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ORIGINAL PAPER

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EXOGENUS 'GA_3' AND 'GA_{4+7}' EFFECTS ON PHENOLOGICAL INDICES, FROST HARDINESS AND QUALITY PROPERTIES OF 'ENGLISH MORELLO' SOUR CHERRY (*Prunus cerasus* L.)

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ABSTRACT

Exogenous gibberellins GA_3 and GA_{4+7} treatments on 'English Morello' sour cherry trees 58 days after anthesis were evaluated in relation to their effect on fruit firmness, size distribution, colour development, titratable acidity to firmness ratio, yield, efficiency indices as well as leaf fall senescence. Moreover in the season following the applications their effect on flower bud characteristics including frost hardiness, fruit set and fruit drop, were examined. Foliar GA_3 treatment had the most positive effect on 'English Morello' fruit size in the year of application and in the following season. GA_{4+7} treatment in the first experimental year increased sour cherry fruit firmness. Both GA_3 and GA_{4+7} delayed leaf senescence process in the autumn. GA_3 application had a positive effect on flower buds survival in comparison to control trees after frost occurred in spring. No GAs subsequent effect on next year fruit set and yield compared to control was found.

Key words: gibberellins, fruit size, firmness, fruit colour, yield

INTRODUCTION

Physiological properties of endogenous and exogenous–gibberellins (GA) in deciduous fruit plants have been most extensively analyzed in relation to bioactive GA_1, GA_3, GA_4, GA_7, as well as GA_9, GA_{12}, GA_{20}, GA_{32} and GA_{34} [Bukovac et al. 1979, Looney et al. 1985, Stephan et al. 1999, Zhao et al. 2010, Csukasi et al. 2011, Yang et al. 2013, Zhang and Whiting 2013]. Based on their functions, in different stages of plant development, applications of exogenous GAs provided technological progress in many agricultural production areas and became a standard procedure for numerous crops. GAs based products, has been applied in vegetable [Knoche and Peschel 2007], floriculture [Cardoso et al. 2012], nursery [Palmer et al. 2011, Pio et al. 2012] and fruit industry. In fruit production GAs application is a tool used for a range of functions: to stimulate vegetative growth [Qureshi et al. 2013], inhibit flower bud formation [Val et al. 2001, Gonzalez-Rossia et al. 2006, Lenahan and Whiting 2008], affect fruit set [Shahin et al. 2010, Galván et al. 2012], reduce fruit surface russeting [Knoche et al. 2011, Curry 2012], induce

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parthenocarpic fruit [Bangerth and Schroeder 1994, Mesejo et al. 2010], dalay fruit maturity [Clayton et al. 2006, Cline and Trought 2007], stimulate rachis elongation for looser fruit clusters [Hed et al. 2011], enhance fruit resistance to pest infestation [Birke et al. 2011] as well as to improve size and internal quality properties of fruit [Basak et al. 1998, Chang and Lin 2006, Zhang and Whiting 2011a, b, Einhorn et al. 2013].

Stone fruit development after pollination consists of three stages: cell division (stage I), pit hardening / embryo development (stage II) and cell enlargement (stage III). Increasing GAs content, in particular gibberellic acid, in a plant at stage II and stage III can modify fruit development delaying its maturity processes [Demirsoy and Bilgener 2000, Pegoraro et al. 2010]. Later fruit ripening is associated with GA effect on cell wall hydrolytic enzymes activity [Kondo and Danjo 2001, Choi et al. 2004] resulting in delayed flesh firmness loss [Clayton et al. 2006] as well as with inhibition of anthocyanin production leading to retarded color change of exocarp [Usenik et al. 2005, Cline and Trought 2007, Zilkah et al. 2008, Zhang and Whiting 2011a, b].

Sour cherry (*Prunus cerasus* L.), unlikely from other crops, is facing strong domination of limited number of genotypes being cultivated on a commercial scale. The few cultivars, of the highest share in global production structure worldwide are: ‘English Morello’ in Poland and Germany [Jadczuk-Tobjasz and Bednarski 2007, Lampe and Hilsendegen 2006], ‘Montmorency’ in United States [Ou et al. 2012], ‘Oblacinska’ in Serbia [Miletić et al. 2009, Fotirić Akšić et al. 2013], ‘Újfehértói fürtös’, ‘Érdi bőtermő’ and ‘Kántorjánosi 3’ in Hungary [Stéger-Máté et al. 2010]. Sour cherry as a non-climacteric fruit has a short period of post-harvest life and a narrow optimum harvest time window. A high share of a single cultivar in a commercial production means that local fresh fruit supply for processing and freezing industry is limited to a very short period. Under these circumstances other than genetic methods of fruit ripening process regulation either by fastening or delaying maturity can play an important role in production strategies. Affecting sour cherry harvest time resulting in sooner and more uniform fruit maturity may be achieved by pre-harvest applications of etephon (2-chloroethyl phosphonic acid) and abscisic acid which stimulate ethylene biosynthesis [Ren et al. 2011, Soczek et al. 1983]. Foliar application of etephon based products is considered a standard procedure in sour cherry production prior to mechanical fruit harvest [Khorshidi and Davarynejad 2010, Smith and Whiting 2010]. It stimulates creation of abscission layer at the juncture between pedicle and fruit reducing the retention force (FRF) minimizing detachment damage.

Research on stone fruit species reaction to exogenous gibberellins focus on: peach (*Prunus persica* (L.) Batsch), plum (*Prunus domestica* L.), apricot (*Prunus armeniaca* L.) and sweet cherry (*Prunus avium* L.). Gibberellic acid (GA_3) application at the straw-yellow fruit skin color stage of development is reported to be a standard practice for sweet cherry production in British Columbia (Canada), the Pacific Northwest of the United States and other sweet cherry producing areas [Kappel and MacDonald 2007]. There are few studies evaluating GAs effect in sour cherry physiology [Buban 1996]. Some of them are limited to evaluation of GAs effect on flower bud formation, fruit set and yield not considering their effect on fruit internal and external properties [Bukovac et al. 1986, Bukovac and Yuda 1991]. The following study concentrates on examining exogenous GA_3 and GA_{4+7} foliar applications effect on dynamics of *Prunus cerasus* ‘English Morello’ (syn. ‘Lutowka’, ‘Schattenmorelle’) fruit quality parameters expression during two seasons – the year of application and a follow-up effect in a subsequent season.

MATERIAL AND METHODS

The research was conducted at Poznan University of Life Sciences, Research Station in western Poland (52°10'N, 18°51'E). *Prunus cerasus* ‘English Morello’ trees grafted on *Prunus mahaleb* (L.) rootstock were planted in spring 2002 in north to south rows in spacing of 3.5 × 1.5 m on proper grey-brown podsolic soil overlaying light boulder clay. The trees were trained to a central leader architecture. Standard management practices (pest, disease and weed control) were followed every season with annual nitrogen soil fertilization dose of 50 $kg \cdot ha^{-1}$ and no irrigation applied. In the 10th and 11th year of experimental trees' growth, evaluation of foliar application of exogenous gibberellins on trees development and crop properties was executed. On the 25-th of June 2011 experimental trees were sprayed with water solution of gibberellic acid isomers: 'GA_3' ('Gibb 3', Globachem nv) and 'GA_{4+7}' ('Gibb Plus', Globachem nv) by air-blast sprayer. The sprays of the GAs were applied to whole canopies at doses of 30 ppm of GA_3 (treatment marked as GA_3) and 30 ppm of GA_{4+7} (treatment marked as GA_{4+7}). The solution amount used equaled 660 l/ha. Applications were performed 58 days after anthesis, at air temperature of 19.0°C. The treated trees as well as untreated control trees were arranged in a randomized complete-block design replicated four times with four trees per replication and from 5 to 8 trees of isolation between blocks. On the of 13-th of July 2011 fruit samples of 85 stemless fruit per tree were collected from randomly selected one year old shoots situated from 150 to 200 cm above ground in outer western canopy zone, the mean weight of every sample was measured with accuracy of 0.01 g directly after which they were subjected to fruit quality analyses. Fruit diameter and mean firmness of individual fruit were measured by Fimtech 2 (Bioworks, USA) with a force thresholds set at 50–350 gms, on a sample of 400 fruits per treatment (100 fruits per block, 25 per tree). The total content of soluble solids (TSS) was measured in juice obtained from randomly selected 10 fruits per tree (160 fruits per treatment) by refractometer PR-101a (Atago Co. Ltd., Japan). The same juice samples (16 per treatment) were used to evaluate titratable acidity (TA) and TSS/TA ratio. Titratable acidity was measured based on the titration of organic acids in juice solution with NaOH (0.1 N) to pH 8.2 by a pH meter (pH 538, WTW, Germany) calibrated with pH 4 and 7 buffers and was expressed as % of malic acid. On the 19-th of July the second collection of fruit samples and analyses of fruit quality attributes were performed according to the same procedures as described above. Yield was determined by weighing all fruit harvested without stems separately from each experimental tree on the 19-th of July increased by the weight of previously collected samples. Efficiency index was calculated as a ratio of individual yield and tree trunk cross sectional area (TCSA) measured on the 28-th of October 2011. Trees vigour was assessed based on TCSA increment during vegetation season as well as on measurements of number and length of all one year old shoots performed on the 28-th of October. At the beginning of leaf drop phase, at BBCH 93 stage, samples of 200 randomly selected leaves were collected from one-year old shoots of each experimental block and their phenological indices reflecting development stage: mean leaf blade weight as well as leaf color were evaluated. Color measurements were done by a colorimeter (Minolta CR-200, Japan) on all collected leaves, with one reading per leaf, and expressed by CIE parameters L* (lightness), a* (redness and greenness) and b* (yellowness and blueness) on a three-dimensional colour space. In 2012 subsequent effect of the 2011 GAs application was examined. In March 2012 a representative three-year old branch at each experimental tree at the height of 150–200 cm was selected and labelled. At BBCH 55 stage all flower and leaf buds were counted within selected branches and at BBCH 59 (hollow ball stage) all flowers were counted to determine average number of flower meristems per flower bud. Following spring frosts with minimum temperature of –4.53°C which occurred on the 9-th of April 2012 (fig. 1), flower buds survival was evaluated at stage BBCH 65 based on determining the share of healthy and frost damaged flower buds on the selected branches. The percent of fruit set vs. total flowers number and percentage of fruit drop were determined, by comparing flower and fruitlet number on selected branches on each tree at stages BBCH 65, BBCH 73 and BBCH 76. Fruit harvest and fruit quality measurements in 2012 were performed on the 20-th of July. Fruit size, fruit firmness, total soluble solids, titratable acidity, as well as yield, efficiency index and TCSA increment in 2012 were analyzed following the procedures of sampling and methods

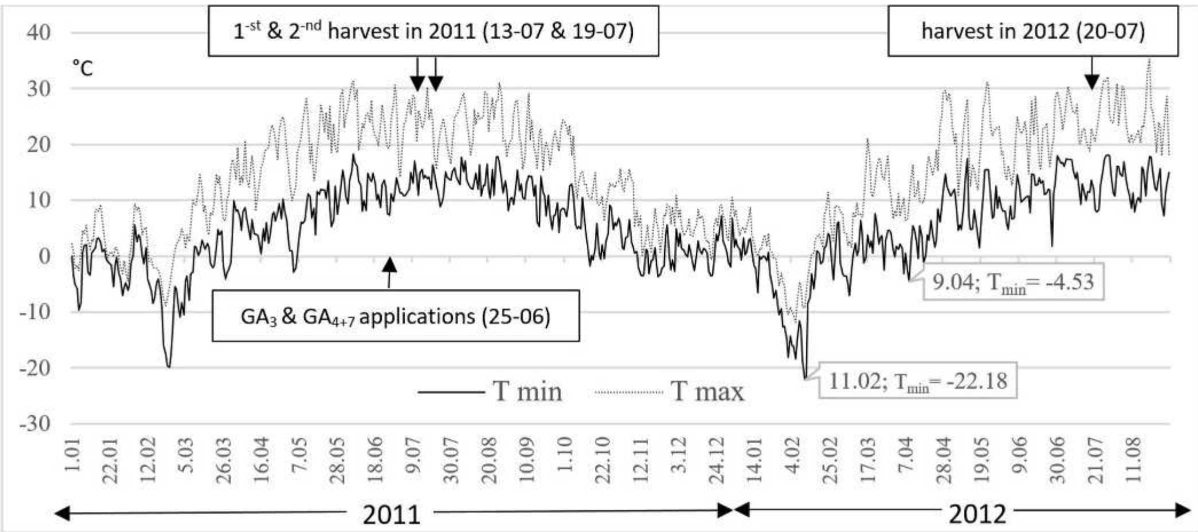


Fig. 1. Minimum and maximum daily air temperatures from January 2011 to August 2012 (2 m above ground level); GA_3 and GA_{4+7} applications and fruit harvest dates

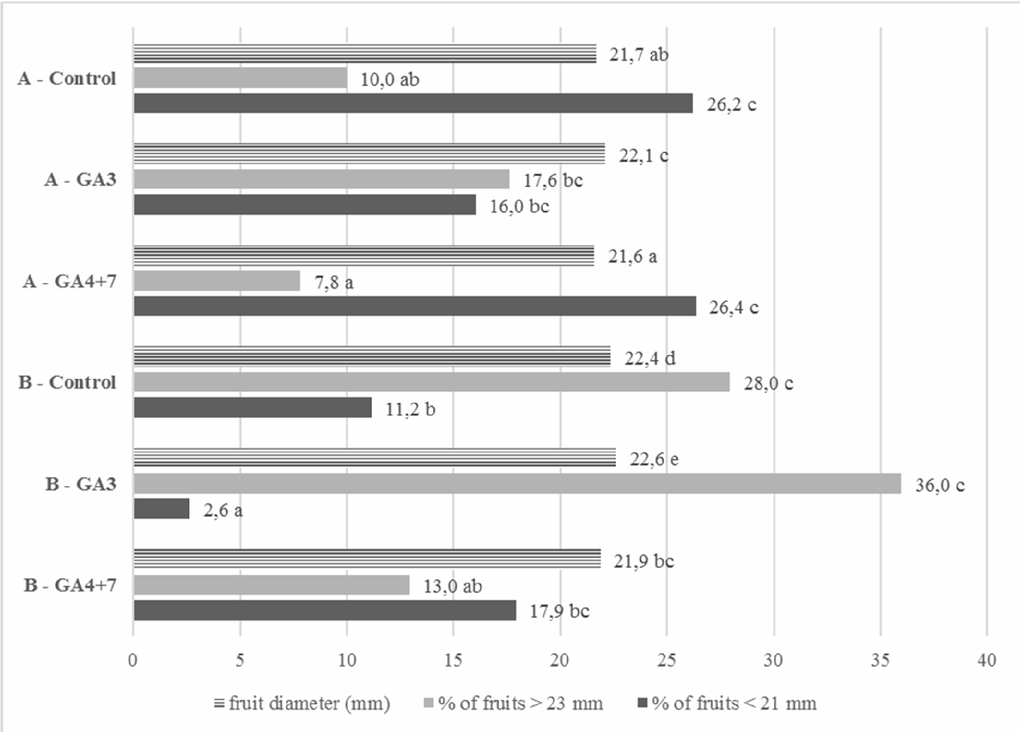
used in 2011 described above. GAs and Control combinations in one-way analyses of variance and treatments and year in two-way analyses of variance were separated by Duncan's Multiple Range test at $P \leq 0.05$. Percentage values were transformed into Bliss grades.

RESULTS AND DISCUSSION

GA_3 increased fruit size essentially in 2011 in relation to Control treatment: from a diameter of 21.7 mm to 22.1 mm during the first harvest and from 22.4 mm to 22.6 mm at the second harvest. In particular on the 13-th of July, the share of fruit with diameter over 23 mm on GA_3 treated trees was 76.1% higher and the share of fruit with diameter below 21 mm was 38.9% lower on GA_3 treated trees compared to control ones. GA_3 application resulted also in significantly lower percentage of fruits with diameter below 21 mm than in control treatment at the second harvest. The effect of GA_{4+7} application on ‘English Morello’ fruit size in 2011 was not significant at the first harvest and negative at the second harvest (fig. 2).

Positive role of exogenous GA_3 application on fruit size in 2011 repeated also in the season following the application. In the second year fruits from the trees treated with both GA_3 and GA_{4+7} had essentially higher mean diameter compared to control ones and had the share of smallest fruits (below 21 mm of diameter) nearly twice lower (fig. 3).

Some reports point at the positive effect of GAs applications at end of stage II – beginning of stage III on stone fruit firmness due to their effect on cell wall hydrolytic enzymes activity [Kondo and Danjo 2001, Choi et al. 2004]. ‘English Morello’ is reported to be a sour cherry cultivar with a low firmness level, poor fresh storage properties when harvested stemless and large fruit size. Generally there is a tendency of slightly lower firmness of the largest stem lessly harvested fruit (fig. 4 – Control). The results showed that despite the fruits from GA_3 treated trees had higher fruit diameter in the year of application, they did not differ significantly in firmness. It means the GA related fruit size rise was not accompanied by firmness drop. The correlation coefficient between fruit diameter and firmness equaled –0.37 and –0.40 in 2011 first and second fruit harvest respectively,



Factor first – treatment, factor second – year

* Two-way analyses of variance; data marked with the same letter within the given feature are not significantly different at α = 0.05 (Duncan's test)

Fig. 2. GA$_3$ and GA$_{4+7}$ effect on cv. English Morello sour cherry fruit diameter on 13.07.2011 (A) and 19.07.2011 (B)

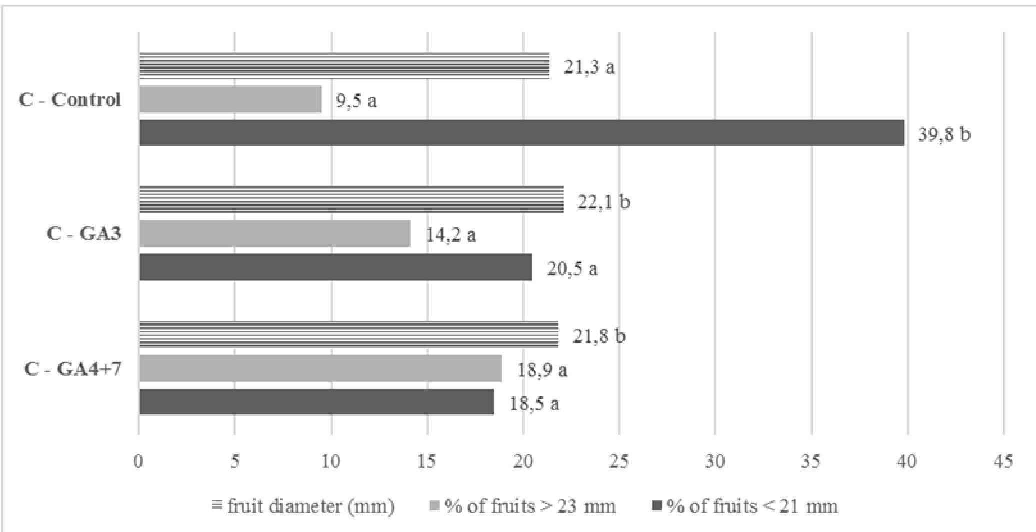


Fig. 3. GA$_3$ and GA$_{4+7}$ effect on cv. English Morello sour cherry fruit diameter on 20.07.2012

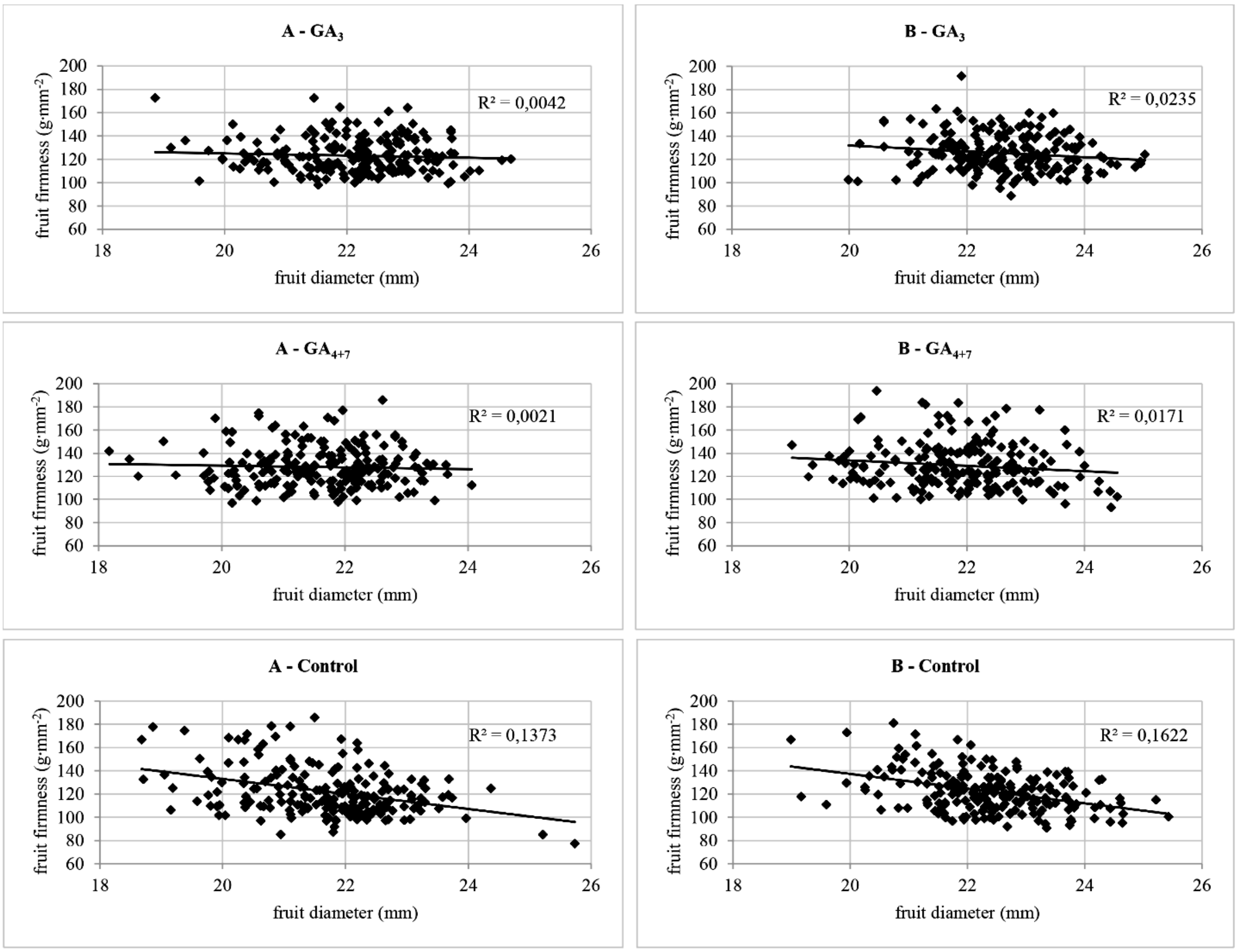


Fig. 4. GA$_3$ and GA$_{4+7}$ effect on cv. English Morello sour cherry fruit diameter and firmness relations on 13.07.2011 (A) and 19.07.2011 (B)

whereas in GAs treatments it was between –0.04 and –0.15. No significant differences in mean fruit firmness between GA$_3$ treatment and Control in 2011 were found, whereas GA$_{4+7}$ applications resulted in higher firmness in both first and second harvest (fig. 4, tab. 1).

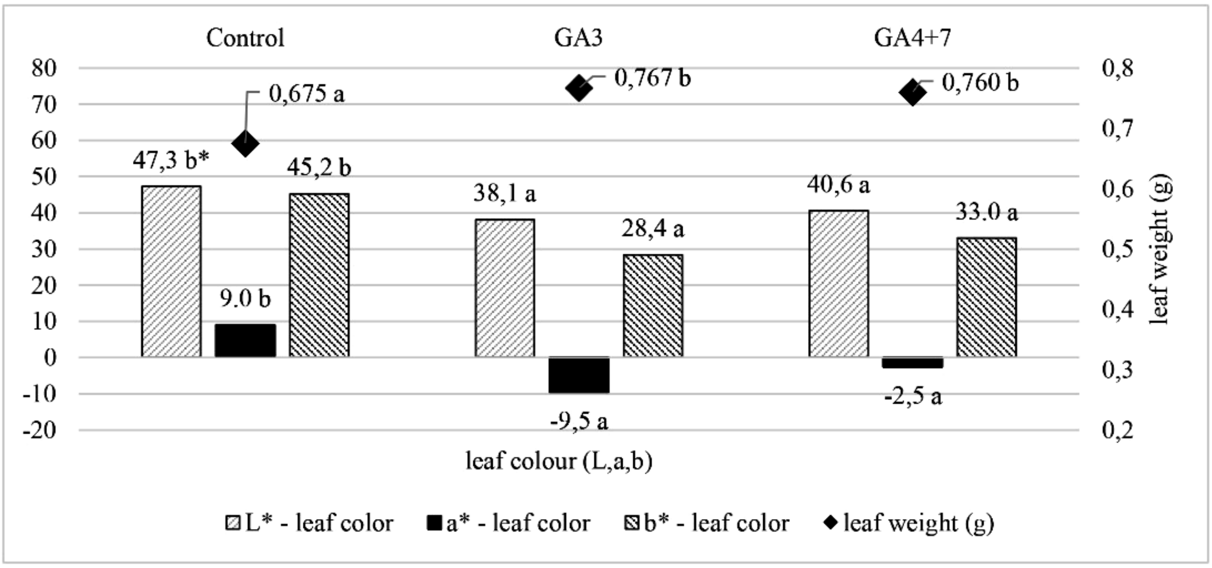
Fruit quality attributes analyzed: total soluble solids and titratable acidity did not appear to depend on the treatment in the study. Similarly no significant differences in yield, efficiency indices vs. trunk cross sectional area and vs. one-year old shoot cumulative length were found, with exception of higher yield on GA$_{4+7}$ vs. GA$_3$ treated trees in 2011 (tabs 1, 2 and 3). The average yield levels in the application year varied from 10.6 $t \cdot ha^{-1}$ to 13.7 $t \cdot ha^{-1}$ and in subsequent season from 11.0 $t \cdot ha^{-1}$ to 12.1 $t \cdot ha^{-1}$ The crop level should be classified as average considering highly intensive sour cherry orchard.

In 2012 frost damages of generative buds were recorded as a result of spring frost event which occurred on the 9-th of April 2012 (fig. 1). The flower buds of experimental trees were affected and the share of frost injured buds on GA$_3$ treated trees was significantly lower than in control treatment. Better

Table 1. GA_3 and GA_{4+7} effect on cv. English Morello sour cherry fruit quality in 2011

		Control	GA_3	GA_{4+7}	Mean values
Fruit diameter (mm)	I – 13.07.2011	21.69 ab*	22.10 c	21.60 a	21.79 a
	II – 19.07.2011	22.37 d	22.62 e	21.89 bc	22.29 b
	mean values	22.03 b	22.36 c	21.74 a	-
Firmness ($g \cdot mm^{-2}$)	I – 13.07.2011	122.08 a	123.11 a	128.23 bc	124.48 a
	II – 19.07.2011	122.32 a	125.38 ab	129.55 c	125.75 a
	mean values	122.20 a	124.25 a	128.89 b	–
Total soluble solids (%)	I – 13.07.2011	14.9 a	15.1 a	15.1 a	15.0 a
	II – 19.07.2011	15.4 a	15.5 a	15.1 a	15.3 a
	mean values	15.1 a	15.3 a	15.1 a	–
Titratable acidity (% malic acid)	I – 13-07-2011	2.3 a	2.2 a	2.1 a	2.2 a
	II – 19.07.2011	2.1 a	2.2 a	2.2 a	2.2 a
	mean values	2.2 a	2.2 a	2.1 a	–
Total soluble solids / titratable acidity	I – 13.07.2011	6.6 a	6.8 a	7.0 a	6.8 a
	II – 19.07.2011	7.2 a	7.0 a	7.0 a	7.1 a
	mean values	6.9 a	6.9 a	7.0 a	–

* Two-way analyses of variance; data marked with the same letter within the given feature are not significantly different at α=0.05 (Duncan’s test)



* One-way analyses of variance; data marked with the same letter within the given feature are not significantly different at α=0,05 (Duncan’s test)

Fig. 5. GA_3 and GA_{4+7} effect on cv. English Morello sour cherry leaf color and leaf blade weight at BBCH 92 stage (28.10.2011)

Table 2. GA_3 and GA_{4+7} effect on English Morello sour cherry fruit quality in 2012

		Control	GA_3	GA_{4+7}
Fruit diameter (mm)	III – 20.07.2012	21.35 a*	22.11 b	21.84 b
Firmness ($g \cdot mm^{-2}$)	III – 20.07.2012	112.23 a	112.17 a	115.73 a
Total soluble solids (%)	III – 20.07.2012	11.8 a	13.6 a	12.8 a
Titratable acidity (% malic acid)	III – 20.07.2012	1.4 a	1.5 a	1.5 a
Total soluble solids / titratable acidity	III – 20.07.2012	8.2 a	9.3 a	8.6 a

* One-way analyses of variance; data marked with the same letter within the given feature are not significantly different at $\alpha = 0.05$ (Duncan's test)

Table 3. GA_3 and GA_{4+7} effect on generative and vegetative characteristics of cv. English Morello sour cherry trees in 2011 and 2012

Generative and vegetative characteristics	Control		GA_3		GA_{4+7}	
			2011			
Yield (kg per tree)	9.28	ab[3]	7.92	a	10.26	b
Efficiency index vs. TCSA[1] ($kg \cdot cm^{-2}$)	0.13	a	0.11	a	0.12	a
Efficiency index vs. 1-YOSL[2] (kgm)	0.25	a	0.26	a	0.23	a
1-YOSL (cm)	131.0	ab	110.3	a	147.3	b
Length (number) of annual shoots (per tree)	28.2	a	28.0	a	29.7	a
			2012			
No of floral meristems per flower bud	3.26	a	3.54	a	3.48	a
% of frost damaged flower buds	12.6	b	8.7	a	9.8	ab
Fruit set (%)	34.5	a	29.8	a	28.6	a
Fruit drop (%)	15.7	a	13.4	a	15.0	a
% of harvested fruit vs. flowers no.	29.0	a	26.7	a	27.9	a
Yield (kg per tree)	8.55	a	8.25	a	9.08	a
Efficiency index vs. TCSA[1] ($kg \cdot cm^{-2}$)	0.12	a	0.12	a	0.12	a
TCSA increment – 2011–2012 (cm^2)	2.18	a	3.00	b	3.38	b

[1] Trunk cross-sectional area
[2] Length of annual shoots
[3] One-way analyses of variance; data marked with the same letter within the given feature are not significantly different at $\alpha=0.05$ (Duncan's test)

survival of flower buds on GA_3 treated trees was however not reflected in the fruit set. Disregarding the treatment the fruit set in relation to total flower number did not differ in 2012. Also the percentage of harvested fruit vs. total flowers number was quite uniform between 26.7 and 29.0% and did not depend on the treatment (tab. 3). Lenahan et al. [2006] point at thinning effect of exogenous GA_3 and GA_{4+7} which may result in changes in the subsequent year flower bud formation e.g. flower vs. flower bud ratio. The analyses of GAs treatments follow-up effect on number of floral meristems per flower bud, expressing yield potential and density, did not present significant differentiation.

Trees vigour evaluations based on two year trunk cross sectional area increment suggest stronger growth of GA_3 and GA_{4+7} treated trees in comparison to control. Also Zhang and Whiting [2011a, b] noticed that GA_3 and GA_{4+7} application had more promotive effect on current shoot vigour of sweet cherry trees when executed 30 days after anthesis than when performed 37 days after. In this trial the measurements of one year shoots length and number which were done in dormant season (with no leaves on) i.e. after post-harvest standard pruning every year did not show the differences in both parameters (tab. 3). Moreover limited GAs applications effect on shoot growth in the study might have been the effect of their application time 58 days after full bloom – the time which took into consideration the phenological development pattern of sour cherry.

Although GAs treatments did not modify fruit maturity indices like TSS and TA, they had essential effect on tree phenology in the fall. Both GA_{4+7} and GA_3 treatments delayed autumn leaf drop. On the 28-th of October 2011, leaves on GAs treated trees were significantly less advanced in color change from green to yellow. Similarly the measurements of leaf blade weight at that stage revealed a modification in leaf senescence process as result of June GAs foliar applications (fig. 5).

CONCLUSIONS

Exogenous GA_3 treatment on ‘English Morello’ sour cherry trees 58 days after anthesis had a positive effect on fruit size in the application season and in the following year.

In the year of GAs application GA_{4+7} resulted in increased firmness of ‘English Morello’ sour cherry fruit. No significant effect of GA_3 and GA_{4+7} on firmness was observed in second year of experiment. Fruit size improvement in GA_3 treatment was not accompanied by as intensive fruit firmness drop as in control treatment.

Both GA_3 and GA_{4+7} application delayed leaf senescence process in the year of applications and increased mean leaf blade weight compared to control.

GA_3 application had a positive effect on flower buds survival in comparison with control trees after spring frost recorded in 2012.

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