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Effect of post-harvest flame-defoliation on strawberry (*Fragaria* × *ananassa* Duch.) growth and fruit biochemical composition

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Abstract

The experiment with strawberry (*Fragaria* × *ananassa* Duch.) cultivar 'Darselect' was carried out at the Research Centre of Organic Farming of Estonian University of Life Sciences (58º21′ N, 26º40′ E). The objective of the present research was to determine the influence of post-harvest defoliation with a directed propane flamer and the combination of flaming and additional application of humic substances on strawberry growth and fruit biochemical composition in two successive years. Defoliation decreased the number of leaves and inflorescences, and crown and root mass. Post-harvest flame-defoliation of two-year-old plants increased the yield in the next year (in 2012), but the succeeding flame-defoliation decreased the yield in the following year (in 2013). The effect of defoliation on fruit biochemical composition was significant, but differed between years and fruit orders. The defoliation treatment increased soluble solids content in primary fruits from 8.9 to 9.3 °Brix, but decreased anthocyanins content in all fruit orders in 2012. Flaming treatment increased the content of phenolics from 181 to 206 mg 100 g^{-1} in primary, but decreased from 268 to 254 mg 100 g^{-1} in tertiary fruits in 2013. Application of humic substance to the defoliated plants increased the number of leaves in 2012, and crown and root mass in 2013, but decreased the yield in both years. The influence of the treatment differed yearly for soluble solids – decreasing the content in 2012, but increasing in 2013. Humic substances had positive influence on the total phenolics and anthocyanins content in both years.

Keywords: anthocyanins, ascorbic acid, humic acids, soluble solids, titratable acids, total phenolics.

Introduction

The tendency towards environmentally friendly cultivation methods producing healthy fruits is constantly increasing. Organically grown strawberries tended to have higher soluble solids/titratable acids ratio indicating somewhat sweeter taste compared to conventional ones (Tõnutare et al., 2009), containing up to 20% more ascorbic acid and phenolics, and anthocyanins (Fernandes et al., 2012). There is data claiming that cultivation systems may have no clear effect on the accumulation of phenolic compounds (Crecente-Campo et al., 2012). Contrary to previous, it has been revealed that environmental conditions affect significantly strawberry quality parameters (Moor et al., 2004; Crespo et al., 2010), and biochemical composition of fruits also (Khanizadeh et al., 2014).

Strawberry plantations are mainly defoliated for renovation and disease control purposes (Wildung, 2000). Experiments with post-harvest defoliation have shown the reducing effect on the incidence of *Botrytis* rot only in one year out of three (Daugaard et al., 2003). Partial leaf removal after planting affects plant leaf rate and photosynthesis related to the physiological changes caused by defoliation (Casierra-Posada et al., 2012). In some cases, post-harvest defoliation treatment has increased the yield in some cultivars (Nestby, 1985), but yields can be affected by changing the extent of defoliation (Albregts et al., 1992). Clearly, it has been found that strawberry plants respond to the post-harvest defoliation differently depending on the cultivar and cropping year (Daugaard et al., 2003; Whitehouse et al., 2009). Some research has been done using the post-harvest straw mulch burning in relation to plant protection which was shown to decrease the occurrence of spider mites (*Tetranycus urticae*) significantly (Metspalu et al., 2000). Flame-defoliation could be used to remove the old strawberry foliage, but this may damage the strawberry crown (Wildung, 2000).

The timing of the defoliation is also important, because strawberry flower bud induction is sensitive to thermo-photoperiod and to some agronomic and nutritional factors (Savini et al., 2005). Plant leaf area is responsible for plant health and vitality due to photosynthesis processes and their influence on the leaf-root ratio. The extent of defoliation treatments was found to affect strawberry flower initiation significantly due to differences in the inductive capacity of leaves of differing maturity (Thompson, Guttridge, 1960). In northern hemisphere, the June-bearing strawberries initiate flower buds form late summer to autumn under short-day conditions. Therefore in cool climate conditions, the defoliation of strawberry plants should be executed at the end of harvest. The growth of new leaves occurs in a week after post-harvest flaming and cultivars with vigorous growth habit are able to recover by the time of flower bud initiation and before dormancy period (Metspalu et al., 2000). Leaves also provide the winter cover in order to protect the buds initiated in the strawberry crown. Flame-defoliation treatment can be an alternative method for mowing, though its effect on strawberry plant growth and fruit quality has been less studied.

For the reason of strawberry plant health and recovery from the defoliation treatment, several studies related to plant nutrition have been conducted. Defoliated plants were found to be affected by fertilization showing decreased content of vitamin C in strawberries (Moor et al., 2004). The utilization of plant bio-stimulants is quite a common practice in organic cultivation. Application of humic acid-containing substances has been used due to their ability to improve plant nutrient availability and therefore plant growth and yield quality (Calvo et al., 2014). Organic amendments such as farm yard manure and vermicomposting-based substances have shown the improving effect on growth-related and yield qualitative parameters (Khalid et al., 2013). Positive effect on organic strawberry fruit quality has been obtained with repetitive humic acid foliar applications from flowering to harvest (Hosseini Farahi et al., 2013). Humic acid treatments have shown a positive effect on assimilation and increased plant nutrient (N and P) uptake (Tehranifar, Ameri, 2014). Any changes in the plant biomass production (such as the number of leaves) may change the nutrient allocation patterns in fruits, which are directly related to yield quality (Correia et al., 2011). Application of liquid humic substances was found to increase strawberry total phenolics, anthocyanins and ascorbic acid contents in strawberry cv. ‘Darselect’ fruits in the following year after planting (Rätsep et al., 2014). On the other hand, the effect of additional humic acid amendments depends on the cultivar, because the role of cultivar properties has been confirmed as the main source of variation in biochemical composition and post-harvest quality (Crespo et al., 2010; Martinez et al., 2015).

In conclusion of the previous, several research problems were pointed out: a directed row flamer could be useful in strawberry plantations in order to remove old leaves causing minor plant damage, but affecting strawberry plant parameters and fruit quality. Moreover, besides environmentally friendly fruit production, under organic cultivation conditions it is possible to obtain considerably high contents of biochemical compounds, in particular total phenolics (Crecente-Campo et al., 2012). The problematic point could be the effect of flame-defoliation on the plant vitality – the post-harvest treatment may damage strawberry plant crowns, therefore affecting strawberry growth, and yield and fruit biochemical composition. For that reason, the application of liquid humic substances was additionally tested on the defoliated plants. There is little data about the effect of defoliation on fruit quality. The objective of the present research was to determine the influence of post-harvest defoliation with a directed propane flamer and the combination of flaming and application of humic substances on strawberry growth and fruit biochemical composition.

Material and methods

Experimental area and treatments. The strawberry experimental plantation was established at the Estonian University of Life Sciences (58º21′ N, 26º40′ E, 68 m a.s.l.). The pre-planting procedures were as follows: weeds were destroyed when the plantation area was covered with black plastic from spring 2009 to the next spring 2010 until ploughing and establishing were carried out; an ecological fertilizer (NPK 4.5-2.5-8) – produced from at least 30% malt germs – was used as pre-establishment soil supplement. Strawberry cultivar ‘Darselect’ frigo-plants were planted in May, 2010 in a one-row system with 50 cm spacing. Strawberry plantation area (including rows) was mulched after planting with 3–5 cm thick organic apple tree bark mulch layer made of leftover branches of organic apple tree canopy pruning. The experimental layout was a randomized block design with three replicates (12 plants per replication).

Flaming was executed in 2011 and 2012, and the following treatments were implemented: control (C), flame-defoliation (FD) and flame-defoliation with humic acid (FD + H) treatment. Control plants were neither defoliated nor fertilized, but water was applied to the control plants at the same time when the other plants received humic substances. Post-harvest FD was executed with a directed row flamer by burning all the leaves of treated plants after fruit harvesting in the second (2011) and third (2012) growth year. The flamer is a product of “Elomestari” (Finland). The machine is working with propane and it has two 20 cm wide burners with covers controlling the heating of the row flamer including mounting for selective (directed) flaming.

Soil and weather conditions. The plantation soil is *Stagnic Luvisol* (FAO soil classification). The soil pHKCl was 6.6 at the time of plantation establishment and humus content was 4.9%. The content of P, K and Mg was high and Ca content was high in the soil (Table 1).

Table 1. The content of soil nutrients, carbon and humus, pH and carbon content in biomass in 2012

Treatment	pH_{KCl}	P mg kg^{-1}	K mg kg^{-1}	Ca mg kg^{-1}	Mg mg kg^{-1}	C %	Humus %	C in biomass C mg^{-1} DW
Control	6.6	256	306	3930	600	3.2	4.9	0.525
Fertilized	6.7	286	347	5131	492	3.0	5.2	0.766

DW – dry weight

Table 2. Average temperatures and total monthly precipitation of 2011–2013 and of long-term average (1971–2000)

Month	Air temperature °C				Precipitation mm			
	2011[1]	2012[1]	2013[1]	1971–2000[2]	2011[1,2]	2012[1]	2013[1,2]	1971–2000[2]
April	5.7	4.6	4.0	4.7	11	45	36	33
May	11.0	11.4	15.5	11.1	58	78	65	53
June	17.2	13.3	17.8	15.1	35	98	29	69
July	20.0	17.7	17.5	16.9	48	80	67	76
August	15.9	14.8	16.6	15.6	55	80	73	80
September	12.3	11.9	10.8	10.4	80	61	38	67

Note. [1] – data was collected from automatic weather station of experimental garden, [1] – data according to the Estonian Weather Service (www.ilmateenistus.ee) database.

In 2011, the temperatures from March to May were 0.5°C warmer than in 1971–2000. The meteorological summer months (from June to August) were remarkably warm exceeding the long-term temperatures up to 2.4°C compared to. Monthly average temperatures in July, August and September were higher than long-term mean, up to 3.1, 0.3 and 1.9 °C, respectively (Table 2). The sum of active temperatures was 1828°C. Precipitation sum was 292 mm which was only 38 mm lower than the long-term mean (330 mm). Rainfall in June, July and August was somewhat lower – 34, 28 and 25 mm, respectively, than many years mean precipitation.

In 2012, the period of active temperatures (above 10°C) started at the beginning of May and ended on the 6th of October. Compared to long-term average, monthly average temperatures were warmer in May, July and September, but the temperatures in June were 1.8°C lower than in 1971–2000 (Table 2). The sum of active temperatures in 2012 was 1967°C. The amount of precipitation was somewhat higher in April, May and June (up to 12, 25 and 29 mm more), but quite at the same level in July, August and September in comparison with many years mean.

In 2013, the active plant growth period started on the 7th of May and ended on the 23rd of September. The sum of active temperatures was 2263°C which is 332°C more than many years average (1936°C). Monthly average temperatures were up to 4.4°C warmer, except April being 0.7°C cooler than long-term mean (Table 2). The precipitation sum from 1 April to 31 October was 352 mm, which was 152 mm less than in 2012 and 86 mm less than the mean (438) of 1971–2000. Rainfall was a bit higher only in April and May compared to long-term mean, while all the other months had lower precipitation rate and in June and September it was even up to 40 and 29 mm lower, respectively.

Measurements and analysis. Strawberry plant growth, yield and fruit composition were investigated in 2012 and 2013. Fruits were picked according to surface colour and fruit order (primary, secondary, tertiary) in clusters, fruit mass and total yield per plant were determined at the same time. Leaves and inflorescences were counted during flowering (in May) in both experimental years. Ten normally developed fully expanded leaves per replication were selected for determining chlorophyll content by using a portable SPAD-502 (Soil Plant Analysis Development) chlorophyll meter ("Minolta", Japan). This method permits a rapid and non-destructive determination of leaf chlorophyll content by measuring leaf transmittance. The dry weight of roots and crowns was determined. In 2013, three plants per replication were dug out; the leaves were cut, and after cleaning from soil the plant crowns and roots were dried until a constant weight was recorded.

All the fruits were picked at the same intervals and in one day for every fruit order and for all the treatments in one year. Yield samples were stored at −20ºC until the analysis. Fruits were analysed separately according to fruit orders. Soluble solids content (SSC) was measured by a refractometer Pal-1 ("Atago Pocket", Japan). Titratable acids content (TAC) was determined by titration method (Mettler Toledo DL 50 Randolino) with aqueous 0.1 M NaOH solution, using phenolphthalein as an endpoint indicator. The TAC was expressed as citric acid g per 100 g of fresh fruits. Soluble solids and titratable acids ratio (SSC/TAC) was calculated. Ascorbic acid content (AAC) was determined iodometrically with the modified Tillman's method. For analysis, a mixture of meta-phosphoric and acetic acid (3% HPO$_3$ + 8% CH$_3$COOH) was added instantly to the pulp to avoid ascorbic acid breakdown in the air. AAC was expressed as mg 100 g^{-1} of strawberry fresh weight (FW). The content of total anthocyanins (ACC) was estimated by a pH differential method. Absorbency was measured with a spectrophotometer UVmini-1240 ("Shimadzu", Japan) at 510 and at 700 nm in buffers at pH 1.0 (HCl 0.1 N) and pH 4.5 (citrate buffer). The results were expressed as mg of pelargonidin-3-glucoside equivalent to 100 g of FW. Total phenolics content (TPC) was determined with the Folin-Ciocalteau phenol reagent method, using a spectrophotometer UVmini-1240 ("Shimadzu", Japan) at 765 nm. TPC was expressed as mg of gallic acid equivalents to 100 g FW of pulp.

Statistical analysis. Strawberry growth related parameters and the effect of treatments according to fruit order were tested by one-way analysis of variance (*ANOVA*), and to evaluate the effect of treatment, the least significant difference was calculated and different letters in tables mark significant differences at $P \leq 0.05$. To evaluate mean effects of the two factors (fruit order in general and treatment as such in average of two experimental years), the two-way analysis of variance (*ANOVA*) was carried out, and marked as non-significant (ns) or using confidence levels as significant at $P \leq 0.05$*, 0.01** or 0.001***, signifying levels of 95, 90 and 99.9 %, respectively. Standard deviations of the mean values are also presented (±SD). Linear correlation coefficients were calculated between variables (n = 7) with the significance of coefficients being $P \leq 0.05$* and $P \leq 0.01$**. The strength of the relationships was estimated as $r \leq 0.3$ (weak), $0.3 \leq r \leq 0.7$ (moderate) and $r \leq 0.7$ (strong). All the data analysed in the present experiment met the assumptions of normality and no additional transformations were made.

Results

Strawberry growth and yield. Flame-defoliation (FD) decreased the leaf number and fruit mass in both experimental years (Table 3). Flaming increased the yield in 2012, but decreased the yield in 2013. SPAD values were decreased by the FD in 2013 only. FD reduced strawberry crown and root mass by up to 67% decreasing from 447 g to 148 g. Humic amendments had positive effect on the fruit mass of defoliated plants in 2012, but negative effect on the number of inflorescences in 2013, and decreased the yield by up to 22% in both years. Application of humic substances increased the crown and root mass of defoliated plants.

Table 3. The effect of flame-defoliation and humic acid treatments on the strawberry cv. 'Darselect' plant and fruit parameters

Year	Treatment	Leaves No. plant^{-1}	Inflorescences No. plant^{-1}	SPAD values	Fruit mass g	Yield g plant^{-1}	Crown and root mass g plant^{-1}
2012	C	40 ± 2.2 a	11 ± 2.2 a	36 ± 1.2 a	31 ± 1.3 a	503 ± 25 b	–
	FD	25 ± 2.3 b	10 ± 1.3 a	35 ± 2.2 a	26 ± 0.6 b	567 ± 28 a	–
	FD + H	38 ± 2.2 a	11 ± 2.3 a	35 ± 2.5 a	30 ± 0.7 a	444 ± 22 c	–
2013	C	62 ± 8.1 a	19 ± 2.1 a	33 ± 1.5 a	15 ± 0.7 a	227 ± 11 a	447 ± 42 a
	FD	31 ± 9.7 b	15 ± 4.2 a	32 ± 2.0 b	13 ± 0.7 b	215 ± 11 b	148 ± 7 c
	FD + H	42 ± 3.7 b	12 ± 1.2 b	32 ± 1.8 b	13 ± 0.7 b	189 ± 10 c	320 ± 40 b

Notes. C – control, FD – flame-defoliation, FD + H – flame-defoliation + humic acids application; different letters mark significant differences at $P \leq 0.05$ according to treatment and characteristics. Standard deviations of the mean values are also presented (±SD).

Strawberry fruit biochemical composition. The influence of defoliation on soluble solids content (SSC) was significant in both experimental years (Table 4). In primary fruits, FD increased the SSC in 2012, but decreased the content in the next experimental year. FD + H decreased SSC by up to 19% in primary and tertiary fruits in 2012, but increased the content by up to 50% in 2013. In average of two years, the mean effect of fruit order was non-significant, while the significance of treatments was at $P \leq 0.001$.

Table 4. The effect of flame-defoliation and humic acid treatments on the strawberry cv. 'Darselect' fruit biochemical composition according to fruit order and the mean effects of treatment and fruit order (2012–2013)

	Fruit order	Treatment	Soluble solids, °Brix	Titratable acids g 100 g^{-1} FW	Soluble solids / titratable acids	Ascorbic acid mg 100 g^{-1} FW	Total phenolics mg 100 g^{-1} FW	Anthocyanins mg 100 g^{-1} FW
2012	Primary	C	8.9 ± 0.2 b	1.03 ± 0.1 b	8.6 ± 0.2 a	116 ± 3.3 a	149 ± 0.2 b	13.2 ± 0.1 b
		FD	9.3 ± 0.1 a	1.36 ± 0.1 a	6.8 ± 0.1 c	103 ± 1.9 b	149 ± 0.2 b	8.5 ± 0.3 c
		FD + H	8.5 ± 0.1 c	1.02 ± 0.1 b	8.4 ± 0.1 b	103 ± 3.2 b	169 ± 0.1 a	14.0 ± 0.2 a
	Secondary	C	8.1 ± 0.1 c	1.02 ± 0.1 b	7.9 ± 0.4 b	92 ± 4.3 a	195 ± 0.2 b	18.2 ± 0.3 b
		FD	8.4 ± 0.2 b	1.14 ± 0.1 a	7.3 ± 0.3 b	99 ± 1.9 a	214 ± 0.4 a	16.4 ± 0.2 c
		FD + H	8.5 ± 0.1 a	0.96 ± 0.1 b	8.9 ± 0.3 a	97 ± 1.6 a	191 ± 0.7 c	19.0 ± 0.2 a
	Tertiary	C	10.9 ± 0.5 a	0.84 ± 0.1 b	13.0 ± 0.9 a	102 ± 5.6 a	195 ± 0.7 b	23.3 ± 0.3 b
		FD	9.6 ± 0.1 b	0.97 ± 0.1 a	10.0 ± 0.5 c	74 ± 3.0 b	178 ± 0.7 c	18.0 ± 0.2 c
		FD + H	8.8 ± 0.1 c	0.80 ± 0.1 b	11.1 ± 0.5 b	65 ± 4.1 c	208 ± 0.4 a	23.6 ± 0.2 a
2013	Primary	C	9.8 ± 0.1 b	1.20 ± 0.1 a	8.2 ± 0.4 b	83 ± 2.1 a	181 ± 0.8 b	6.8 ± 0.3 b
		FD	8.8 ± 0.7 c	1.15 ± 0.1 b	7.6 ± 0.7 b	81 ± 5.4 a	206 ± 2.3 a	9.4 ± 0.5 a
		FD + H	11.0 ± 0.1 a	1.15 ± 0.1 b	9.5 ± 0.3 a	81 ± 1.5 a	183 ± 3.9 b	6.5 ± 0.2 b
	Secondary	C	9.3 ± 0.1 c	1.37 ± 0.1 a	6.8 ± 0.2 a	104 ± 2.0 a	212 ± 1.7 c	10.3 ± 0.4 a
		FD	9.6±0.1 b	1.31 ± 0.1 a	7.3 ± 0.4 a	91 ± 2.9 b	229 ± 1.1 b	10.4 ± 0.3 a
		FD + H	10.0±0.4 a	1.31 ± 0.1 a	7.6 ± 0.4 a	68 ± 21 c	233 ± 3.0 a	10.5 ± 1.2 a
	Tertiary	C	8.3 ± 0.2 b	1.58 ± 0.1 a	5.2 ± 0.1 b	72 ± 2.5 b	268 ± 2.1 b	9.5 ± 0.3 c
		FD	7.8 ± 0.5 c	1.49 ± 0.1 a	5.3 ± 0.5 b	73 ± 2.0 b	254 ± 2.1 c	10.5 ± 0.4 b
		FD + H	11.7 ± 0.4 a	1.47 ± 0.1 a	8.0 ± 0.4 a	87 ± 3.4 a	280 ± 0.5 a	15.4 ± 0.6 a
Mean effect of fruit order			ns	*	**	***	***	***
Mean effect of treatments			***	***	***	***	***	***

Notes. C – control, FD – flame-defoliation, FD + H – flame-defoliation + humic acids application. Different letters mark significant differences at $P \leq 0.05$ among control and different treatments in different fruit orders; ns, *, **, *** – non-significant or significant at $P \leq 0.05$, 0.01 or 0.001, respectively. Standard deviations (±SD) of the mean values are also presented.

FD treatment increased titratable acids content (TAC) by up to 32%, while FD + H decreased it in all fruit orders in 2012 (Table 4). FD and FD + H decreased TAC in primary fruits in 2013. Mean effect of fruit order on TAC was statistically significant at $P \leq 0.05$ and treatments at $P \leq 0.001$. SSC/TAC was influenced due to FD in all fruit orders, while FD + H affected the ratio only in 2012 compared to FD. In 2013, the effect of FD + H was positive in primary and tertiary fruits, increasing the ratio up to 33% compared to C and FD. Mean effect of fruit order was significant for the SSC/TAC at $P \leq 0.01$ and treatments at $P \leq 0.001$.

FD and FD + H decreased the ascorbic acid content (AAC) by up to 11% in primary and 36% in tertiary fruits in 2012 (Table 4). The same effect was noticed in secondary fruits in 2013, while FD + H had positive effect on the tertiary fruits. There was no effect of any experimental treatments on AAC in primary strawberry fruits. Treatments and fruit order had significant mean effect on AAC at $P \leq 0.001$.

Total phenolics content (TPC) was significantly affected by treatments in all fruit orders (Table 4). FD + H increased TPC in primary and tertiary fruits but decreased it in secondary fruits in 2012. In 2013, TPC increased by up to 12% by FD in primary fruits but decreased significantly in tertiary fruits compared to FD + H. Mean effect of fruit order and treatments on TPC was significant at $P \leq 0.001$. ACC was significantly lower in FD for all fruit orders in 2012 (Table 4). Humic acid amendments increased ACC significantly in defoliated plants in all fruit orders in 2012. In the next experimental year, ACC was increased by FD in primary and by FD + H in tertiary fruits (from 10.5 to 15.4 mg 100 g^{-1} FW). The mean effect of fruit order and treatments on ACC was statistically significant at $P \leq 0.001$.

The correlation analysis showed moderate positive correlation between TAC and the number of leaves ($P \leq 0.05$) and inflorescences ($P \leq 0.05$); moderate negative correlation was found between TAC and SPAD ($P \leq 0.01$), and TAC and yield ($P \leq 0.01$) (Table 5). Moderate positive relationship was found between SSC/TAC and SPAD value ($P \leq 0.05$). TPC correlated moderately negatively with SPAD ($P \leq 0.01$) and yield ($P \leq 0.01$). Strong negative correlation was observed between ACC and number of leaves ($P \leq 0.01$), while ACC had positive moderate correlation with SPAD ($P \leq 0.05$) and with yield ($P \leq 0.01$). No correlation was found between the mass of roots and crowns and fruit biochemical parameters.

Table 5. Correlation coefficients (*r*) between the plant parameters and fruit biochemical parameters of strawberry cv. 'Darselect'

Strawberry plant parameters	Soluble solids, °Brix	Titratable acids g 100 g^{-1}	Soluble solids / titratable acids	Ascorbic acid mg 100 g^{-1}	Total phenolics mg 100 g^{-1}	Anthocyanins mg 100 g^{-1}
Leaves No. $plant^{-1}$	0.232 ns	0.564 *	−0.404 ns	−0.129 ns	0.310 ns	−0.729 **
Inflorescences No. $plant^{-1}$	0.379 ns	0.511 *	−0.244 ns	−0.001 ns	0.260 ns	−0.290 ns
SPAD values	−0.182 ns	−0.646 **	0.515 *	−0.065 ns	−0.592 **	0.566 *
Yield g $plant^{-1}$	−0.26 ns	−0.641 **	0.442 ns	0.259 ns	−0.561 *	0.671 **
Crown and root mass g $plant^{-1}$	0.277 ns	0.195 ns	0.054 ns	0.336 ns	−0.076 ns	−0.054 ns

ns – correlation coefficients between variables with the significance coefficients being non-significant; significance at $P \leq 0.05$* and $P \leq 0.01$**

Discussion

Strawberry growth and yield. FD treatment decreased the number of leaves in both experimental years and the mass of crowns and roots eventually. Photosynthesis processes after FD treatment were inhibited up to one week as all the leaves were removed. Inhibited processes caused the delay in the growth of new leaves, and due to the growth delay, the competition between foliage development and flower bud initiation occurred. Thompson and Guttridge (1960) have pointed out that strawberry leaves of different maturity are able to reduce flower bud initiation and under some conditions mature leaves can act as inhibitors compared to immature leaves. In the present experiment, the negative tendency in 2013 may be explained not only by repetitive FD treatment but also by the ageing of strawberry plants and their sympodial growth; crowns had arisen above the soil surface, being more influenced by different treatments, especially by flame-defoliation.

Application of humic substances had positive effect on the leaf growth of defoliated plants only in one experimental year out of two. The effect of FD + H on the number of inflorescences became evident only in the next year after the second treatment. Regarding other plant characteristics, the effect of FD + H tended to be decreasing or showing no effect at all. According to literature, humic and fulvic acids can interact with soil nutrients and elicit physiological responses in plants leading to increased plant growth (Calvo et al., 2014). However, in the present experiment additional amendments of humic substances could have contributed to the plant recovery by improving plant nutrient uptake and therefore favoured plant growth. Still, the impact of applying humic substances to the flame-defoliated plants differed yearly as affecting the plant characteristics probably in relation to changed plant physiological processes and soil nutrient level. It has been indicated that strawberry flower bud induction is sensitive to different agronomic and nutritional factors (Savini et al., 2005). Enhanced nutrient uptake probably led to increased nitrogen assimilation which promoted plant growth and inhibited the flower bud initiation. Although additional fertilization can increase flower bud induction successfully only from a low soil nutrient base

(Breen, Martin, 1981), additional fertilization of already fertile soil can inhibit flower bud formation and reduce fruit yield. In the present experiment, high contents of soil nutrients, and additional humic acids could have had negative influence on strawberry plant growth parameters and yielding. Moreover, also the decreased number of inflorescences by FD + H treatment probably affected the strawberry yield.

In the present experiment, the strawberry yields under organic production conditions were in the case of FD + H and FD treatment respectively, in the first year from 444 to 567 g plant^{-1}, but in the second experimental year only from 189 to 215 g plant^{-1}. In conventional strawberry experimental plantations, the average yields obtained using additional fertilization were from 336 to 427 g plant^{-1} (Moor et al., 2009), which agrees with our results. According to Estonian economic analysis, an average strawberry yield, which would be still profitable, is approximately 4400 kg ha^{-1} (Vahejõe et al., 2010). In the present experiment, the average strawberry yield in the first year would have been from 6660 to 8580 kg ha^{-1}, which is higher than the Estonian average, but in the next year from 2835 to 3225 kg ha^{-1} being remarkably lower. The latter indicates that the repeated usage of strawberry flame-defoliation may not be agronomically profitable.

Strawberry fruit biochemical composition. FD had significant influence on strawberry fruit biochemical composition, but the treatment-caused variations in fruit composition differed yearly. This could have been caused by differences in weather conditions, but also by the age of the strawberry plants. Earlier findings showed that the effect of growing methods on fruit quality parameters depended significantly on environmental conditions (Moor et al., 2004; Crespo et al., 2010). In the present experiment, the temperatures in June 2013 were warmer by up to 3.1ºC than in previous years and long-term mean. Moreover, rainfall was higher in May compared to many years mean, but significantly lower in June and July during the flowering and fruiting. According to literature, plants need to adapt to changing environmental conditions for survival in increasing pressure due to biotic and abiotic stressors (Van den Ende, El-Esawe, 2014). Therefore, in addition to the effect of FD as a thermal treatment, also air temperature fluctuations and variable precipitation rates in the present study could have caused differences in plant response and hence influenced the effect of defoliation.

The interaction between flame-defoliation and humic acids application became evident. Combined treatment of FD + H increased the SSC by up to 50% in the last experimental year compared to plants that were defoliated only. Dadashpour and Jouki (2012) concluded the availability of plant nutrients as a significant factor for influencing SSC and SSC/TAC, especially in the case of higher absorption of nitrogen. Moor et al. (2004) have found that fertilization of defoliated plants had no effect on the content of soluble solids and variations in the results depend on the cultivar and plant age. Humic substances had positive influence on the TPC and ACC. The effect may be a result of the additional treatment with humic substances that advanced the defoliated plants' recovery by increasing the mass of strawberry crowns and roots. This, in turn, could have promoted nutrient uptake and therefore, increased the TPC and ACC in both experimental years too. Our findings correspond with those of Wang and Lin (2003) who reported the significant influence of improved plant nutrient uptake on the accumulation of phenolic compounds in strawberries. In addition to previous, interactions between treatments may have occurred due to plant age, as was also found by Tõnutare et al. (2009) – the content of anthocyanins showed the tendency of increasing in three-year-old plantation.

Strawberry fruit biochemical composition was found to be fruit order dependent. Anttonen et al. (2006) have also found up to 2-fold differences in accumulation of phenolic compounds and anthocyanins between fruit orders which have been related to fruit size. In the current study, in the last year of fruiting, the TPC was increased significantly in primary fruit order, but decreased in tertiary fruits due to FD which can be also related to fruit mass and fruit maturation conditions. In 2012, primary fruits were picked three weeks earlier than tertiary fruits, and in 2013 the interval was two weeks. During fruiting in 2012, the temperatures between June and July did not differ significantly, but precipitation was 38 mm higher in July. In 2013, the temperatures in June were up to 4ºC lower compared to July, which shows that tertiary fruits had warmer conditions for maturation than for example in year 2012, while the amounts of rainfall were similar in both experimental years.

The correlation analysis showed that the influence of defoliation on strawberry biochemical characteristics was related to plant growth parameters, and TAC, TPC and ACC were most affected biochemical compounds. TAC had positive correlation with the number of leaves which shows that higher number of leaves led to the increased titratable acids. This is in an agreement with Correia et al. (2011), who reported that TAC was positively related to the fresh weight of above-ground biomass and number of leaves in some strawberry cultivars. On the other hand, SSC/TAC showed positive correlation with SPAD, consequently higher chlorophyll content in leaves affected fruit taste, enhancing strawberry sweetness. Increased nitrogen application rate has been reported to increase the sugar content of strawberries which has also been correlated with increased leaf nitrogen content (Hargreaves et al., 2008). The TPC was found to be related to yield and leaf chlorophyll content (SPAD), but whether it is due to the increased plant nutrient availability and therefore increased nitrogen content in the leaves, it needs further investigations. In the present experiment, differences in leaf growth did not affect TPC but greater number of leaves decreased the ACC significantly. Anttonen et al. (2006) have found that vigorous growth and the shading effect of the leaves can be named as the main factors diminishing ACC. Therefore it can be assumed that in the present experiment, the higher number of leaves affected the ACC negatively, while the higher SPAD had positive effect.

Conclusions

1. Flame-defoliation decreased strawberry plant leaf number and fruit mass. Soluble solids content was increased in primary fruits in the first experimental year, and anthocyanins were reduced in all fruit orders in the succeeding year after the treatment. Total phenolics content was increased in primary, but decreased in tertiary fruits in the second experimental year.

2. The application of humic substances to the defoliated plants increased the number of leaves in the first year, and increased the crown and root mass eventually. The yield was decreased in both years. Soluble solids were decreased in the first experimental year, but increased in the next year. The content of total phenolics and anthocyanins was affected positively in both years.

3. From the previous it can be concluded that post-harvest defoliation, and defoliation in combination with humic substances affected strawberry plant growth parameters and fruit biochemical composition too, but the direction of the influence differed yearly and was also fruit order dependent. On the basis of the yield results obtained, the post-harvest flame-defoliation could be recommended for use in organic strawberry plantations for single treatment in the second growth year. Further experiments may be required to investigate the influence of flame-defoliation on different strawberry cultivars and fruit quality parameters.

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Braškių defoliacijos liepsna po derliaus nuėmimo įtaka augalų augimui ir vaisių biocheminei sudėčiai

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Santrauka

Bandymas su daržinės braškės (*Fragaria* × *ananassa* Duch.) veisle ‘Darselect’ buvo atliktas Estijos gyvybės mokslų universiteto Ekologinio ūkininkavimo tyrimų centre (58º21′ N, 26º40′ E). Tyrimo tikslas – nustatyti braškių defoliacijos tiesioginės propano liepsnos įrenginiu ir liepsnos bei papildomo huminių medžiagų derinio įtaką augalo augimui ir vaisių biocheminei sudėčiai dvejus metus iš eilės. Defoliacija sumažino lapų bei žiedynų skaičių ir vainikų bei šaknų masę. Dvimečių augalų defoliacija liepsna po braškių derliaus nuėmimo padidino vaisių derlių (2012), tačiau vėlesnė defoliacija sumažino kitų (2013) metų derlių. Defoliacijos poveikis vaisių biocheminei sudėčiai buvo esminis, tačiau skyrėsi tarp metų ir vaisių skynimo eiliškumo. Defoliacija padidino tirpių sausųjų medžiagų kiekį pirmojo skynimo vaisiuose nuo 8,9 iki 9,3 °Brix, bet sumažino antocianinų kiekį visų skynimų vaisiuose 2012 m. Apdorojimas liepsna fenolinių junginių kiekį padidino nuo 181 iki 206 mg 100 g^{-1} pirmojo skynimo vaisiuose, tačiau sumažino jų kiekį nuo 268 iki 254 mg 100 g^{-1} trečiojo skynimo vaisiuose 2013 m. Huminių medžiagų naudojimas defoliuotiems augalams padidino lapų skaičių 2012 m. ir vainikų bei šaknų masę 2013 m., tačiau abiem metais sumažino derlių. Tirpių sausųjų medžiagų kiekis skyrėsi kiekvienais metais – 2012 m. jų kiekis sumažėjo, tačiau 2013 m. padidėjo. Abiem metais huminės medžiagos turėjo teigiamos įtakos bendram fenolinių junginių ir antocianinų kiekiui.

Reikšminiai žodžiai: antocianinai, askorbo rūgštis, bendras fenolių kiekis, huminė rūgštis, tirpios kietosios dalelės, titruojamos rūgštys.