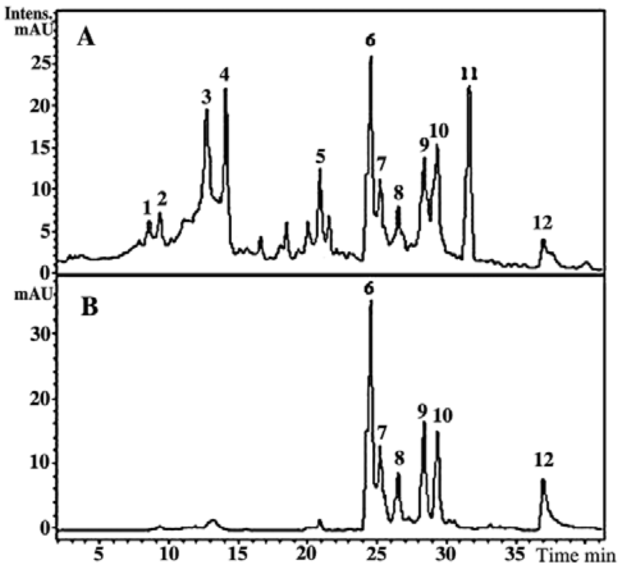
Figure 1. Weather conditions in summer 2006 and 2007 in South Estonia

In the apple peel extract six major polyphenols (catechin, chlorogenic acid (3-*O*-caffeoylquinic), quercitrin (quercetin-3-rhamnoside), quercetin-3-galactoside, quercetin and phloridzin (phloretin-2-*O*-glucoside)) were identified by their chromatographic retention times and MS^2 fragmentation spectra when compared to the respective parameters of commercial standards. Standard chemicals for identifying and quantifying catechin, quercetin, quercetin-3-rhamnoside (quercitrin), quercetin-3-galactoside (hyperin), 3-*O*-caffeoylquinic acid (chlorogenic acid) and phloretin-2-*O*-glucoside (phloridzin) as well as formic acid were sourced from "Sigma-Aldrich Buchs" Co. (Switzerland). Methanol and HPLC grade were obtained from "Rathburn Chemicals" Ltd. (Walkerburn, UK). Six more polyphenols were putatively identified, but not quantified, by their fragmentation spectra. Sample UV-chromatograms at two wavelengths are presented in Figure 2. (-)-epicatechin was not quantified because of the same retention time with cyanidin-3-glucoside.

Peel (approximately 1 mm thick and weighing 0.2 g) at room temperature was cut from ten apples in three replications per cultivar, and rapidly transferred into a tube with 1 ml 0.01 M HCl in methanol. The samples were shaken for 30 minutes at 40 rpm using "Biosan Multi RS60". After shaking, the extract was transferred to another tube before second and third re-extractions of the peel using the same procedure as described above. The total extract (3 ml) was centrifuged by "Eppendorf Centrifuge 5810R" ("Eppendorf" AG, Germany) for 15 minutes at 4000 rpm. The extract was then cooled to 15ºC and 1 ml of extract was filtered through 0.45 µm "Millex-FH" filter and sealed into an airtight glass capsule ("Agilent", USA).

"Agilent 1100 Series HPLC" device equipped with a reversed phase Zorbax 300SB-C18 column (2.1 × 150 mm, 5 µm particle size – "Agilent", USA), photodiode array detector and an electrospray ionization ion trap MS/MS detector ("1100 Series LC/MSD Trap-XCT", "Agilent Technologies", Germany), operated in negative mode ionization (m/z interval 100–1000 amu, target mass – 400 amu) was used for identification and quantification of polyphenols from the filtered peel extract. The column was eluted at 0.3 ml per minute with gradient of 0.1% aqueous formic acid (solvent A) and of acetonitrile (solvent B) from 5 to 95%. The *HPLC 2D ChemStation* software with a *ChemStation Spectral SW* module ("Agilent Technologies", Germany) was used for the process guidance and primary processing of the results. The concentration of polyphenols in the apple peel was calculated according to respective calibration curves obtained using standard compounds and expressed as mg 100 g^{-1} fresh weight (FW). The estimated content of total polyphenols was calculated as the areas under chromatographic curves (AUC) at λ = 280 nm.

Statistical analyses. One-way analysis of variance was used for testing the effect of the rootstock or cultivar on the polyphenols concentration. The means were separated by the least significant difference (LSD) test and differences at $P \leq 0.05$ were considered statistically significant.



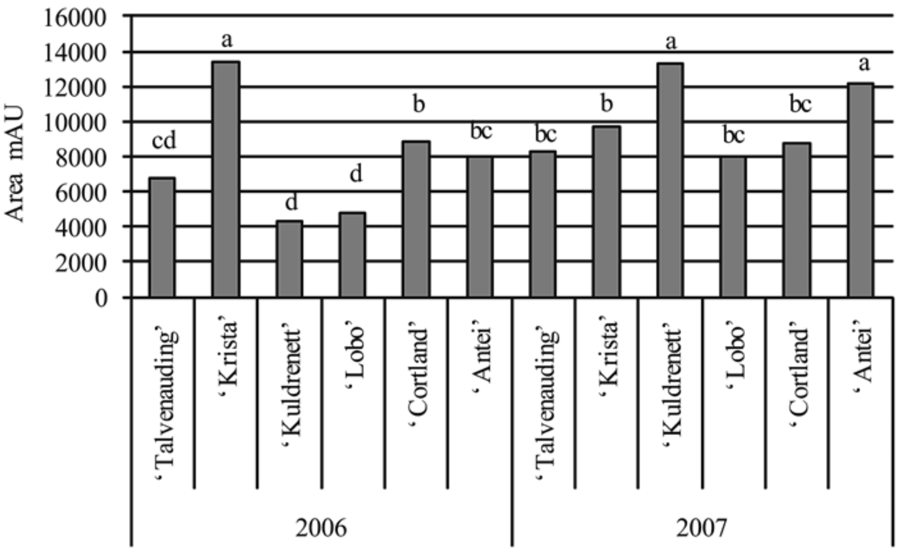
Note. Peak identifications: 1) (+)-catechin ($[M-H]^-$ = 289), 2) chlorogenic acid ($[M-H]^-$ = 353), 3) procyanidin B2 ($[M-H]^-$ = 577), 4) (-)-epicatechin ($[M-H]^-$ = 289) and cyanidin-3-glucoside ($[M-H]^-$ = 447), 5) feroylquinic acid ($[M-H]^-$ = 367), 6) quercetin-3-galactoside ($[M-H]^-$ = 463), 7) quercetin-3-glucoside ($[M-H]^-$ = 463), 8) quercetin pentoside isomer 1 ($[M-H]^-$ = 433), 9) quercetin pentoside isomer 2 ($[M-H]^-$ = 433), 10) quercitrin ($[M-H]^-$ = 447), 11) phloridzin ($[M-H]^-$ = 435), 12) quercetin ($[M-H]^-$ = 301).

Figure 2. UV-chromatograms of the peel extract of 'Antei' (2007) at 280 (A) and 370 (B) nm

Results and discussion

The effect of the genotype. The estimated content of total polyphenols varied significantly among the years and cultivars. In 2006, the total content of polyphenols was significantly higher in 'Krista', but in 2007 in 'Liivi Kuldrenett' and 'Antei' (Fig. 3). No significant differences were seen among other cultivars. The mean effect of the cultivar showed significantly higher content of total polyphenols in 'Krista'.

In the current study, phloridzin and quercitrin were the most abundant polyphenols in the apple peel. Polyphenols occurred in the following decreasing order: phloridzin > quercitrin > catechin > chlorogenic acid > quercetin galactoside > quer-



Note. Mean values followed by the same letter are not significantly different at $P \leq 0.05$.

Figure 3. The estimated content of total polyphenols measured at λ = 280 nm in the apple peel of different cultivars grown in 2006 and 2007 in Estonia

cetin. Genotype had no significant effect on the order of polyphenols in the apple peel. These results are similar to those found by Lu and Foo (1997). D'Abrosca et al. (2007) also found that apple peel was rich in phloridzin, phloretin-2′-xyloglucoside, catechin and epicatechin.

The effect of genotype on the content of polyphenols was significant, but differed between the years (Table 1). In 2006, 'Krista' had significantly higher concentration of phenolic compounds, except for chlorogenic acid and phloridzin, compared to other cultivars. In 2007, 'Krista' had significantly lower concentrations catechin, quercetin and quercetin galactoside and no significant differences among other polyphenols compared to 2006.

Table 1. The concentration of phenolic compounds (mg 100 g^{-1} FW) in the peel of different apple cultivars grown in 2006 and 2007 in Estonia

Cultivars	CAT	CHL	PHL	QERT	QGAL	QTRI
			2006			
'Talvenauding'	59.0g	41.5de	149.8d	17.7c	16.3c	91.5d
'Krista'	123.9b	52.5cd	134.2de	117.6a	50.4a	127.1c
'Liivi Kuldrenett'	45.3g	27.9e	108.0e	5.2d	1.1e	26.9f
'Lobo'	52.2g	25.0e	120.0e	5.6d	1.7de	36.0ef
'Cortland'	70.9f	47.3d	169.9cd	35.0b	17.5c	77.6d
'Antei'	88.8de	41.2de	152.9cde	17.5c	12.5c	64.2de
			2007			
'Talvenauding'	93.0de	131.6a	294.9a	7.0d	33.0b	198.2b
'Krista'	98.0de	69.0c	141.1de	11.1d	37.1b	101.5c
'Liivi Kuldrenett'	118.4bc	123.7a	293.4a	22.5c	16.8c	270.0a
'Lobo'	81.3ef	68.5c	192.9c	5.8d	5.8de	126.4c
'Cortland'	106.0cd	104.6b	279.1ab	6.8d	9.2d	113.7c
'Antei'	157.9a	160.2a	249.5b	8.5d	39.5b	203.4b
$LSD_{0.05}$	18.3	19.2	41.2	7.9	7.9	31.1

Notes. Mean values followed by the same letter are not significantly different at $P \leq 0.05$. CAT – catechin, CHL – chlorogenic acid, PHL – phloridzin, QERT – quercetin, QGAL – quercetin galactoside, QTRI – quercetrin.

In 2006, 'Antei' had significantly lower concentration of catechin and 'Liivi Kuldrenett' significantly lower content of quercetin. However in 2007, the content of those polyphenols was markedly higher compared to other cultivars and the year 2006.

The experiment demonstrated a large difference in the concentration of polyphenols in different apple cultivars showing the important influence of genotype. The largest variation of concentrations between the cultivars occurred in quercetin and quercetin galactoside. In 2006, 'Krista' and 'Lobo' had a 23-fold difference in quercetin concentration. Earlier experiments with 19 apple cultivars in Poland have shown a 9-fold difference in chlorogenic acid (Łata et al., 2009) and an experiment with three apple cultivars in Japan reported a 15-fold difference in catechin content (Kondo et al., 2002). Researchers have indicated the beneficial effect of quercetin on human health (Knekt et al., 2002; Ansari et al., 2009). The current study showed that the concentration of quercetin significantly differed among cultivars and years. In 2006, the concentration of quercetin and quercetrin was significantly higher in 'Krista' and in 2007, in 'Liivi Kuldrenett' (Table 1). The cultivar experiment showed that the apple peel with higher content of total polyphenols had also higher content of flavonols and catechin. Wojdyło et al. (2008) also demonstrated that genotypes with the highest phenolic concentration had simultaneously high content of flavonols.

Yearly different weather conditions had an impact also: a significantly higher concentration of phenolic compounds was found in 2007 (Fig. 3). We can assume that increased content of polyphenols in 2007 was caused by longer sunlight hours, less precipitation and higher temperature in August 2007 compared to the August 2006. Genetic background, developmental stage and environmental factors, such as nutrient availability, temperature and particularly light influence the synthesis of polyphenols (Saure, 1990). Sunlight induces many enzymes involved in flavonoid synthesis (Treutter, 2001). In 2007, all the apples had a better red skin colour and higher polyphenol concentration (except quercetin). Even the yellow apples of 'Liivi Kuldrenett' had a red blush, which might have been due the 68% more sun light hours in August 2007 compared to the same period in 2006. Better light conditions before harvesting probably caused higher polyphenols concentration in the apple peel in 2007. These results are in agreement with Hagen et al. (2007) who found that the red to green colour values of the apple skin correlated very well with the peel content of phenolic compounds and ascorbic acid in 'Aroma' apples. They also implied that the colour of the apple skin might provide useful information about the health value of the apples.

The effect of the rootstock. The area of a peak indicating the estimated content of total polyphenols in the rootstock experiment ranged from 6781 to 15029 mAU min. No significant differences were seen between the years and vegetative rootstocks. The estimated content of total polyphenols was significantly lower only in 'Talvenauding' apples grafted on 'Antonovka' seedlings (data not shown).

The rootstocks significantly influenced the concentrations of different polyphenols in the apple peel (Table 2). Apples grown on vegetative rootstocks had significantly higher concentration of polyphenols compared to those grown on 'Antonovka' seedlings. In 2006, the concentrations of quercetin (82.5 mg 100 g^{-1} FW) and quercetin galactoside (46.3 mg 100 g^{-1} FW) in the apples grown on B.396 were significantly higher compared to the apples grown on M.26 where the concentration of same compounds were 71.7 mg 100 g^{-1} FW and 30.9 mg 100 g^{-1} FW, respectively. In 2007, the content of catechin, quercetin galactoside and quercitrin was higher in apples grown on B.396.

In both experimental years, significantly higher concentrations of phloridzin were detected in the peel of apples grown on vegetative rootstocks (Table 2). Also in both years there were no significant differences between vegetative rootstocks and 'Antonovka' seedling in chlorogenic acid concentration. Significantly higher concentration of quercetin galactoside was measured in apples grown on B.396 in both years. In 2006, more significant differences between vegetative rootstocks and 'Antonovka' seedling occurred. Significantly higher concentrations of catechin, phloridzin, quercetin and quercetin galactoside were measured in apples grown on vegetative rootstocks.

Table 2. The concentration of phenolic compounds (mg 100 g^{-1} FW) in the peel of 'Talvenauding' apples grown on different rootstocks in 2006 and 2007 in Estonia

Rootstock	CAT	CHL	PHL	QERT	QGAL	QTRI
			2006			
Seedling	59.0c	41.5b	149.8d	17.7c	16.3c	91.5d
B396	91.0b	50.8b	217.4c	82.5a	46.3a	116.8d
M26	111.5b	58.8b	195.1c	71.7b	30.9b	128.1d
			2007			
Seedling	93.0b	131.6a	294.9b	7.0d	33.0b	198.2c
B396	143.1a	139.8a	370.0a	16.1cd	43.7a	386.0a
M26	106.8b	129.9a	338.1a	10.6cd	28.8b	289.1b
$LSD_{0.05}$	25.4	23.3	36.5	9.7	8.9	68.0

Notes. Mean values followed by the same letter are not significantly different at $P \leq 0.05$. CAT – catechin, CHL – chlorogenic acid, PHL – phloridzin, QERT – quercetin, QGAL – quercetin galactoside, QTRI – quercetrin.

Previous research conducted in Estonia demonstrated that dwarfing vegetative rootstocks successfully inhibit the growth of vigorous cultivars (Haak, 2003). Since 'Talvenauding' is a vigorous cultivar, the dwarfing effect of B.396 and M.26 might have had a beneficial influence on the concentrations of polyphenols, because canopies with weaker shoot growth provide better light conditions for fruits. According to Hagen et al. (2007) the concentration of flavonoids and the level of total phenols are higher in the peel of sun-exposed apples compared to shade-grown apples.

The rootstocks also significantly influence the intake of mineral elements from the soil. Studies have shown that the content of macronutrients in the leaves depend on the rootstock (Poniedziałek et al., 1993). Sotiropoulos (2008) found a higher concentration of N in the leaves of 'Imperial Double Red Delicious' grown on the seedling compared to the rootstocks M.7 and MM.106. Dris and Niskanen (1998) found a positive correlation between fruit and leaf N concentration and Awad and de Jager (2002) found a negative correlation between nitrogen and apple skin cyanidin-3-galactoside and total flavonoids concentration. Fallahi and Simons (1993) found that trees on M.7 had significantly lower leaf and fruit N contents which resulted in a darker fruit colour than that of fruits from trees on M.26 rootstocks, both at harvest and after storage. The intake of mineral elements from the soil is also significantly influenced by the soil moisture. It is also observed that rootstocks with different vigour have different ability to extract soil water (Lo Bianco et al., 2008). Less vigorous rootstocks have weaker root system and are therefore usually more susceptible to the weather conditions. However, in the current experiment it was observed that apples grown on vegetative rootstocks had higher content of most polyphenols compared to the seedling rootstock irrespective of the yearly different weather conditions.

Conclusions

1. The current study confirmed that the genotype is a major factor influencing the concentration of polyphenols, since a 23-fold difference in polyphenol concentrations between the cultivars was observed. The obtained results may be used in the selection of apple genotypes suitable for Estonia and other countries with similar climate that have improved nutritional quality for fresh consumption. Among the studied cultivars, 'Krista' and 'Liivi Kuldrenett' had significantly higher concentrations of quercetin and quercitrin compared to all other cultivars.

2. The hypothesis about the influence of the rootstock on the apple polyphenols was confirmed: both vegetative rootstocks promoted formation of most polyphenols in 'Talvenauding' fruits. Among dwarfing rootstocks, B.396 was superior compared to M.26 in terms of increasing quercetin galactoside content in the peel of 'Talvenauding' apples. Further research is needed for other rootstocks and other cultivars. Yearly different weather conditions affected polyphenol content in apples, but fruits grown on vegetative rootstocks had higher content of most polyphenols compared to the seedling rootstock irrespective of the weather.

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Genotipo ir poskiepių įtaka atrinktų obels veislių polifenolių sudėčiai Estijoje

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Santrauka

Tyrimų, atliktų 2006–2007 m., tikslas – nustatyti pagrindinių polifenolių koncentraciją ir bendrą polifenolių kiekį Estijoje auginamų šešių obels (*Malus* x *domestica* Borkh.) veislių: 'Talvenauding', 'Krista', 'Liivi Kuldrenett', 'Lobo', 'Cortland' ir 'Antei', vaisių odelėje. Be to, tirta poskiepių B.396 bei M.26 ir veislės 'Antonovka' sėjinukų įtaka polifenolių kiekiui veislės 'Talvenauding' obuoliuose. Efektyvioji skysčių chromatografija (ESCH) buvo taikyta nustatant šių polifenolių kiekį: katechinų, chlorogeninės rūgšties, floridzinų, kvercetinų, kvercetinų galaktozido ir kvercitrinų. Polifenoliai buvo identifikuoti taikant tandeminę masių spektrometriją (MS/MS).
Polifenolių kiekį lėmė veislės genotipas, poskiepis ir metų oro sąlygos. Tarp veislių labiausiai skyrėsi kvercetino ir kvercetinų galaktozido koncentracija. Veislių 'Krista' ir 'Lobo' kvercetino koncentracija skyrėsi 23 kartus ir sudarė atitinkamai 117,6 ir 5,8 mg 100 g^{-1} žalios masės. Obuolių odelėje polifenolių kiekis mažėjančiai pasiskirstė taip: floridizinai > kvercitrinai > katechinai > chlorogeninė rūgštis > kvercetinų galactozidas > kvercetinai; genotipas šiam pasiskirstymui neturėjo įtakos. Veislės, pasižyminčios didesniu kiekiu katechinų ir flavonolų, taip pat turėjo didesnį kiekį polifenolių. Polifenolių koncentracija veislės 'Talvenauding' obuoliuose buvo didesnė vaismedžių su vegetatyviniais poskiepiais, palyginti su vaismedžių su sėkliniais poskiepiais obuoliais.

Reikšminiai žodžiai: *Malus* x *domestica*, katechinai, chlorogeninė rūgštis, floridizinas, flavonolai, ESCH.