

# A during-infection spray strategy using sulphur compounds, copper, silicon and a new formulation of potassium bicarbonate for primary scab control in organic apple production

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**Abstract** In a field experiment conducted over two growing seasons, the effectiveness and phytotoxicity of inorganic fungicides such as sulphur, lime sulphur, copper, silicon and Armicarb (a new formulation of potassium bicarbonate) was compared with water for the control of primary apple scab infections in Belgium on high, medium and low scab-susceptible cultivars (cvs ‘Pinova’, ‘Pirouette’ and ‘Reinette des Capucins’, respectively). In order to drastically reduce the amount of fungicide applied in the orchard, two approaches were used: (1) a strategy involving spraying during the infection process, before fungal penetration and (2) a tunnel sprayer machine for treatment applications. Under field conditions highly favourable for disease, low rates of elemental sulphur (31.8 and 38.6 kg ha<sup>-1</sup> year<sup>-1</sup> in 2005 and 2006, respectively) combined with low rates of copper (2.1 kg ha<sup>-1</sup> year<sup>-1</sup> in both years) provided the best scab control and reduced scab severity on the fruits of cv. ‘Pinova’ by 97 and 98% compared with water control in 2005 and 2006, respectively. Lime sulphur was much more effective than wettable sulphur and appeared to be efficient at temperatures below 10°C,

but its effectiveness against apple scab decreased if the treatments were applied 12–24 h later than in the ‘during-infection’ spray strategy. Armicarb used alone significantly reduced apple scab severity on the leaves and fruits of the three cultivars compared with the water control. Its effectiveness was as good as wettable sulphur applied using the same timing and dosage. Silicon reduced apple scab on fruits very slightly, but not on leaves. The amounts of wettable sulphur, lime sulphur, copper, silicon and potassium bicarbonate used in this experiment to control apple scab were not phytotoxic, did not increase fruit russet, did increase the yield of each cultivar and did not affect summer density of the beneficial *Typhlodromus pyri*. The potential and limitations of ‘during-infection’ spraying as a protection strategy against apple scab in organic farming are discussed.

**Keywords** Alternative control · Disease management · Lime sulphur · Natural substances · Polygenic resistance · *Typhlodromus pyri* · *Venturia inaequalis*

## Introduction

Over the past decade, public concern about pesticide residues on fruit and in the environment have generated much interest in organic apple production. One of the main components of successful organic apple production in a humid environment is the

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control of scab (*Venturia inaequalis*). Only a few approved chemical compounds are available for disease control under organic guidelines, based mainly on sulphur and copper. Copper is effective against apple scab but, for environmental reasons, a new EU Council Regulation (EC No 473/2002) allows only a reduced input of copper fungicides; in some countries the use of copper fungicides is no longer allowed. Because no new options have been proposed, the remaining ones for apple scab control are the use of elemental sulphur and lime sulphur products (Holb et al. 2003). Sulphur compounds are often less effective than copper-based compounds, especially under cold weather conditions, and apple scab control requires large amounts of sulphur compounds to compensate for copper. Several studies have shown that the repeated application of large amounts of sulphur compounds has ecotoxicological and phytotoxic side effects (Mills 1947; Tweedy 1981; Holb et al. 2003; Palmer et al. 2003). Therefore, organic apple growers under humid climate conditions have to contend with the results of using large amounts of sulphur compounds which could lead to leaf phytotoxicity, reduced fruit quality (Holb et al. 2003) and undesirable effects on beneficial fauna (Kreiter et al. 1998). Typically, in an organic apple orchard, 10–26 sprays are applied against apple scab in each season, depending on cultivar susceptibility, weather conditions and the amount of inoculum (Holb 2006).

Various strategies have been proposed to reduce fungicide applications on apple. Several studies have shown that early warning systems based on disease forecasting models that give timely information of apple scab infection periods have the potential to limit the use of fungicides (MacHardy 1996; Trapman and Polfliet 1997; Hindorf et al. 2000). An after-infection programme can significantly reduce fungicide applications for scab control (Funt et al. 1990; Holb et al. 2003). However, organic growers have not widely adopted this new technology, including the after-infection spray approach, probably because of the lack of (1) compounds with curative properties and (2) an accurate local warning system. Most sulphur compounds have poor curative properties; the exception is lime sulphur, which might have good curative properties against apple scab (Holb et al. 2003; Montag et al. 2005). Although the use of lime sulphur is permitted under EU regulations for organic production (EEC No

2092/91), it is currently not allowed to be used in Belgium. Therefore, the ‘during-infection’ spray strategy, involving spraying during the infection process, would be a promising scab control approach using compounds with poor curative properties and requiring fewer treatments than preventive control.

Another promising way to significantly reduce the use of fungicides for scab control would be to expand the cultivation of scab-resistant cultivars (Ellis et al. 1998; MacHardy et al. 2001). Monogenic resistance cannot be considered durable. New scab races, virulent to the *Vf* gene, exist in most European countries (Gessler et al. 2006). Apple breeding programmes based on polygenic resistance are therefore now of greater interest (Lateur et al. 1999; Gessler et al. 2006).

Other options for controlling apple scab may be to use natural substances as fungicides that have no known adverse effects on the environment and human health. Bicarbonates are one of several control options now attracting attention. They are common food additives allowed in many applications under European and North American regulations and they have been used against several plant pathogens (Horst et al. 1992) and recently against apple scab (Beresford et al. 1996; Schulze and Schönherr 2003; Ilhan et al. 2006; Jamar and Lateur 2007; Jamar et al. 2007). The use of soluble silicon in horticulture, as a protective agent against several fungal pathogens, has also been reported (Belanger et al. 1995).

The objectives of this study were: (1) to evaluate the relative effectiveness of inorganic fungicides such as wettable sulphur, lime sulphur, copper, potassium bicarbonate and silicon for primary scab control, and (2) to evaluate the effectiveness of the ‘during-infection’ spray strategy using reduced amounts of fungicide. Effectiveness, phytotoxicity and effects on yield, fruit quality and *T. pyri* populations were studied on high, medium and low scab-susceptible cultivars in a modern apple orchard system.

## Materials and methods

### Orchard design and equipment

The study was conducted in 2005 and 2006 in a well-maintained experimental apple orchard (3.5×1.5 m) planted in 2002 at Gembloux, Belgium. A split-plot

design based on six randomized blocks was used. Each block comprised six rows (plots) of 18 dwarf trees. The plots consisted of six trees of cv. 'Pinova', six trees of cv. 'Rubinstep-Pirouette' and six trees of cv. 'Reinette des Capucins' grafted onto M9 rootstocks. The cultivars were randomized to subplots within the plots. The cvs 'Pinova', 'Pirouette' and 'Reinette des Capucins' were reported to be high, medium and low scab-susceptible cultivars, respectively (Jamar and Lateur 2007).

The trees were grown according to organic production standards (Anon. 2007). The orchard soil was heavy loam containing 1.2% C and each year it received 1,000 kg ha<sup>-1</sup> of organic fertilizers (5% N) and 1,000 kg ha<sup>-1</sup> of hydrated lime for pH enhancement. Any significant soil nutrient limitation was registered. Orchard maintenance included a centrifugal training system (Simon et al. 2006). The trees reached an average of 3 and 3.25 m in 2005 and 2006, respectively. For weed control under the tree rows, a cover-crop machine was successfully used four times a year. Grass alleys between the tree rows were kept short. Leaf analysis revealed B, Zn, Mn deficiency, therefore six correcting foliar treatments were applied during the growing season in both years.

Potential infection periods, based on Mills criteria, were recorded in the field using a METY computer-based weather recorder (Bodata Co. Ltd, Dordrecht, The Netherlands) connected to a RIMpro scab warning system (Trapman and Polfliet 1997) from 15 March to harvest in both 2005 and 2006. The scab warning system calculated the infection periods based on the hourly detected meteorological data, the modified Mills table (MacHardy and Gadoury 1989), the simulation of ascospore release and the effect of previously used sprays. Furthermore, the local climatic forecasts were registered daily in the RIMpro for infection risk extrapolations.

### Treatments

The experiment was conducted on 648 trees, involving six experimental spray programmes of treatment in both years. Each treatment was applied to 108 trees (36 trees per cultivar). The treatments were randomised in plots within each of the six blocks. Each treatment combination occurred once in each block. The experimental orchard could be considered as homogeneous at the beginning of the

experiment. The treatments were applied with a tunnel sprayer (Munckhof, 5961 CV Horst, The Netherlands) to prevent spray-drift and to reduce pesticide dispersal. In order to achieve various treatments in a single run, the sprayer was fitted with six individual tanks.

The six spray programmes in both years were as follows: (1) water control (Control); (2) 1.6% potassium bicarbonate (0.8% during flowering; PB); (3) 1.6% sulphur from wettable sulphur (0.8% during flowering) + 1% calcium hydroxide (WS); (4) 1.6% sulphur from lime sulphur (0.8% during flowering; LS); (5) 0.1% potassium silicate in 2005 (PSi) and 1.6% sulphur from lime sulphur (0.8% during flowering) under delayed spray timing in 2006 (LSd); and (6) 1.6% sulphur from wettable sulphur (0.8% during flowering) + 0.16% copper from the hydroxide form before flowering (WSCu; Table 2). The same treatments were applied in the same plots in both years except for potassium silicate (PSi), which was replaced by delayed lime sulphur (LSd) application in 2006. Calcium hydroxide was added to the wettable sulphur to obtain as much calcium in the wettable sulphur treatments (WS) as in the lime sulphur treatments (LS and LSd).

All treatments were applied at a low spray rate of 300 l ha<sup>-1</sup>. The treatment timings, defined as the number of hours multiplied by the mean temperature in °C (degree-hours) between the onset of rain (associated with infection) and the time of application, were registered for each treatment in both years (Table 1). In both 2005 and 2006, the treatments were applied during each potential primary infection period identified by the RIMpro scab warning system. They were applied during the infection process, after ascospore inoculation and before hyphal penetration (<300 DH). Thus, treatments were applied just before or at the beginning of the infection risk periods detected by the RIMpro scab warning system. To anticipate whether the primary infection periods were associated with the infection periods forecast by the RIMpro, the extrapolation system using the short-term weather forecasts was used. The treatments were applied on 30 May 2005 and on 9 April, 30 May and 15 June 2006, although RIMpro did not consider primary infection risk on those days; potential infections were then based on revised Mills criteria. A delayed spray programme, consisting of spraying 12–24 h after the 'during-

**Table 1** Degree-hours (DH) between the onsets of the rain period associated with primary infection risks and the time of spraying

2005						2006						
Scab infections					DH	Scab infections					DH	
Date	F <sup>a</sup>	T <sup>b</sup>	Mills <sup>c</sup>	RIM <sup>d</sup> value	Early spray	Date	F	T	Mills	RIM value	Early spray	Delayed spray
27 March	C3	9.4	L	93	62	30 March	C3	9.1	S	123	90	90
14 April	D	9.8	M	1,018	85	09 April	D	7.8	L	0	50	194
17 April	E	5.7	S	1,044	125	14 April	D	7.9	S	396	269	386
25 April	E2	9.9	S	138	220	01 May	E2	6.2	M	71	170	269
29 April	F2	11.6	S	197	180	08 May	F2	10.7	M	13	189	302
03 May	G	10.1	M	113	253	18 May	H	12.4	S	92	129	219
08 May	H	6.1	M	44	270	20 May	HI	9.7	L	160	258	376
30 May	I	8.1	M	0	290	22 May	I	10.6	S	165	242	364
						30 May	I	7.2	M	0	215	335
						15 June	J	14.7	S	0	228	372

<sup>a</sup>F = Tree growth stages according to the Fleckinger-growth stage scale: C3 green leaf tip, D green bud, E early tight cluster, E2 tight cluster, F2 full bloom, G petal fall, H first fruit set, I fruit setting, J fruit swelling

<sup>b</sup>T = mean temperature between the onset of the rain associated with the infection and the time of spraying

<sup>c</sup>L low, M moderate, S severe infections according to the revised Mills criteria

<sup>d</sup>Infection risks (day values) according to RIMpro 2005 and 2006, respectively

**Table 2** Description of the spray programmes used in 2005 and 2006

Spray programme	Trademark (Manufacturer)	Active ingredient (%)	Active ingredient application rate (%)	Total a.i. amount (kg ha <sup>-1</sup> year <sup>-1</sup> ) <sup>a</sup>	
				2005	2006
Control	Water control	— <sup>b</sup>	—	—	—
PB	Armcarb®100 (Helena Chemical Company, USA)	Potassium bicarbonate (85%)	1.6 <sup>c</sup> (0.8) <sup>d</sup>	31.9	38.6
WS	Thiovit jet (Syngenta Agro, France)	Elemental sulphur (80%)	1.6 (0.8)	31.9	38.6
	Supercalco 95 (Carmeuse, Belgium)	Calcium hydroxyde (98%)	1.0 (1.0)	21.0	25.2
PSi	Potassium silicate solution (Sigma-Aldrich, Belgium)	Potassium silicate (34%)	0.1 (0.1)	1.3	—
LSd	Polisolfurio di Calcio (Polisenio, Italy)	Elemental sulphur (23%)	1.6 (0.8)	—	38.6
LS	Polisolfurio di Calcio (Polisenio, Italy)	Elemental sulphur (23%)	1.6 (0.8)	31.9	38.6
WSCu	Thiovit jet (Syngenta Agro, France)	Elemental sulphur (80%)	1.6 (0.8)	31.9	38.6
	Kocide WG (Griffin Europe, Belgium)	Copper (under hydroxide form) (40%)	0.16	1.3	1.3

For each spray programme, ten and twelve treatments (300 l ha<sup>-1</sup>) were applied during the growing seasons, from March to harvest, in 2005 and 2006, respectively, including one treatment during flowering and two treatments during summer in both years

<sup>a</sup>Amount of active ingredient (a.i.) applied during the growing seasons, from March to harvest, including a 30% saving with the tunnel sprayer process. Each year, all spray programmes received two additional post-harvest applications of 0.2% copper, mainly for European canker (*Nectria galligena*) control

<sup>b</sup>No values are applicable

<sup>c</sup>Application rate apart from flowering

<sup>d</sup>Values in brackets are application rate during flowering; for the WSCu treatment, the Kocide WG was applied only before flowering

infection' spraying, was also implemented in 2006 with lime sulphur (LSd).

Two additional treatments were applied on two secondary infection risk periods during the summer (on 11 and 18 July in 2005 and on 12 July and 14 August in 2006). All spray programmes received two post-harvest applications of 0.2% copper (from the hydroxide form) in both 2005 and 2006, mainly for European canker (*Nectria galligena*) control. Insect control was the same for all treatments and followed standard European organic guidelines (Anon 2007).

The applied products included: wettable sulphur (Thiovit jet, 80%, Syngenta Agro, Saint Cyr l'Ecole Cedex, France), copper hydroxide (Kocide WG, 40%, Griffin Europe, Zaventem, Belgium), lime sulphur (Polisolfurio di Calcio, 23% of elemental sulphur, Polisenio, Lugo, Italy), calcium hydroxide (Supercalco 95, 97.7%, Carmeuse, Seilles, Belgium), potassium silicate (Soluble potassium silicate, 34%, Sigma-Aldrich, Bornem, Belgium) and potassium bicarbonate (Armcarb<sup>1</sup>100, 85%, Helena Chemical Company, Collierville, TN, USA; Table 2).

The common formulation of lime sulphur products contains a mixture of calcium polysulphide and a small amount of calcium thiosulphate. The Polisolfurio di Calcio contains 23% of elemental sulphur which was considered as the active ingredient content in this paper. Armcarb<sup>®</sup> 100 is a new formulation registered in the USA by the Environmental Protection Agency (EPA) and can be used in organic farming systems in this country. It was chosen for this study for its effectiveness under greenhouse conditions and its adapted formulation for foliar applications (Jamar et al. 2007).

A recovery system that included a continuous recycling process in the tunnel sprayer led to an average of 30% being saved on the applied spray mixtures when spraying under moderate wind speed ( $\leq 15 \text{ km h}^{-1}$ ) in a 6-year-old apple orchard. The amounts of active ingredients applied per ha and per year, including a 30% saving, are indicated in Table 2. The amount of elemental sulphur used annually, for the WS, LS, LSd, WSCu spray programmes, was 31.9 and 38.6 kg ha<sup>-1</sup> in 2005 and 2006, respectively. For the WSCu spray programme, an additional 1.3 kg ha<sup>-1</sup> of copper was applied during the growing seasons in both years. The rate of potassium bicarbonate in the PB spray programme was also 31.9 and 38.6 kg ha<sup>-1</sup> in 2005 and 2006, respectively (Table 2).

## Scab incidence and severity assessment

Disease assessments were made on leaves and fruits in both years. For leaf severity assessments, 10 shoots per tree were recorded on 17 and 20 June in 2005 and 2006, respectively. Observations were made on 10 older leaves per shoot. A 1–9 scale was used whereby: 1=no scab lesions; 2= $\leq 1\%$  infected leaves with at least one lesion; 3= $\leq 5\%$  infected leaves with at least one lesion; 4=5–50% infected leaves with at least one lesion; 5= $\geq 50\%$  leaves with lesions with  $\leq 5\%$  leaf area spotted; 6=5–25% leaf area spotted; 7=25–50% leaf area spotted; 8=50–75% leaf area spotted; and 9=maximum infection, leaves black with scab (Lateur and Blazek 2002).

The disease assessment on fruit was made on harvested fruits, from 15 to 31 October in both years. The percentage of diseased fruit was assessed on the whole yield collected per plot. Fruit incidence (FI) was calculated as the proportion of infected fruits with at least one scab lesion. Scab severity on fruits was assessed on the whole yield from each plot according to a scale of 1 to 6 based on a standard diagram method reported by Croxall et al. (1952) whereby: 1=no scab; 2=0–1%; 3=1–5%; 4=5–20%; 5=20–50%; and 6= $\geq 50\%$  fruit surface covered by scab. Fruit severity (FS) was defined as the mean proportion of the fruit surface covered by scab and was calculated using the following equation:

$$FS = \frac{n1 \times 0}{nt} + \frac{n2 \times 0.5}{nt} + \frac{n3 \times 2.5}{nt} + \frac{n4 \times 12.5}{nt} + \frac{n5 \times 35}{nt} + \frac{n6 \times 75}{nt}$$

where  $n1$  to  $n6$  represent the number of fruits in each category;  $nt$  represents the total number of fruits; and the coefficients 0, 0.5, 2.5, 12.5, 35 and 75 represent the median of the lower and upper boundaries of classes 1 to 6, respectively.

## Yield, fruit quality and leaf phytotoxicity

Fruits were harvested on 20 and 21 September and 14 October in 2005 and on 21 and 25 September and 17 October in 2006 for cvs 'Pirouette', 'Reinette des Capucins' and 'Pinova', respectively. Yield was characterised by the weight of all harvested fruits and was classified into four size categories (<60; 60–



70; 70–80 and >80 mm) when all fruits per plot were collected. The number of harvested fruits associated with yield was assessed for each plot. No hand thinning had been done during the growing season for any cultivars.

Fruit russet was registered for the whole yield, after harvest, from 15 to 31 October in both years. Fruit russet was assessed according to EPPO/OEPP standards based on a scale of 1 to 4 whereby: 1=no russet; 2=< 10%; 3=10–30%; and 4=30–100% russet on the fruit surface area. The fruit russet severity index (FR) was calculated using the following equation:

$$FR = \frac{n1 \times 0}{nt} + \frac{n2 \times 5}{nt} + \frac{n3 \times 20}{nt} + \frac{n4 \times 65}{nt}$$

where  $n1$  to  $n4$  represent the number of fruits in each category;  $nt$  represents the total number of fruits; and the coefficients 0, 5, 20 and 65 represent the median of the lower and upper boundaries of classes 1 to 4, respectively.

The percentage and weight of the first-class fruit were determined. In our experiment, the first-class fruit was defined as fruits with a scab severity of <1% (category 1 and 2), russet <10% (category 1 and 2) and size >60 mm (category 2, 3 and 4), irrespective of all other parameters.

Leaf phytotoxicity observations were made on five spur-leaf clusters per tree on 3 and 5 June in 2005 and 2006, respectively. Phytotoxicity was assessed on the whole leaf lamina following EPPO/OEPP standards. Leaf phytotoxicity assessments were rated on a 0–5 scale, as follows: 0=no damage; 1=leaf size 60–80% of normal size and no leaf necroses; 2=leaf size < 60% of normal size and with brown margins (<3% leaf necroses); 4=leaf < 60% of normal size and 3–6% leaf necroses; and 5=bumpy small leaf and >6% leaf necroses, as described by Holb et al. (2003).

#### Effects on the predatory mite *Typhlodromus pyri* population

The predatory mite *Typhlodromus pyri* (Acari: Phytoseiidae), particularly useful for the biological control of phytophagous mites such as *Panonychus ulmi* and *Aculus schlechtendali*, was imported from an IPM orchard into the experimental orchard in 2002. The introduction of *T. pyri* was achieved in August by hanging a 1 year-old branch from the IPM orchard inside each tree of the experimental orchard in order

to allow a free propagation of the predatory mite. A regular distribution of the *T. pyri* population was registered in June 2003 and 2004. The residual effects of treatments, applied to control apple scab, on *T. pyri*, *P. ulmi* and *A. schlechtendali* densities were evaluated on cvs ‘Pinova’, ‘Pirouette’ and ‘Reinette des Capucins’ in both 2005 and 2006. The treatments were the same as described earlier. Mite density assessments were made on 17 May, 10 June and 29 July in 2005 and on 15 May, 7 June and 31 July in 2006. For each assessment, 108 leaves per treatment (36 leaves per cultivar) were collected and observed in the laboratory. One leaf per tree was taken at shoulder height from a 1 year-old shoot in the external part of the canopy. Zeiss magnifying glass binoculars were used to count adult mites on the lower surface of each leaf.

#### Data analysis

The years were analysed separately for each variable. The percentage data were transformed in arcsine before performing an analysis of variance. No transformation was carried out for other measures. The data were analysed using SAS software version 9.1 (SAS Institute, Cary, North Carolina, USA) and the Student–Newman–Keuls multiple range test was applied to determine whether the differences between treatments were significant. All the statistical evaluations were conducted at a significance level of  $P=0.05$ .

## Results

#### Infection periods

In 2005 and 2006 there were 8 and 10 Mills infection periods, respectively, recorded from the end of March to mid-June in which one infection period occurred during flowering, whereas the RIMpro scab warning system identified only seven potential infection periods in both years. The primary infection periods, based on the revised Mills criteria, were severe in three and five instances, moderate in four and three instances and low in one and two instances in 2005 and 2006, respectively (Table 1). There was heavy disease pressure during the primary infection seasons, as revealed by the high scab infection rates recorded in untreated cv. ‘Pinova’ plots (Control) in both years (Table 3).

**Table 3** Effects of low-rate treatments on apple scab on cvs ‘Pinova’, ‘Pirouette’ and ‘Reinette des Capucins’ in 2005 and 2006 (June and harvested fruits assessments)

Treatments <sup>a</sup>	Pinova			Pirouette			Reinette des Capucins		
	LSv <sup>b</sup>	FI	FS	LSv	FI	FS	LSv	FI	FS
	(1–9)	(%)	(%)	(1–9)	(%)	(%)	(1–9)	(%)	(%)
<b>2005</b>									
Control	6.8 a <sup>c</sup>	84.5 a	6.2 a	3.1 a	45.6 a	1.2 a	1.7 a	0.0	0.0
PSi	6.3 a	81.3 a	4.5 b	3.0 a	46.3 a	0.8 ab	1.6 a	0.0	0.0
PB	1.9 b	55.3 b	1.8 c	1.4 b	28.1 b	0.4 b	1.2 b	0.0	0.0
WS	2.2 b	45.4 c	0.9 cd	1.5 b	24.4 bc	0.3 b	1.0 b	0.0	0.0
LS	1.8 b	27.0 d	0.4 d	1.2 b	15.1 bc	0.2 b	1.0 b	0.0	0.0
WSCu	1.1 c	18.4 e	0.2 d	1.1 b	11.0 c	0.1 b	1.0 b	0.0	0.0
<i>F</i> -test	***	***	***	***	***	*	*	ns	ns
<b>2006</b>									
Control	7.0 a	99.1 a	18.7 a	3.7 a	38.8 a	2.2 a	2.4 a	0.0	0.0
PB	4.9 b	81.3 b	7.7 b	2.5 b	21.3 b	0.7 b	1.2 b	0.0	0.0
WS	4.6 c	82.5 b	9.1 b	2.6 b	21.1 b	0.5 b	1.3 b	0.0	0.0
LSd	3.2 d	65.9 c	2.2 c	2.2 c	8.3 c	0.2 c	1.1 b	0.2	0.0
LS	2.7 e	45.0 d	1.2 c	2.0 d	4.2 d	0.0 c	1.1 b	0.0	0.0
WSCu	2.7 e	21.1 e	0.4 c	1.9 d	2.1 d	0.0 c	1.1 b	0.0	0.0
<i>F</i> -test	***	***	***	***	***	***	***	ns	ns

\* $P \leq 0.05$ \*\* $P \leq 0.01$ \*\*\* $P \leq 0.001$ 

<sup>a</sup> Control water-treated plots, PSi potassium silicate, PB potassium bicarbonate, WS wettable sulphur, LS lime sulphur, LSd lime sulphur in delayed spray timing, WSCu wettable sulphur + copper hydroxide (see Table 2)

<sup>b</sup> LSv = leaf severity using a 1–9 scale, where 1 = no scab and 9 = maximum scab infection; June assessment shown here. FI = fruit incidence: proportion of total harvested fruits with at least one spot. FS = fruit severity: mean scabbed area or mean proportion of fruit surface covered by scab

<sup>c</sup> Values within columns followed by different letters are significantly different ( $P \leq 0.05$ ) according to the Student–Neuwman–Keuls multiple range tests. *F*-test = ns (non-significant) at  $P > 0.05$

## Apple scab assessments

Apple scab symptoms present in the untreated plots in the orchard gave the following ratings for the cultivars: high, medium and low scab-susceptible for cvs ‘Pinova’, ‘Pirouette’ and ‘Reinette des Capucins’, respectively (Table 3).

In both years, all the treatments significantly reduced apple scab compared with the untreated control for all cultivars. In both years, the combined copper and wettable sulphur treatments (WSCu) gave the best apple scab control on both leaves and fruits for all cultivars. With this treatment, the scab severity on fruits of the scab-susceptible cv. ‘Pinova’ was reduced by 97% and 98% compared with the water control in 2005 and 2006, respectively (Table 3). In 2005 and 2006, the lime sulphur treatments (LS)

resulted in significantly lower scab damage on both the leaves and fruits of cv. ‘Pinova’ compared with wettable sulphur treatments (WS), while the same amount of elemental sulphur was applied. The delayed lime sulphur treatment (LSd) was less effective than the ‘during-infection’ lime sulphur treatment (LS), although the timing of the applications differed by no more than 0–144 degree-hours (DH) (Table 1). For the medium scab-susceptible cv. ‘Pirouette’, lime sulphur treatments (LS) resulted in almost the same level of scab control achieved by the combined copper and sulphur treatments (WSCu). In most cases, for the scab-susceptible cv. ‘Pinova’, the combined sulphur and copper treatment (WSCu) gave better control than lime sulphur (LS).

In 2005 and 2006, potassium bicarbonate treatments (PB) significantly reduced apple scab severity

on leaves and fruits compared with the water control (Table 3). In most cases, potassium bicarbonate (PB) was as effective as the wettable sulphur treatment (WS), using the same amount of active ingredients for both treatments. In 2005, the potassium silicate treatments (PSi) at 0.1%, using the ‘during-infection’ spray strategy, did not reduce scab severity on leaves, but did reduce it very slightly on fruits.

#### Yield, fruit quality and leaf phytotoxicity

None of the treatments adversely affected leaves in either year (no phytotoxicity, leaf size reduction or necrotic damage). All scores ranged between 0 and 0.3 on the 0–5 scale used (data not shown). None of the treatments adversely affected fruit russet compared with the untreated control in either 2005 or 2006. The average fruit russet severity index (FR) was 3.2 and 3.5% for cvs ‘Pinova’ and ‘Pirouette’ and 5.8 and 6.2% for cv. ‘Reinette des Capucins’ in 2005 and 2006, respectively (data not shown). Fruit russet was slightly higher on cv. ‘Reinette des Capucins’; this cultivar is genetically more prone to russet than the two other cultivars.

In most cases, compared with the untreated control, all treatments significantly increased overall yield per tree, reduced the proportion of fruits smaller than 60 mm and increased the amount of first-class fruit (Table 4). For cv. ‘Pinova’, the yield values on plots treated with WSCu were 2.6 and 4.6 times higher than in the control plots in 2005 and 2006, respectively. For cv. ‘Reinette des Capucins’, with a very low scab rate, the sulphur-based treatments (WS, LS, LSd, WSCu) also increased yield compared with the non-sulphur-based treatments (Control, PSi and PB), largely as a result of the effects on fruit number per tree rather than on mean fruit weight (Table 4). The lime sulphur treatment (LS) did not affect mean yields  $\text{ha}^{-1}$  compared with wettable sulphur treatments (WS and WSCu; Table 4).

#### Effects on the predatory mite *Typhlodromus pyri* population

In both 2005 and 2006, winter observations of 2- and 3 year-old branches did not show any eggs of *P. ulmi*. Assessments of the leaves in the spring and summer did not reveal the presence of any *P. ulmi* populations under any treatments. July assessment of

*A. schlechtendali* showed that all the sulphur treatments significantly reduced its density to 0.1 individuals per leaf compared with an average of two individuals per leaf from untreated plots in both years (data not shown). All the treatments slightly reduced the predatory *T. pyri* density level in June 2005 and in May and June 2006, but not in May and July 2005 or in July 2006 compared with the untreated control (Table 5). There was no significant difference in the *T. pyri* density level among the lime sulphur (LS, LSd), wettable sulphur (WS) and wettable sulphur combined with copper (WSCu) treatments in either 2005 or 2006.

#### Discussion

The results of this study clearly demonstrate that the ‘during-infection’ spray strategy, was effective in controlling apple scab with a reduced amount of fungicides such as sulphur, lime sulphur, copper and potassium bicarbonate. Our results show that it was possible to produce 75% of marketable fruit ( $35.3 \text{ ton ha}^{-1}$  of class 1) from the scab-susceptible cv. ‘Pinova’ from a 5 year-old orchard with no more than 38.6 kg of elemental sulphur and 2.1 kg of copper (from the hydroxide)  $\text{ha}^{-1} \text{ year}^{-1}$ , whereas no more than 2% of marketable fruits ( $0.2 \text{ ton ha}^{-1}$  of class 1) were obtained from untreated plots (Table 4). These amounts of fungicides are about 70% below the amounts usually used to control apple scab in organic production under humid climate conditions (Ellis et al. 1994; Holb et al. 2003; Palmer et al. 2003). Up to  $110 \text{ kg ha}^{-1} \text{ year}^{-1}$  of elemental sulphur combined with  $8 \text{ kg ha}^{-1} \text{ year}^{-1}$  of copper were used for scab control in organic apple production in The Netherlands (Holb and Heijne 2001).

Fungicides were applied shortly after rainfall associated with primary scab infections. Treatments were applied during the infection process, sometimes on drying leaves. Ascospores had been discharged, susceptible tissue was present and, in most cases, the minimum temperature and leaf-wetness conditions for infection accorded with the revised Mills criteria, but penetration of the cuticle had not yet occurred (Table 1). Below 250 DH after rainfall, few ascospores reach the stadium of penetration, and a relevant portion will reach this stage only after 300 DH (Smereka et al. 1987; MacHardy 1996). Sprays below 300 DH can be considered to be applied during the



**Table 4** Effect of treatments on overall yield per tree, total fruit number per tree (FN), proportion of fruits < 60 mm (<60), and Class 1 yield (Class 1) of 4- and 5-year-old trees of cvs 'Pinova', 'Pirouette' and 'Reinette des Capucins' in 2005 and 2006, respectively

Treatments <sup>a</sup>	cv. 'Pinova'				cv. 'Pirouette'				cv. 'Reinette des Capucins'			
	Yield (kg tree <sup>-1</sup> )	FN	<60 (%)	Class 1 <sup>b</sup> (ton ha <sup>-1</sup> ; %)	Yield (kg tree <sup>-1</sup> )	FN	<60 (%)	Class 1 (ton ha <sup>-1</sup> ; %)	Yield (kg tree <sup>-1</sup> )	FN	<60 (%)	Class 1 (ton ha <sup>-1</sup> ; %)
<b>2005</b>												
Control	4.2 a <sup>c</sup>	31 a	12 a	1.7 (22) a	8.1	43	6.5 a	9.2 (60)	8.7 a	51 a	3	15.9 (96) a
PSi	7.3 ab	52 ab	11 a	3.5 (25) ab	8.1	42	3.0 ab	10.0 (65)	11.2 bc	64 abc	3	18.0 (95) a
PB	8.3 b	52 ab	2 b	8.0 (51) b	8.0	34	0.6 b	11.5 (76)	10.3 ab	60 ab	2	19.0 (97) a
WS	12.7 c	76 c	3 b	13.9 (59) c	10.2	53	2.0 b	15.3 (79)	13.3 c	79 c	3	24.3 (96) b
LS	11.7 c	68 bc	3 b	17.3 (78) c	9.2	42	1.0 b	15.6 (89)	12.8 c	70 bc	3	23.1 (95) b
WSCu	11.0 bc	69 bc	3 b	18.2 (87) c	9.7	43	0.5 b	17.7 (93)	11.5 bc	63 abc	5	20.8 (95) ab
<i>F</i> -test	*	*	**	***	ns	ns	**	ns	*	*	ns	*
<b>2006</b>												
Control	5.4 a	48 a	63 a	0.2 (02) a	13.8 a	90	10	17.6 (67) a	15.4 a	97 a	8	26.6 (91) a
PB	11.5 b	89 b	52 a	5.5 (25) b	16.2 ab	94	3	25.5 (83) b	15.4 a	84 a	5	27.4 (93) a
WS	12.4 b	123 bc	51 a	6.6 (28) b	17.7 abc	106	9	28.2 (84) b	19.5 ab	116 ab	8	34.1 (92) ab
LSd	19.4 c	154 cd	31 b	19.2 (52) c	19.2 bc	119	3	34.3 (94) c	23.6 bc	137 b	7	41.3 (92) bc
LS	23.1 d	185 d	31 b	28.1 (64) d	19.5 bc	116	6	35.2 (95) c	23.7 bc	148 b	5	42.3 (94) bc
WSCu	24.8 d	196 d	22 b	35.3 (75) d	21.2 c	124	3	38.7 (96) c	25.6 c	144 b	6	45.2 (93) c
<i>F</i> -test	***	***	**	***	**	ns	ns	***	**	**	ns	**

\* $P \leq 0.05$ \*\* $P \leq 0.01$ \*\*\* $P \leq 0.001$ <sup>a</sup>For an explanation of treatments and dosages see Table 2<sup>b</sup>Class 1 includes fruits with scab severity <1%, size >60 mm and russet <10%. The calculation base for yield (ton ha<sup>-1</sup>) was 1,900 trees ha<sup>-1</sup>. Values in brackets are the proportion of class 1 compared with total yield<sup>c</sup>Values within columns with different letters differ significantly according to the Student–Neuroman–Keuls multiple range tests ( $P \leq 0.05$ ). *F*-test = ns (non-significant) at  $P > 0.05$ 

infection process (before infection), on germinating spores possibly already with appressoria; however not yet with the formation of the primary stroma which allows the fungus to be protected by the plant cuticle.

The poorer result registered with the delayed spray programme (LSd) showed that the timing of treatment application must be close to the onset of infection, even with lime sulphur.

The 'during-infection' spray strategy has several important advantages compared with the preventive (before rainfall) spray strategy; these include (1) less washing effect, (2) greater treatment effectiveness, (3) avoidance of unnecessary treatments (Funt et al. 1990; Holb et al. 2003) and (4) in hot seasons, applying treatments during less sunny periods. However, there are potential problems with this strategy, such as spraying in windy weather being unsuitable for 'low-volume' spraying, delaying a spray during an

extended rainy period and spray-timing within a few hours after the onset of the rain.

The low rate of copper (0.16%) added to the wettable sulphur treatments before flowering gave far better scab control than sulphur alone. This suggests that dilute copper could be very effective under cold weather conditions in early primary scab infection periods. In both 2005 and 2006, the lime sulphur treatment was slightly less effective than the combined wettable sulphur and copper treatment. However, lime sulphur was more effective than wettable sulphur used alone, confirming the results of previous studies (Mills 1947; Ellis et al. 1994; Holb et al. 2003).

The efficiency of bicarbonate salts in controlling apple scab, as reported here and in previous studies (Schulze and Schönherr 2003; Ilhan et al. 2006; Jamar and Lateur 2007; Jamar et al. 2007), together with the

**Table 5** Effects of treatments on adult *Typhlodromus pyri* density in May, June and July 2005 and 2006

Treatments <sup>a</sup>	Number of <i>T. pyri</i> per 100 leaves <sup>c</sup>					
	2005			2006		
	May	June	July	May	June	July
Control	44	83 a <sup>d</sup>	70	51 a	56 a	76
PB	48	69 b	64	37 b	38 b	66
PSi or LSd <sup>b</sup>	42	70 b	66	32 b	31 b	71
WS	40	43 cd	55	33 b	33 b	64
LS	47	55 c	63	28 b	22 b	69
WSCu	46	39 d	59	31 b	30 b	61
<i>F</i> -test	ns	***	ns	*	**	ns

\* $P \leq 0.05$ \*\* $P \leq 0.01$ \*\*\* $P \leq 0.001$ <sup>a</sup> For an explanation of treatments and dosages see Table 2<sup>b</sup> PSi in 2005 and LSd in 2006<sup>c</sup> Values are the means of six replicates, each replicate including 18 leaves (six leaves per cultivar)<sup>d</sup> Values within columns with different letters differ significantly according the Student–Neuroman–Keuls multiple range tests ( $P \leq 0.05$ ). *F*-test = ns (non-significant) at  $P > 0.05$ 

improvement of other disease-control treatments using bicarbonate salts (Horst et al. 1992), suggest that this compound could be introduced in apple disease management. The fact that the compounds are ubiquitous in nature, naturally present in human food, available to the general public for non-pesticide uses and required for normal functions in human, animal, plant and environmental systems imply that this simple compound is appropriate for organic production systems. However, potassium bicarbonate acts as a contact fungicide and is not likely to be systemic or curative. Greenhouse experiments have shown that 1% Armicarb is 95% effective in controlling apple scab, but a long-lasting effect cannot be expected (Jamar et al. 2007). Bicarbonates are quickly converted into ineffective compounds and are highly water-soluble, and they will be washed off the leaves by a small amount of precipitation. They therefore require frequent spray applications well-targeted in the infection risk periods. So far, no data are available on the effectiveness of potassium bicarbonate under low temperature conditions. Activity below 10°C is a prerequisite if copper is to be replaced. Our results indicated that applications of Armicarb alone during the growing season were not effective enough against scab and suggested that it must be supplemented. In our experimental conditions, silicon had a very poor

effect on apple scab. However, Belanger et al. (1995) reported that there is cumulative evidence that increased silicon absorption offers protection against various fungal diseases.

The impact of the disease was stronger in 2006 than in 2005 because (1) weather conditions were more favourable for scab infections in 2006 and (2) several untreated and poorly treated plots in 2005 led to heavy disease pressure during the 2006 primary infection season. Since several sanitation practices were reported to reduce the potential ascospore dose (Holb 2006), autumn leaf-shredding and early spring leaf-burying were carried out between the two growing seasons in order to limit the influence of the previous year. In addition, the risk of early scab epidemics initiated by over-wintered conidia is high in organic orchards (Holb et al. 2005a) and this could explain the relatively lower effectiveness of wettable sulphur and potassium bicarbonate used alone in the primary 2006 scab season compared with 2005. That means that the results registered in 2006 were probably influenced by treatments applied in the previous year. Some of the scab damage observed on harvested fruits possibly arose from secondary scab infection, as only two summer sprays were applied, especially in plots where primary scab control was partial (MacHardy 1996; Holb et al. 2005b).

Although some authors have reported poorer leaf appearance with sulphur and copper treatments (Palmer et al. 2003; Holb et al. 2003), the amount of active substances used in our study to control apple scab did not induce any phytotoxic effects, plant damage or yield fall. However, in our experiment the use of copper was avoided during and after flowering, the treatments were applied with a tunnel sprayer at a low rate of about 300 l ha<sup>-1</sup> and the treatment frequencies and fungicide doses were limited, particularly during flowering.

On the low scab-susceptible cv. 'Reinette des Capucins', sprays based on sulphur caused a significant increase in yield per tree (Table 4). Such positive effects of sulphur compounds on yields cannot be explained by the control of apple scab or other apple diseases such as powdery mildew (*Podosphaera leucotricha*) because the infection levels on untreated plots were very low, including at a later stage, in both years. These results contrast with earlier studies showing that sulphur applications reduced yield and fruit numbers (Mills 1947; Holb et al. 2003; Palmer et al. 2003). The application of elemental sulphur to crops is increasingly advocated as a way of overcoming deficiency in this key nutrient, and sulphur deficiency has recently become a widespread nutrient disorder in crops, largely due to restrictions on fossil fuel burning (Schnug 1998; Williams and Cooper 2004). A chemical analysis of leaves from cv. 'Reinette des Capucins' collected on 25 June 2006, previously washed with acid solutions, showed that the leaf dry extracts from the WS, LS, LSd and WSCu treatments contained 0.38% of sulphur while the leaf dry extracts from the PB and control treatments contained 0.30% of sulphur ( $P < 0.001$ ).

The absence of the phytophagous mite *P. ulmi* and the very low density of *A. schlechtendali* during the two growing seasons might be associated with the very high density of the predator *T. pyri* observed throughout the orchard in both years. The slight and temporary reduction of *T. pyri* on treated plots in June might be correlated with periods with higher treatment frequencies. The treatments might have had harmful effects on *T. pyri*, but the reduction of *T. pyri* during these periods of treatments might also be due to the decrease of prey availability. Sulphur compounds can have harmful effects on phytoseiids (Kreiter et al. 1998). However, other studies have

reported predator mite population tolerance of treatments with sulphur compounds, probably due to the development of tolerant strains with an acquired resistance to sulphur (Markoyiannaki-Printzioui et al. 2000). An earlier study (Beresford et al. 1996) showed that bicarbonate salts did not reduce predator mite numbers or disrupt biological mite control.

In this study it was clearly shown that (1) the 'during-infection' spray strategy using reduced amounts of either lime sulphur or wettable sulphur combined with copper was very effective against primary scab infections; (2) copper and lime sulphur were efficient when temperatures were below 10°C; (3) lime sulphur was more effective than wettable sulphur; (4) lime sulphur effectiveness against apple scab decreased if the treatments were applied 12–24 h later than in the 'during-infection' spray strategy; (5) potassium bicarbonate was effective against apple scab and as effective as wettable sulphur; (6) the sulphur-based treatments increased yield even with a low scab-susceptible cultivar; (7) inorganic fungicide doses and frequencies used to reduce apple scab severity on fruits by about 98% were not phytotoxic, did not adversely affect yield and did not affect summer *T. pyri* density; and (8) the amount of copper for scab control could be reduced on medium and low scab-susceptible cultivars compared with high scab-susceptible cultivars.

As lime sulphur is not allowed in Belgium, copper is still needed for apple scab control under apple organic production in the country. Currently, lime sulphur appears to be the sole remaining option for replacing copper when temperatures are below 10°C in organic farming, and therefore scab management in Belgium would be compromised if there were new European or national regulations restricting the use of copper. The present study has demonstrated the potential of controlling apple scab with reduced and non-damaging amounts of inorganic fungicides using accurate timing of treatments and a spray machine.

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## References

- Anon. (2007). Council Regulation (EEC) No 2092/91 of 24 June 1991 on organic production of agricultural products and indications referring thereto on agricultural products and foodstuffs. Retrieved September 30, 2007 from European Union Web site: <http://europa.eu/scadplus/leg/en/lvb/l21118.htm>
- Belanger, R. R., Bowen, P. A., Ehret, D., & Menzies, J. G. (1995). Soluble silicon: Its role in crop and disease management of greenhouse crops. *Plant Disease*, 79, 329–336.
- Beresford, R. M., Wearing, C. H., Marshall, R. R., Shaw, P. W., Spink, M., & Wood P. N. (1996). Slaked lime, baking soda and mineral oil for black spot and powdery mildew control in apples. Proceedings of the 49th New Zealand Plant Protection Conference. Quality Hotel Rutherford, Nelson, New Zealand, pp. 106–113, August.
- Croxall, H. E., Gwynne, D. C., & Jenkins, J. E. E. (1952). The rapid assessment of apple scab on fruit. *Plant Pathology*, 2, 89–92.
- Ellis, M. A., Ferree, D. C., Funt, R. C., & Madden, L. V. (1998). Effects of an apple scab-resistant cultivar on use patterns of inorganic and organic fungicides and economics of disease control. *Plant Disease*, 82, 428–433.
- Ellis, M. A., Madden, L. V., Wilson, L. L., & Ferree, D. C. (1994). Evaluations of organic and conventional fungicide programs for control of apple scab in Ohio. *Ohio Agricultural Research Development Centre, Research Circular*, 298, 63–68.
- Funt, R. C., Ellis, M. A., & Madden, L. V. (1990). Economic analysis of protectant and disease-forecast-based fungicide spray programs for control of apple scab and grape black rot in Ohio. *Plant Disease*, 74, 638–642.
- Gessler, C., Patocchi, A., Sansavini, S., Tartarini, S., & Gianfranceschi, L. (2006). *Venturia inaequalis* resistance in apple. *Critical Reviews in Plant Sciences*, 25, 473–503.
- Hindorf, H., Rövekamp, I. F., & Henseler, K. (2000). Decision aids for apple scab warning services (*Venturia inaequalis*) in Germany. *OEPP/EPPO Bulletin*, 30, 59–64.
- Holb, I. J., & Heijne, B. (2001). Evaluating primary scab control in organic apple production. *Gartenbauwissenschaft*, 66, 254–261.
- Holb, I. J., De Jong, P. F., & Heijne, B. (2003). Efficacy and phytotoxicity of lime sulphur in organic apple production. *Annals of Applied Biology*, 142, 225–233.
- Holb, I. J., Heijne, B., & Jeger, M. J. (2005a). The widespread occurrence of overwintered conidial inoculum of *Venturia inaequalis* on shoots and buds in organic and integrated apple orchards across the Netherlands. *European Journal of Plant Pathology*, 111, 157–168.
- Holb, I. J., Heijne, B., Withagen, J. C. M., Gall, J. M., & Jeger, M. J. (2005b). Analysis of summer epidemic progress of apple scab at different apple production systems in the Netherlands and Hungary. *Phytopathology*, 95, 1001–1020.
- Holb, I. J. (2006). Effect of six sanitation treatments on leaf litter density, ascospore production of *Venturia inaequalis* and scab incidence in integrated and organic apple orchards. *European Journal of Plant Pathology*, 115, 293–307.
- Horst, R. K., Kawamoto, S. O., & Porter, L. L. (1992). Effect of sodium bicarbonate and oils on the control of powdery mildew and black spot of roses. *Plant Disease*, 76, 247–251.
- Ilhan, K., Arslan, U., & Karabulut, O. A. (2006). The effect of sodium bicarbonate alone or in combination with a reduced dose of tebuconazole on the control of apple scab. *Crop Protection*, 25, 963–967.
- Jamar, L., & Lateur, M. (2007). Strategies to reduce copper use in organic apple production, ISHS. *Acta Horticulturae*, 737, 113–120.
- Jamar, L., Lefrancq, B., & Lateur, M. (2007). Control of apple scab (*Venturia inaequalis*) with bicarbonate salts under controlled environment. *Journal of Plant Diseases and Protection*, 115, 221–227.
- Kreiter, S., Sentenac, G., Barthes, D., & Auger, P. (1998). Toxicity of four fungicides to the predacious mite *Typhlodromus pyri* (Acari: Phytoseiidae). *Journal of Economic Entomology*, 91, 802–811.
- Lateur, M., Wagemans, C., & Populer, C. (1999). Evaluation of fruit tree genetic resources as sources of polygenic scab resistance in apple breeding. ISHS. *Acta Horticulturae*, 484, 35–42.
- Lateur, M. & Blazek, J. (2002, May). Evaluation descriptors for *Malus*. Report of a Working Group on *Malus/Pyrus*. Second meeting, Dresden-Pillnitz, Germany. International Plant Genetic Resources Institute, Rome, Italy, pp. 76–82.
- MacHardy, W. E., & Gadoury, D. M. (1989). A revision of Mills's criteria for predicting apple scab infection periods. *Phytopathology*, 79, 304–310.
- MacHardy, W. E. (1996). *Apple scab, biology, epidemiology and management*. St Paul, Minnesota, USA: APS Press.
- MacHardy, W., Gadoury, D. M., & Gessler, C. (2001). Parasitic and biological fitness of *Venturia inaequalis*: Relationship to disease management strategies. *Plant Disease*, 85, 1036–1051.
- Markoyiannaki-Printzioui, D., Papaioannou-Souliotis, P., Zeginis, G., & Giatropoulos, C. (2000). Observations on acarofauna in four apple orchards of Central Greece. I. Incidence of pedoclimatic conditions and agricultural techniques on phytoseiid mites (Acari: Phytoseiidae). *Acarologia*, 41, 109–126.
- Mills, W. D. (1947). Effects of sprays of lime sulphur and of elemental sulphur on apple in relation to yield. *Cornell Experiment Station*, 273, 38 p.
- Montag, J., Schreiber, L., & Schönherr, J. (2005). An in vitro study on the postinfection activities of hydrated lime and lime sulphur against apple scab (*Venturia inaequalis*). *Journal of Phytopathology*, 153, 485–491.
- Palmer, J. W., Davies, S. B., Shaw, P. W., & Wünsche, J. N. (2003). Growth and fruit quality of 'Braeburn' apple (*Malus domestica*) trees as influenced by fungicide programmes suitable for organic production. *New Zealand Journal of Crop and Horticultural Science*, 31, 169–177.
- Schnug, E. (1998). *Sulphur in agroecosystems*. Dordrecht: Kluwer.
- Schulze, K., & Schönherr, J. (2003). Calcium hydroxide, potassium carbonate and alkyl polyglycosides prevent spore germination and kill germ tubes of apple scab (*Venturia inaequalis*). *Journal of Plant Diseases and Protection*, 110, 36–45.

- Simon, S., Lauri, P. E., Brun, L., Defrance, H., & Sauphanor, B. (2006). Does manipulation of fruit-tree architecture affect the development of pests and pathogens? A case study in an organic apple orchard. *Journal of Horticultural Science & Biotechnology*, 81, 765–773.
- Smereka, K. J., MacHardy, W. E., & Kausch, A. P. (1987). Cellular differentiation in *Venturia inaequalis* ascospores during germination and penetration of apple leaves. *Canadian Journal of Botany*, 65, 2549–2561.
- Trapman, M., & Polfliet, C. M. (1997). Management of primary infections of apple scab with the simulation program RIMpro: Review of four years field trials. *IOBC Bulletin*, 20, 241–250.
- Tweedy, B. G. (1981). Inorganic sulphur as a fungicide. *Residue Reviews*, 78, 44–68.
- Williams, J. S., & Cooper, R. M. (2004). The oldest fungicide and newest phytoalexin—A reappraisal of the fungitoxicity of elemental sulphur. *Plant Pathology*, 53, 263–279.