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Integrated control of brown rot blossom blight by combining approved chemical control options with *Aureobasidium pullulans* in organic cherry production

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ABSTRACT

In a 3-year study, the effectiveness of copper hydroxide, lime sulphur, and their combinations with *Aureobasidium pullulans* were investigated for blossom blight control in organic sour cherry production. The effect of treatments on phytotoxicity on spur-leaf clusters and yield was also determined. A major aim was to evaluate the integrated control approach of the combined A. *pullulans* treatments to reduce allowed chemical damage on spur-leaf clusters. Among fungicide treatments suitable for organic production, copper hydroxide and lime sulphur alone were most effective for blossom blight control when applied three times (at closed blossom, full bloom, and petal fall) during bloom. Both treatments were not as effective as the conventional standard, caused more damage on spur-leaf clusters, but significantly increased crop yield compared to the untreated control. *A. pullulans* treatments, applied three times during bloom (at closed blossom, full bloom, and petal fall), resulted in significantly higher blossom blight incidence but lower phytotoxicity compared to copper hydroxide or lime sulphur treatments applied alone. All *A. pullulans* treatments controlled blossom blight significantly better in all years and increased crop yield in two out of three years compared to the untreated plots. The need for an integrated approach to improve blossom blight management in organic sour cherry production is discussed. This is the first in-depth study on the multiyear effects of *A. pullulans* applications on blossom blight control in comparison with organically approved plant protection compounds of copper or lime sulphur for organic sour cherry production.

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1. Introduction

Brown rot blossom blight, caused by *Monilinia laxa* (Aderhold & Ruhland) Honey is a devastating disease of sour cherry (*Prunus vulgaris* Mill.). The disease is endemic in Europe (Wormald, 1954; Tamm, 1994; Holb and Schnabel, 2005; Everhart et al., 2011), and is distributed in all major areas of fruit production with a humid and moderate warm climate (Byrde and Willetts, 1977). In rainy springs, *M. laxa* causes severe blossom blight (from 30 to 70%) in sour cherry orchards in Hungary (Holb, 2006; Holb and Schnabel, 2005; Holb et al., 2008). Depending on weather conditions, blossom blight can be controlled with one to three applications of protectant or/and systemic fungicides during the bloom period in conventionally and IFP-grown stone fruit orchards (Ogawa et al., 1985; Osorio et al., 1994; Holb, 2004; Holb and Schnabel, 2005).

Concerns about pesticide residues have generated increased interest in environmentally friendly plant protection, and especially, in organically grown fruits (e.g. Reganold et al., 2001; Holb et al., 2003; Tamm et al., 2004; Holb, 2006). However, brown rot under favourable conditions is difficult to control, and especially, in organic production systems (e.g. Larena et al., 2005; Holb and Schnabel, 2005). The International Federation of Organic Agriculture Movements (IFOAM) fully implemented their standards for organic crop production outlining the criteria that must be met to label agricultural products as “organic” (Anon., 1998a, 2009). In contrast to conventional fruit production, only natural products, such as farmyard manure, compost, soluble rock powder, copper and sulphur-based products, botanical soaps, botanical insecticides, microorganisms, sanitation practices, and biological products are

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permitted in organic fruit production (Anon., 1998a, 2009). From these options, copper-based products, elemental and lime sulphur are considered to be most effective for blossom blight control (Holb and Schnabel, 2005; Holb, 2006; Holb et al., 2008; Holb et al., 2013). Holb and Schnabel (2005) and Holb (2006) showed that elemental sulphur had low efficacy against the disease but reduced dosages of lime sulphur and copper showed significantly better control compared to untreated control. However, lime sulphur and copper compounds caused significant blossom toxicity, especially under wet conditions during the blooming period. These results indicated that allowed chemicals in organic cherry production are not as effective as in conventional or integrated production and in addition they are more phytotoxic. Therefore, it would be necessary to find other means of control options (e.g. sanitation and biological control) to resolve both problems that arise in the brown rot blossom blight control of organic sour cherry production.

Previous studies showed some promising results on some selected microbes against blossom and twig blight caused by *M. laxa* or *Monilinia fructicola*, such as *Epicoccum nigrum*, *Epicoccum purpurascens*, and *Aureobasidium pullulans* (e.g. Madrigal et al., 1994; Wittig et al., 1997). One of the promising microbes was the yeast-like fungus *A. pullulans* against blossom blight. Most cherry studies on *A. pullulans* focused on control efficacy against post-harvest rot of cherry. Only few studies evaluated efficacy of *A. pullulans* against blossom blight of cherries in small-scale trial studies (Wittig et al., 1997; Whipps and McQuilken, 2009; Obenaus et al., 2010). Wittig et al. (1997) in a three-year study in the US showed that two to three applications of *A. pullulans* during the blooming period reduced blossom blight relative to nontreated blossoms by 13–54% on 25-year old conventionally grown trees of sweet cherry cultivar Royal Anne. Recently a commercial biological product Boni Protect® forte containing *A. pullulans* was registered in Germany (www.bio-ferm.com/de/index.php?id=199), which is recommended for the control of fruit rots in strawberries and stone fruits (Whipps and McQuilken, 2009), as well as for the control of Monilia blossom and twig blight in stone fruits. A preliminary one-year report of Obenaus et al. (2010) in Germany showed that four applications of *A. pullulans* (Boni Protect forte) during the blooming period reduced blossom blight relative to nontreated blossoms by 78% on sour cherry cultivar Schattenmorelle in an organically grown orchard. Previous results indicated that *A. pullulans* may have acceptable efficacy against blossom blight of brown rot; however, no detailed study is available on multiyear effects of *A. pullulans* applications on blossom blight reductions in comparison with organically approved plant protection compounds of copper or lime sulphur. In addition, no data available on phytotoxicity and yield responses of applications of *A. pullulans vs* copper or lime sulphur during the bloom period in organic sour cherry production.

The aim of this three-year study was to compare the effectiveness and phytotoxicity of the treatments of copper hydroxide, lime sulphur, and their combinations with *A. pullulans* for blossom blight control in organic sour cherry production. The effect of treatments on yield was also determined. A major aim was to evaluate the integrated control approach of the combined A. *pullulans* treatments to reduce allowed chemical damage on spur-leaf clusters.

2. Materials and methods

2.1. Orchard and plant material

The study was conducted in an organic sour cherry orchard at Eperjeske (Hungary), located at 47°31′60″N and 21°37′60″E, during 2008, 2009 and 2010. The 5.8 ha orchard consisted of 19 rows with a distance of 6 m between trees and 4 m within a row. The orchard was planted in 1997 with self-fertile sour cherry cultivars including eight rows of cultivar (cv.) Érdi bőtermő. Cultivars were grafted on *Prunus mahaleb* rootstock. Trees have been grown according to the Hungarian organic production guidelines (Anon., 1997a) derived from IFOAM (International Federation of Organic Agriculture Movements) standards (Anon., 1998a, 2009). The guidelines have been applied since planting of the orchard in 1997. The orchard soil type was brown forest soil with alternating layers of clay substance. Trees were ca. 3–3.5 m tall during the three-year assessment period. Intra-row spacing between branches in the crown of adjacent trees was ca. 0.1–0.3 m and between adjacent rows was ca. 2.8 m. A 0.5 m wide strip of bare soil was maintained in the rows and grass was grown in the spacings between rows. The orchard was not irrigated. Treatments and observations were made on cv. Érdi bőtermő. The cultivar is susceptible to blossom blight (Holb and Schnabel, 2005). Cultivar Érdi bőtermő belongs to the early blooming group (Nyéki et al., 2003) and its fruit maturity dates range from June 13 to 22 (Soltész, 1997).

2.2. Environmental monitoring

Rainfall (mm day^{-1}) and mean daily temperature (°C day^{-1}) were recorded during the test periods in 2008, 2009 and 2010 using a Metos Compact agrometeorological station (Pessl Instrument GmbH, Weiz, Austria) from 15 April to 30 May in each year. Weather station was placed in the middle of the orchard and sensors were established at a 1.5 m height from the ground.

2.3. Fungicide treatments

Fungicide treatments were performed in the springs of 2008, 2009 and 2010. Each fungicide treatment was replicated five times (single tree replications). The whole experiment was set up as a split-plot design [split (year) – plot (fungicide)] according to Gomez and Gomez (1984). Fungicide treatments were: 1) untreated control (Untreated), 2) conventional fungicide (Conv.), 3) lime sulphur – reduced rates during bloom period (Lime-S), 4) copper

Table 1
Application rates in percentage active ingredient of fungicide treatments at various bloom stages of sour cherry (Eperjeske, Hungary, 2008–2010).

Fungicide treatments[b]	Bloom stages[a]				
	Bud break (BBCH 53)	Before bloom (BBCH 54)	Closed blossom (BBCH 59)	Full bloom (BBCH 65)	Petal fall (BBCH 70)
Conv.	–	–	0.12; 0.03[c]	0.12; 0.03	–
Co-Hydr.[d,e]	1.5	1.5	0.6	0.03	0.05
Lime-S[e]	1.5	1.0	1.5	1.0	1.1
Po-Carb.	0.5	0.5	–	–	–
A. pull.[e]	–	–	0.06	0.06	0.06

[a] Application timing: BBCH 53 = bud break, BBCH 54 = before bloom, BBCH 59 = closed blossom, BBCH 65 = full bloom, BBCH 70 = petal fall according to BBCH growth stage scale for stone fruits (Meier et al., 1994).

[b] Codes of treatments are: Conv. = conventional fungicide: vinclozoline and penconazole (Ronilan DF and Topas 100 EC, respectively), Co-Hydr. = copper hydroxide (Funguran-OH 50 WP), Lime-S = lime sulphur (Tiosol), Po-Carb. = potassium carbonate (Omni Protect), and *A. pull.* = *Aureobasidium pullulans* (BoniProtect forte).

[c] Conventional fungicide treatment was made with vinclozoline (0.12%) in 2008 and penconazol (0.03%) in 2009 and 2010.

[d] Copper hydroxide was selected from the copper compounds because according to Richardson (1997), compounds such as Cu_2O, $Cu(OH)_2$ and $CuCl_2 \cdot 3Cu(OH)_2$ had greater protective value than $CuSO_4$ as even though they are sparingly soluble they provide sufficient soluble copper to be toxic to fungi without adversely affecting the host.

[e] The selection of reduced dosages for copper hydroxide and lime sulphur sprays were based on results of Holb and Schnabel (2005).

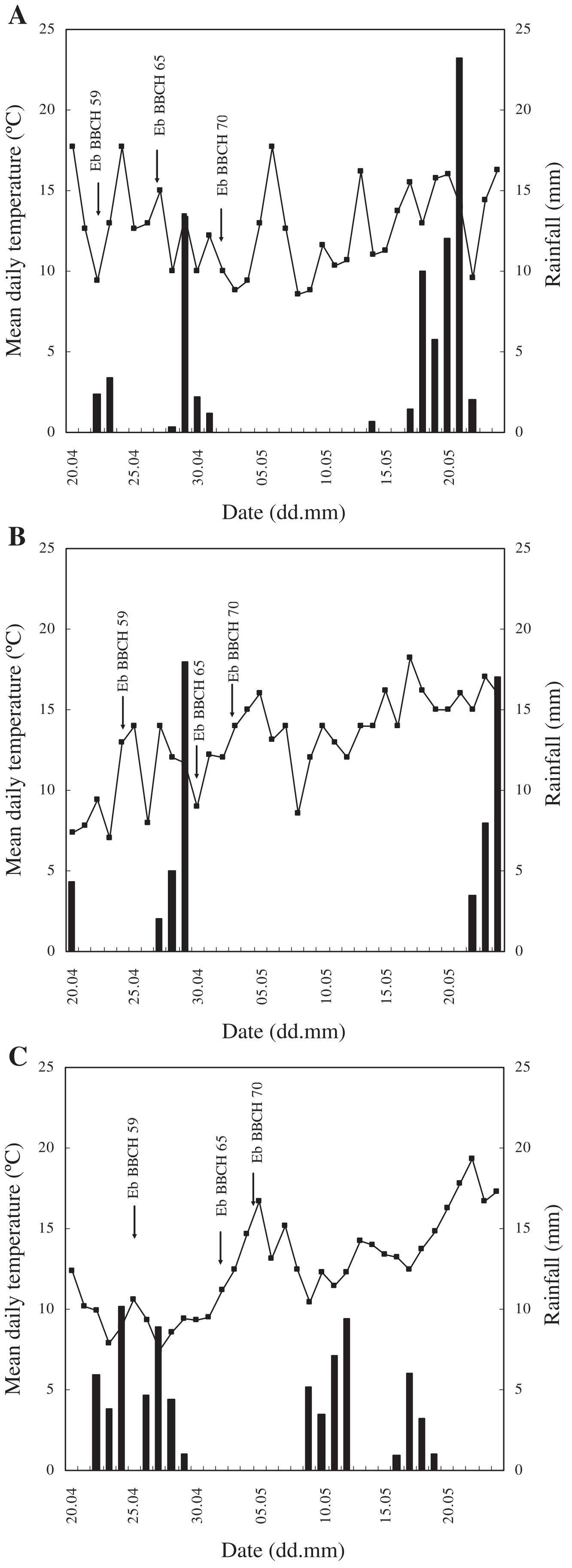
A
B
C
Mean daily temperature (ºC)
Rainfall (mm)
Date (dd.mm)
0
5
10
15
20
25
20.04
25.04
30.04
05.05
10.05
15.05
20.05
Eb BBCH 59
Eb BBCH 65
Eb BBCH 70

Table 2
Brown rot blossom blight incidence (%) caused by *Monilinia laxa* in sour cherry cultivar Érdi bőtermő in an organic sour cherry orchard (Eperjeske, Hungary, 2008–2010).

Fungicide treatments[a]	Application timing[b]	2008	2009	2010
Untreated	–	58.9[c] e[d]	42.8 e	54.9 e
Conv.	BBCH 59 and 65	4.9 a	3.9 a	4.4 a
Lime-S	BBCH 53, 54, 59, 65, 70	23.4 b	17.5 b	25.3 b
Co-Hydr.	BBCH 53, 54, 59, 65, 70	25.6 b	19.9 b	22.6 b
Lime-S + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	33.1 cd	25.2 c	36.2 cd
Co-Hydr. + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	30.7 c	24.7 c	32.8 c
Po-Carb. + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	37.1 d	31.4 d	39.4 d
	LSD$_{0.05}$[e]	4.9	5.1	6.2

[a] For explanation of treatment codes, see Table 1.

[b] Application timing: BBCH 59 = closed blossom, BBCH 65 = full bloom, BBCH 70 = petal fall according to BBCH growth stage scale for stone fruits (Meier et al., 1994).

[c] Brown rot incidence data shown are back-transformed means from arcsine $[\%]^{1/2}$ values.

[d] Values within columns followed by different letters are significantly different.

[e] LSD$_{0.05}$ = least significant differences at $P = 0.05$ level.

Table 3
Phytotoxicity on sour cherry cultivar Érdi bőtermő in a sour cherry orchard (Eperjeske, Hungary, 2008–2010).

Fungicide treatments[a]	Application timing[b]	2008	2009	2010
Untreated	–	0.6[c] a[d]	0.7 a	0.7 a
Conv.	BBCH 59 and 65	0.7 a	0.8 a	0.7 a
Lime-S	BBCH 53, 54, 59, 65, 70	2.5 c	2.6 b	2.6 c
Co-Hydr.	BBCH 53, 54, 59, 65, 70	2.2 bc	2.3 b	2.5 c
Lime-S + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	1.7 b	1.4 a	1.4 a
Co-Hydr. + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	1.5 b	1.5 a	1.5 a
Po-Carb. + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	1.5 b	1.3 a	1.5 a
	LSD$_{0.05}$[e]	0.7	0.8	0.8

[a] For explanation of treatment codes, see Table 1.

[b] For explanation of application timing, see Table 2.

[c] Phytotoxicity assessments were rated according to the scale: 0 = no damage; 1 = cluster size 60–80% of normal size, no necroses on cluster; 2 = cluster size less than 60% of normal size, no necroses on cluster; 3 = cluster size less than 60% of normal size and necroses on cluster less than 3%; 4 = cluster size less than 60% of normal size and 3–6% cluster necroses; and 5 = cluster size less than 60% of normal size and necroses on cluster more than 6%. Phytotoxicity data shown are back-transformed means from arcsine $[\%]^{1/2}$ values.

[d] For explanation, see Table 2.

[e] For explanation, see Table 2.

hydroxide – reduced rates during bloom period (Co-Hydr.), 5) lime sulphur in combination with *A. pullulans* (Lime-S + *Aureobasidium pull.*), 6) copper hydroxide in combination with *A. pullulans* (Co-Hydr. + *A. pull.*), and 7) potassium carbonate in combination with *A. pullulans* (Po-Carb. + *A. pull.*). Application rates and times are listed in Table 1. Applied products were: Funguran-OH 50 WP (77% copper hydroxide, Spiess-Urania Chemicals GmbH, Hamburg, Germany), Tiosol (29% calcium polysulphide, Tiosol Ltd., Kistelek, Hungary), Boni Protect forte 7.5 × 10^9 cfu g^{-1}, *A. pullulans* DSM14940 and DSM14941, Bio-Protect GmbH, Konstanz, Germany), Omni Protect (99% potassium carbonate K_2CO_3, Bio-ferm GmbH, Tulln, Germany), Ronilan DF (vinclozoline, 50%, BASF Hungaria Ltd., Budapest, Hungary), and Topas 100 EC (penconazol, 100 gr l^{-1}, Syngenta Ltd., Budapest, Hungary). All sprays were applied with a handheld spray gun (EMPASS, Huneschans 18 3905 XL Veenendaal, The Netherlands) with a ceramic hollow cone at 1.1–1.2 MPa with a volume of 1000 l ha^{-1}. All treatments received 0.5% wettable sulphur (Kumulus S 80% wettable sulphur, BASF Hungaria Ltd., Budapest, Hungary) at 7–16 day intervals, depending on weather conditions, after fruit set (BBCH 71) until the middle of July. A final spray of 2% copper hydroxide (Funguran-OH 50 WP) was applied in mid-August in all treatments and in all years.

2.4. Brown rot assessment

For each treatment and year, disease assessment was based on the percentage of blighted twigs 2 weeks after the petal fall application. One hundred randomly selected twigs from each quadrant of a tree were examined for disease symptoms as described previously (Osorio et al., 1994; Anon., 1998b). A blossom was considered to be diseased when the petals, calyx, and less than 1/3 of pedicel were necrotic (Tamm et al., 1995). A twig without flower was considered to be diseased if a blighted leaf was present. Values from the quadrants were averaged to obtain the percentage of diseased twigs per tree.

2.5. Assessment of phytotoxicity and yield

Phytotoxicity observations were made on 50 spur-leaf clusters (including vegetative and generative plant parts) per tree after two weeks of petal fall in all years as described in the study of Anon. (1997b). Briefly, phytotoxicity assessments were rated according to the scale: 0 = no damage; 1 = cluster size 60–80% of normal size, no necroses on cluster; 2 = cluster size less than 60% of normal size, no necroses on cluster; 3 = cluster size less than 60% of normal size and necroses on cluster less than 3%; 4 = cluster size less than 60% of normal size and 3–6% cluster necroses; and 5 = cluster size less than 60% of normal size and necroses on cluster more than 6%.

Yield was determined by the weight of all fruit per tree and treatment at harvest. Fruit yield was recorded on June 15, 18 and 14 in 2008, 2009 and 2010, respectively.

2.6. Statistical analyses

Brown rot incidence and phytotoxicity data were transformed to *angular* ($Y = \text{arcsine } [\%]^{1/2}$) before analysis. No transformation was necessary for yield data. Brown rot incidence, phytotoxicity, and yield data were analysed by split-plot ANOVA to evaluate the main effects of fungicide treatment, year, and their interactions. For brown rot incidence, phytotoxicity and yield data sets, significant *F*-tests ($P < 0.05$) were followed by a Least Significance Difference (LSD)-test for comparison of fungicide treatment means using LSD$_{0.05}$ values. Standard errors of differences (SED) and degrees of freedom (df) are given in the tables if appropriate. Genstat 5 Release 4.1 statistical package (Lawes Agricultural Trust, IACR, Rothamsted, UK) was used for all analyses. In order to quantify relationship between phytotoxicity and yield, Pearson's correlation coefficients were calculated among the two measures. Correlation analyses were done separately for the three years using Genstat 5 Release 4.1.

3. Results

3.1. Environmental monitoring

Rainfall was 22.9 mm and mean daily temperature was 12.6 °C during a 10-day period of bloom, in 2008 (Fig. 1A). In 2009, rainfall was 25 mm and mean daily temperature was 11.7 °C during a 9-day period of bloom (Fig. 1B). In 2010, rainfall was 19 mm and mean daily temperature was 10.3 °C during a 10-day period of bloom (Fig. 1C).

Fig. 1. Rainfall (bars), mean daily temperature (squares) and application timing of fungicide treatments [BBCH stages according to Meier et al. (1994)] of cultivar Érdi bőtermő (Eb) during 2008 (A), 2009 (B), and 2010 (C) in Eperjeske, Hungary. Bloom stages are: BBCH 59 = closed blossom, BBCH 65 = full bloom, BBCH 70 = petal fall.

3.2. Brown rot incidence

Analyses of variance on brown rot incidence values indicated significant differences amongst years ($P = 0.002$) and treatments ($P = 0.025$). There was no significant year × treatment interaction ($P = 0.067$).

The conventional fungicide treatment provided the best blossom blight control in each year (Table 2). Both copper and lime sulphur fungicide treatments resulted in improved blossom blight control ($P < 0.05$) when applied five times (at BBCH 53, 54, 59, 65 and 70) compared to untreated control. Among fungicide treatments suitable for organic production, the best control was achieved with copper hydroxide and lime sulphur treatments applied alone in each year. Copper hydroxide treatments applied five times, did not differ significantly ($P < 0.05$) from the corresponding lime sulphur treatments. All *A. pullulans* treatments, applied three times during bloom (at BBCH 59, 65 and 70) combined with lime sulphur, copper hydroxide or potassium carbonate applied twice before bloom (at BBCH 53 and 54), resulted in significantly higher blossom blight incidence compared to lime sulphur or copper hydroxide treatments applied five times. However, all *A. pullulans* treatments controlled blossom blight significantly better ($P < 0.05$) than the untreated control.

3.3. Phytotoxicity

Analyses of variance on phytotoxicity values indicated significant differences amongst years ($P = 0.004$) and treatments ($P = 0.017$). There was no significant year × treatment interaction ($P = 0.078$).

The conventional and the three treatments of *A. pullulans* in 2009 and 2010 did not cause significantly more damage on spur-leaf clusters ($P < 0.05$) compared to untreated control (Table 3). However, *A. pullulans* in 2008 caused significantly more damage on spur-leaf clusters ($P < 0.05$) compared to untreated control. Lime sulphur and copper hydroxide negatively affected spur-leaf clusters in all years, in those treatments where these fungicides were applied five times compared to either untreated or *A. pullulans* treatments in 2009 and 2010.

3.4. Fruit yield

Analyses of variance on yield data indicated significant differences amongst years ($P = 0.003$) and treatments ($P = 0.031$). There was no significant year × treatment interaction ($P = 0.094$).

The conventional fungicide treatment provided the highest yield in each year (Table 4). All treatments had significantly higher yield ($P < 0.05$) in 2008 and 2010 compared to untreated control. However, in 2009, all *A. pullulans* treatments combined with lime sulphur, copper hydroxide and potassium carbonate as well as the treatment of lime sulphur applied five times resulted in significantly not different yield compared to untreated plots.

3.5. Relationships between phytotoxicity and yield

Pearson's correlation coefficients showed that phytotoxicity and yield did not correlate significantly to each other ($r = 0.196$ and $P = 0.5767$ for 2008, $r = 0.208$ and $P = 0.5378$ for 2009 and $r = 0.185$ and $P = 0.6123$ for 2010).

4. Discussion

None of the treatments were as effective as the conventional fungicide treatment, but all treatments of lime sulphur, copper hydroxide and *A. pullulans* significantly reduced blossom blight when applied three times during bloom compared to nontreated plots. Although these treatments caused low to moderate phytotoxicity, they significantly increased crop yield compared with the untreated controls in two out of the three years.

Table 4

Yield (kg $tree^{-1}$) of sour cherry cultivar Érdi bőtermő in a sour cherry orchard (Eperjeske, Hungary, 2008–2010).

Fungicide treatments[a]	Application timing[b]	2008	2009	2010
Untreated	–	10.1[c] a[d]	11.7 a	10.1 a
Conv.	BBCH 59 and 65	15.7 c	16.2 c	15.5 c
Lime-S	BBCH 53, 54, 59, 65, 70	13.4 bc	14.2 abc	13.1 b
Co-Hydr.	BBCH 53, 54, 59, 65, 70	13.8 bc	14.4 bc	13.9 bc
Lime-S + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	12.7 b	12.6 ab	12.8 b
Co-Hydr. + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	12.6 b	12.9 ab	12.5 b
Po-Carb. + *A. pull.*	BBCH 53, 54 + BBCH 59, 65, 70	12.8 b	12.3 ab	12.7 b
	$LSD_{0.05}$[e]	2.4	2.5	2.3

[a] For explanation of treatment codes, see Table 1.

[b] For explanation of application timing, see Table 2.

[c] Yield data shown are back-transformed means from arcsine $[\%]^{1/2}$ values and represent kg $tree^{-1}$.

[d] For explanation, see Table 2.

[e] For explanation, see Table 2.

All *A. pullulans* combinations applied three times during bloom were less effective for blossom blight control than copper hydroxide and lime sulphur alone but they also caused less damage on spur-leaf clusters. Our results provide control alternatives for brown rot blossom blight by combining *A. pullulans* with allowed chemicals, although none of the treatments controlled blossom blight effectively compared to conventional standard such as described before (Holb and Schnabel, 2005; Holb et al., 2013). Only few previous studies evaluated efficacy of *A. pullulans* applied during bloom period of cherries (Wittig et al., 1997; Obenaus et al., 2010; this study). Wittig et al. (1997) showed that three applications of *A. pullulans* during the bloom period reduced blossom blight of sweet cherry relative to nontreated blossoms by 13–54%, while Obenaus et al. (2010) on sour cherry presented 78% blossom blight reduction relative to untreated plots when *A. pullulans* were applied four times during the bloom period. However, our study demonstrated an intermediate range of *A. pullulans* efficacy (between 51 and 72%) against brown rot blossom blight of sour cherry compared to untreated control. Efficacy differences may due to several factors such as *M. fructicola* and not *M. laxa* were assessed on larger and older trees in the study of Wittig et al. (1997) which were grown not under organic standards; while the results of Obenaus et al. (2010) were based on one year experimental data only and on a different sour cherry cultivar grown under dissimilar ecological conditions compared to this study.

In our experimental orchards, high inoculum pressures were combined with disease-conducive weather conditions which caused severe blossom blight epidemics in all years. Under such conditions sulphur and copper compounds as well as antagonistic microbes had relatively poor efficacy against blossom blight of cherries (e.g. Wittig et al., 1997; Holb and Schnabel, 2005; Holb, 2006; Holb et al., 2008; Obenaus et al., 2010), as it was also demonstrated in this study. Under lower disease pressures and less disease-conducive weather conditions, efficacy *A. pullulans* treatments may increase while spur-leaf cluster phytotoxicity remains low. Under such less favourable conditions, numbers of *A. pullulans* sprays are likely to be reduced to two or one occasion between closed bloom stage and petal fall compared to the three applications used in this study. Our results also indicated that the copper hydroxide, lime sulphur or potassium carbonate compounds in the *A. pullulans* treatments had no additional effects either on brown rot control efficacy or phytotoxicity. This indicates that cherry growers have the opportunity to select any of these compounds for *A. pullulans* spray applications which are the best suit to cultivar, ecological and weather conditions of the orchard.

In this study, three applications of *A. pullulans* during the bloom period caused significantly less damage compared to copper hydroxide or lime sulphur treatments, but they caused significantly more damage on spur-leaf clusters in two out of the three years compared to either untreated control or conventional fungicide treatments (Table 3). Copper fungicides are known to cause phytotoxicity in organic cherry production (Tamm et al., 2004; Holb and Schnabel, 2005). Lime sulphur phytotoxicity is dependent on the availability of moisture, if leaf wetness or relative humidity increases it can also enhance the phytotoxic effects of lime sulphur (Subhash, 1988; Tate et al., 2000; Trapman, 2001; Holb et al., 2003; Holb and Schnabel, 2005). *A. pullulans* had also previously identified as russet-inducing species on apple (Matteson Heidenreich et al., 1997) and on pear fruits (Spotts and Cervantes, 2002) but as it is shown in this study, the phytotoxic effect of combined *A. pullulans* treatments was lower on sour cherry leaf cluster compared to copper and lime sulphur compounds applied alone. Gildemacher et al. (2004, 2006) demonstrated that *A. pullulans* stimulates russet formation on apple fruits but the level of russet was dependent on *A. pullulans* isolates, the presence of other yeast species and fungicide use. Further studies also revealed that *A. pullulans* is able to use cutin from the apple fruit skin as a sole carbon source. Due to the degradation of the apple cuticle, the epidermis of the fruit is exposed, which triggers the formation of periderm (cork) tissue, which shows in the form of russeting (Goffinet et al., 2002, 2006). However, the study of Kunz and Haug (2006) showed that two or three applications of *A. pullulans* during bloom period did not increase fruit russet on apple cultivars Jonagored and Golden Delicious. Our sour cherry study, in agreement with the study of Kunz and Haug (2006), indicated that three applications of *A. pullulans* during bloom period did not increase phytotoxicity on spur-leaf clusters including young cherry fruits.

In this study, treatments of lime sulphur, copper hydroxide and *A. pullulans* applied at three times during bloom increased yield in two out of the three years compared to untreated control (Table 4). Holb and Schnabel (2005) also showed that sour cherry fruit yield was higher in copper hydroxide and lime sulphur treatments applied two or three times during bloom, compared to untreated plots, if the disease pressure was high. In this study, yield was not significantly higher in the treatments of lime sulphur or copper hydroxide applied five times compared to *A. pullulans* treatments despite the fact that blossom blight reduction was higher in those treatments of lime sulphur or copper hydroxide. However, phytotoxicity was lower in the *A. pullulans* treatments than on trees treated with lime sulphur or copper hydroxide alone. The phenomena of blossom blight reduction *vs* phytotoxicity may equalize so that the yield performs equally in the trees treated with either *A. pullulans* or lime sulphur or copper hydroxide.

5. Conclusions

In summary, three applications of *A. pullulans* may adequate against blossom blight control in organic sour cherry production compared to the corresponding lime sulphur or copper hydroxide treatments with the respect of efficacy, phytotoxicity and yield responses. However, none of the treatments controlled blossom blight effectively compared to conventional standard. This result implies that an improved integrated control strategy is needed for blossom blight control in organic sour cherry production. Other control methods (e.g. more effective biocontrol agents, inducers of systemic acquired resistance) in combination with sanitation, cultivar selection, and/or fungicide applications may provide more adequate blossom blight control in organic sour cherry production.

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