

Full Length Research Paper

Effects of concentration, frequency of application and runoff of foliar-applied soluble silicon on powdery mildew of zucchini

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The effect of concentration, frequency of application and runoff of foliar-applied soluble silicon (Si) on the severity of powdery mildew (PM) of zucchini (*Cucurbita pepo* L.) was evaluated. Soluble Si (250 to 1000 mgL⁻¹), together with Break-Thru[®] (0.25 ml/L), was sprayed onto zucchini plants at frequencies of 1 to 3 times per week. The leaves of all plants were inoculated with a known concentration of conidia of *Podosphaera xanthii* 2 days after the sprays were applied. The effect of runoff was determined by covering some of the pots with polyethylene sheets, while others were left uncovered. Spraying Si onto zucchini plants reduced the severity of PM significantly. The efficacy of Si was improved by increasing the spray frequency. The best results were obtained when the frequency of the treatment was increased, and when Si was allowed to reach the root zone of the plants. When Si was applied onto leaves, direct contact between the spray and the pathogen seemed to be the main mechanism of action involved in disease control, and part of the spray (that is, the runoff) was absorbed by the plant roots, and subsequently played an important role in the health of the plants.

Key words: *Podosphaera xanthii*, powdery mildew, soluble silicon, zucchini.

INTRODUCTION

Powdery mildew (PM) of many crops causes significant losses in yield if not managed properly (Romero et al., 2003, 2004). Soluble silicon (Si) has been investigated by many authors (Epstein, 1994, 1999; Bélanger et al., 1995, 1997; Liang et al., 2005) as an environmentally safe option for PM management.

Foliar applications of Si have shown promising results in controlling PM of several crops (Bowen et al., 1992; Menzies et al., 1992; Reynolds et al., 1996; Liang et al., 2005; Palmer et al., 2006; Guével et al., 2007). They demonstrated possibility of replacing or supplementing the fungicides treatments against PM disease with soluble Si sprays. The efficacy of Si can also be

improved by increasing the frequency of application as this increases the deposition of the element on the phylloplane. The level of disease control that can be achieved using this approach is, however, limited due to a lack of information on the optimum concentration.

Furthermore, when a foliar spray is used, there is always runoff, which is intercepted by the soil. If the Si treatment is absorbed by the roots, as a quasi-essential element, it may play a role in the health of the plant. Therefore, to assess the direct effect of foliar application of Si on PM, it is important to differentiate the impact of runoff from the total effect of the treatment. Understanding the effect of such components can determine whether foliar applications of Si should be more or less effective as a soil drench.

The objectives of this study were to determine the effects of Si as a foliar application on the severity of PM, to determine the optimum concentration and application

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Table 1. Analysis of variance showing factorial interactions between concentration, frequency and runoff of foliar-applied silicon on severity of powdery mildew of zucchini.

Source of variation	Trial 1		Trial 2	
	F-value	P-value	F-value	P-value
Concentration	1.03	0.389	0.84	0.481
Frequency	32.56	< 0.001***	25.89	< 0.001***
Runoff	21.86	< 0.001***	5.19	0.027*
Concentration*Frequency	2.26	0.053	3.54	0.006**
Concentration * Runoff	0.33	0.805	1.84	0.153
Frequency*Runoff	0.13	0.877	0.48	0.621
Concentration * Frequency * Runoff	0.90	0.503	3.06	0.013*
CV (%)		18.4		24.42

Values with * = significant at $p < 0.05$; ** = significant at $p < 0.01$; *** = significant at $p < 0.001$.

frequencies of Si on controlling PM disease, and study the impact of runoff spray on the efficacy of foliar sprays of soluble Si against PM disease.

MATERIALS AND METHODS

Preparation of plants

Seeds of zucchini (*Cucurbita pepo*, F1-Hybrid Partenon) were obtained from Starke Ayres Seeds (Pty) Ltd., planted in seedling trays filled with composted pine bark and kept in a greenhouse at a temperature of $26 \pm 2^\circ\text{C}$ and relative humidity of 75 to 85%. Trays were irrigated with a NPK mixed fertilizer [NPK 3:1:3 at 0.5 gL^{-1}] + $[\text{Ca}(\text{NO}_3)_2$ at $0.5 \text{ gL}^{-1}]$ for 2 weeks using overhead irrigation. Immediately the germinated seedlings fully developed a second leaf, they were transplanted into pots filled with composted pine bark and transferred to another greenhouse operating at similar environmental conditions. The plants were irrigated with the same nutrient solution using drip irrigation until the end of the experiment.

Preparation of *Podosphaera xanthii* for inoculation

Leaves of zucchini plants, naturally infected with PM fungus were collected and the causal organism was identified as *P. xanthii* (Castagne) according to McGrath and Thomas (1996). Healthy plants of zucchini were inoculated by dusting the leaves with *P. xanthii* conidia and kept in a separate greenhouse to be used as source of inoculum. After three weeks of inoculation of these plants, conidia of *P. xanthii* were harvested using previous techniques (Askary et al., 1998; Dik et al., 1998). To ensure a high level of conidial viability and to maintain the same age of inoculum, old conidia were removed by shaking the source leaves 24 h before inoculation. Infected leaves were then immersed in sterilized distilled water and shaken to remove the conidia. Conidia were counted using a haemocytometer and the concentration was adjusted to 10^3 conidia mL^{-1} . Finally, seedlings were inoculated by spraying 3 to 5 mL of conidial suspensions of *P. xanthii* onto the leaves of each seedling using a hand sprayer within two hours of counting. Inoculation was done 2 days after Si was sprayed onto plants.

Application of soluble silicon

As a source of soluble Si, potassium silicate (K_2SiO_3) (product name K2550 containing 20.5 to 20.9% SiO_2 and 8.0 to 8.15% K_2O)

was supplied by PQ Silicas (SA). Concentrations of Si were calculated according to the product's information and sprayed onto zucchini plants at five different levels (0, 250, 500, 750 and 1000 mg L^{-1}) until runoff. Treatments were applied at a frequency of 1 to 3 times per week. To prevent the impact of the runoff of Si spray on the disease severity, the lower part of the plants and their pots were covered by polyethylene sheets to play as a waterproof barrier to stop any Si spray from reaching the root zone of the plants. The aerial parts of the plants were left open for spray. Other pots were left uncovered, allowing the drift and runoff of spray applications to reach to the rhizosphere.

For all treatments, 0.25 ml/L Break-Thru® (BK) was used as a surfactant. Distilled water containing only BK was sprayed at the same frequencies as the control treatment. All the treatments were applied in the late afternoon in order to minimize dryness of the leaves due to heat. Disease severity, calculated as percentage of leaf area infected, was evaluated for all the treatments 3 weeks after inoculation.

Disease assessment and statistical analysis

The experiment was conducted twice, with each treatment having three replications. The treatments were arranged in a factorial randomized complete block design. Analysis of Variance (ANOVA) was performed using a factorial treatment structure. Where the CV% was $> 20\%$, the original data were transformed using a square root transformation. Interactive effects of application concentration, frequency and runoff of Si on the severity of PM was analysed using GenStat® Statistical Analysis Software (GenStat, 2006). Treatment means were separated using Fisher's protected least significant difference (LSD). The percentage of disease reduction by each treatment was calculated based on the disease severity of the Untreated Control as a reference. Finally, polynomial regression analysis was performed on the percentage of disease reduced by different levels of Si applied on uncovered and covered pots at 3 different frequencies.

RESULTS

In both experiments, application of Si reduced the disease severity significantly. The effect of concentration on the efficacy of soluble Si sprays was not significant (Table 1). After spraying the plants with Si at different tested concentrations (250 to 1000 mgL^{-1}) for 3 weeks, the disease severity was reduced by 42 to 61% and 75 to

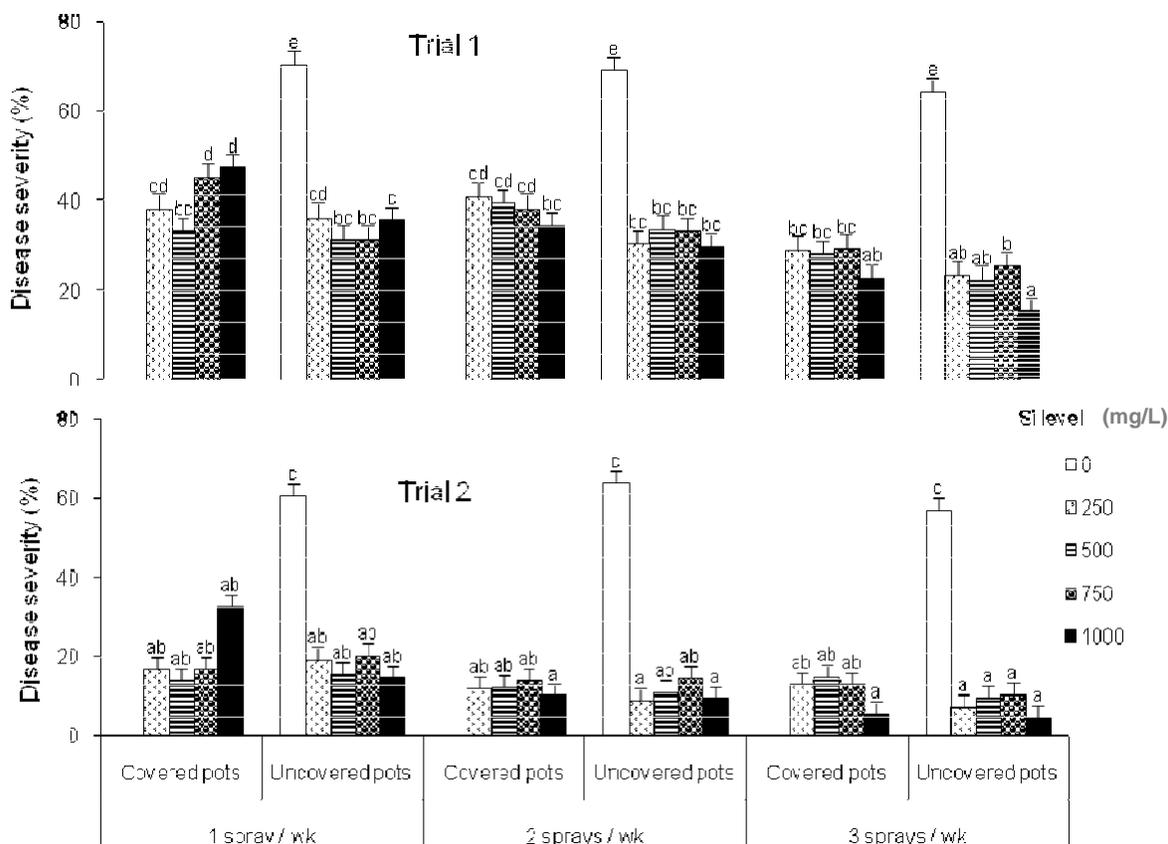


Figure 1. Histograms showing the effects of spraying Si on the severity of powdery mildew of zucchini, when applied at various concentration and frequencies on plants grown under greenhouse conditions in uncovered and covered pots in two different trials.

79% in the first and second trials, respectively. In both experiments, no significant difference was found between the different tested concentrations of Si on the severity of the disease.

In both trials, spray frequency had a significant effect on the efficacy of Si in controlling PM. Using the same concentration of Si, efficacy of the treatment was increased initially by 30% and almost doubled in the subsequent experiment when the spray frequency was tripled. Even in the control treatment, which contained only BK, the disease level was reduced when the treatment was sprayed at higher frequency (Figures 1 and 2).

The comparison between uncovered and covered pots on PM severity showed that drift and runoff of foliar-applied Si had a significant impact on disease control (Table 1). In both trials, efficacy was improved significantly when Si was sprayed onto plants in uncovered pots. An overall increase of 17% (Trial 2) and 18% (Trial 1) in disease reduction occurred on plants in uncovered pots, where Si was allowed to reach the rhizosphere, compared to the covered pots where the spray was restricted to the phylloplane (Figure 2).

In both trials, the interaction of Si concentration and

frequency was significant. Interactive effects of concentration and frequency on the efficacy of Si was significant in the second experiment, but not in the first experiment. However, trends of these two experiments showed that the best results were obtained when Si was used at higher concentrations with higher frequencies (that is, 3 times per week). For instance, when Si was applied 3 times a week at 1000 mgL^{-1} , then its efficacy was increased by more than two - three times (Figures 1 and 2).

Interactive effects of Si concentration and application method were not significant on the efficacy of Si. However, improved efficacy was obtained when treatments with higher levels of Si were sprayed on plants in uncovered pots. Interaction of frequency and runoff on the efficacy of Si was not significant. In both trials, efficacy of Si was improved when sprayed onto plants in uncovered pots at higher frequencies. Increasing the frequency from 1 to 3 times per week increased the efficacy of the treatment by a mean of 35% in the first experiment and nearly 100% in the second experiment. Trends of the two trials also showed that the increase in efficacy was slightly higher when sprays were applied to plants in the uncovered pots.

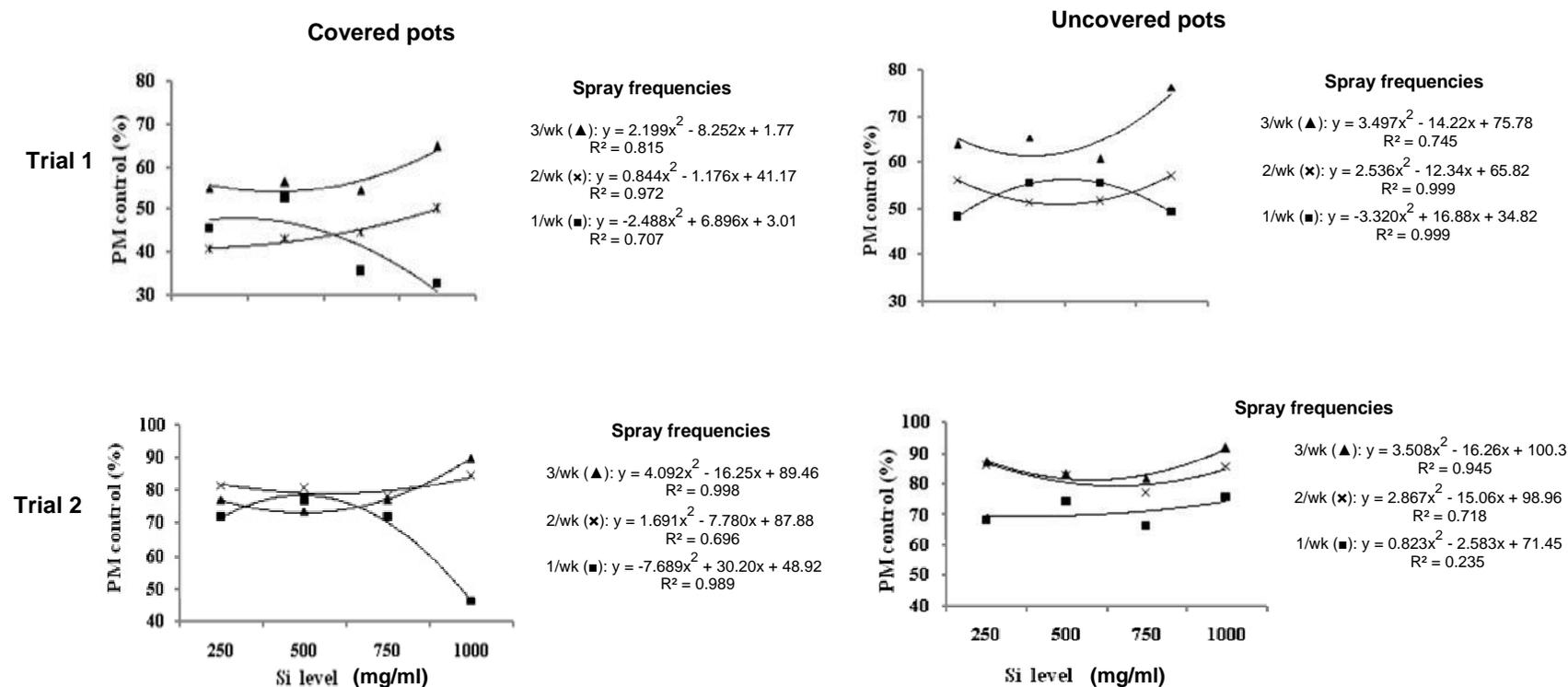


Figure 2. Relationship between Si concentrations and frequency of sprays applied in uncovered or covered pots on powdery mildew control of zucchini plants grown under greenhouse conditions in two different trials.

Interaction of concentration, frequencies and application method was only significant on the second experiment. However, individual observations showed that the best results were obtained when Si was sprayed at higher frequencies to plants in uncovered pots (Figures 1 and 2).

DISCUSSION

In this study, spraying soluble Si onto zucchini

leaves reduced the severity of PM significantly. This findings confirmed a previous research on the use of Si sprays for the control of PM of cucumber (Menzies et al., 1992; Liang et al., 2005), grape (Bowen et al., 1992; Reynolds et al., 1996), muskmelon (Menzies et al., 1992), strawberry (Kanto et al., 2004; Palmer et al., 2006), wheat (Guével et al., 2007) and zucchini (Menzies et al., 1992).

In most cases, foliar applications of Si provides less disease control compared to when the element is fed to the plant in a nutrient solution or

as a soil amendment (Liang et al., 2005; Guével et al., 2007). However, in some plants, foliar application provides better efficacy than supplying the element to the roots. For instance, Bowen et al. (1992) found that spraying Si at 1000 mgL^{-1} onto grape leaves reduced the severity of PM significantly; whereas, treating the plant with a Si-amended solution did not. This is because some crops are unable to transport Si from their root to shoots through their vascular system (Bélanger et al., 1995).

The mode of action of Si in reducing severity of

PM when applied as a foliar treatment may be different from when it is fed through the roots (Wang and Galletta, 1998). Spraying Si on cucumber can result in the formation of a coating (film) on the leaves (Menzies et al., 1992), which would act as a physical barrier, preventing the penetration of fungal hyphae into the host (Bowen et al., 1992). Guével et al. (2007) showed that foliar application of Si has a direct effect on PM over and above any effects mediated by the plant, this was as a result of root amendments, which may lead to induced resistance. A direct action by Si sprays on PM was confirmed by our results when Si was sprayed onto plants where the element was restricted from reaching the roots of the plants. When KSi is used against PM, the active ingredient of the spray appears to be Si (Menzies et al., 1992).

Results of this study showed that in the range of 250 to 1000 mgL⁻¹, the level of Si in the spray solution had little impact on the efficacy of Si in PM control. Severity of PM was significantly reduced by all treatments, indicating that application of Si at lower concentrations could provide the optimum disease control, while minimizing the cost of control. Observations by Menzies et al. (1992) on the use of Si against PM of different hosts showed that the efficacy of a foliar spray of Si at 1000 mgL⁻¹ was equivalent to a solution of 100 mgL⁻¹ applied as a soil amendment.

In this study, the efficacy of Si was improved by increasing the spray frequency. When the application frequency was increased from 1 to 3 times per week, efficacy of Si was doubled. This agrees with the report of Reynolds et al. (1996), who increased the efficacy of Si sprays against PM of grape (*Uncinula necator* (Schwein.) Burrill) by increasing the spray frequency. Increasing application frequencies could increase the total amount of Si deposited on the surface of the leaf, resulting in better efficacy. Spraying Si at a concentration of 250 mgL⁻¹ with a frequency of 3 times per week could be expected to result in a lower amount of Si being deposited on the leaf than if it was applied once a week at a concentration of 1000 mgL⁻¹. However, this study showed that spraying Si at 250 mgL⁻¹ x 3 wk⁻¹ was more effective than spraying 1000 mgL⁻¹ x 1 wk⁻¹. Even for the adjacent treatment, which did not contain Si, disease severity was reduced by increasing the frequency of spray treatment. Therefore, when spray frequency is increased, there must be other factors that are involved in improving the efficacy of Si other than the total amount of the element deposited on the phylloplane. The wetting agent (BK) and increased leaf wetness could have their own impact on the pathogen. Break-Thru[®] was shown to have a direct effect on PM restricting expansion of its colonies and collapsing conidia and hyphae of the pathogen (Tefagiorgis, 2008, unpublished data). Although extended periods of leaf wetness can favour infection of zucchini by several foliar pathogens, it has a negative effect on the development of PM by inhibiting germination of the conidia (Sakurai and

Hirata, 1959; Bushnell and Rowell, 1967). Most importantly, increasing the frequency of applications could improve the continuity of Si supply to plants roots through runoff. Even though the major factors remain unknown, this study demonstrated that better disease control could be achieved by spraying Si at lower concentrations with increased spray frequencies instead of applying higher concentrations of Si at lower frequencies.

When the same concentration of Si was sprayed at the same frequencies per week, the severity of PM recorded from plants grown in uncovered pots was less than that of plants grown in covered pots. This was because, in open pots, part of the spray solution was intercepted by the soil as a result of drift and runoff. Once the Si solution was in the rhizosphere of the plant, it could be absorbed by the roots and translocated to different parts of the plant. Reviews by Epstein (1994, 1999, 2001) showed that adding Si into the nutrient solution or adding it to the soil benefits plants by providing protection against pathogens through physical barriers and triggering plant resistance against pathogens, and ameliorating other biotic and abiotic stresses of the plant. However, when KSi is sprayed onto the leaf, it may be deposited on the surface of the leaf without penetrating into the plant (Buck et al., 2008). Therefore, the possible mechanisms of protection it provides might have been through direct contact with the pathogen and alteration of the chemical properties of the leaf such as pH, which could lead to changes in osmotic properties of the leaf surface (Liang et al., 2005). Using scanning electron micrographs, Bowen et al. (1992) observed an active accumulation of Si around the appressoria of the PM fungi when Si was sprayed onto leaves of cucumber plants. However, Reynolds et al. (1996) suggested that exogenously applied silicates act to augment the activity of their endogenous counterparts, resulting in the accumulation of the element around the pathogen.

In most research conducted on the management of PM using foliar-applied Si, the effects of the drift or runoff of soluble Si on the disease has not been considered. However, as seen in this study, such drift and runoff can have a significant impact on the health of the plant and give a confounded conclusion because different modes of action can be involved, once the element is absorbed by the plant via its roots. For instance, foliar application of Si did not reduce severity of PM of strawberry (Palmer et al., 2006), but enhanced growth of the plant by increasing its chlorophyll content (Wang and Galletta, 1998). Therefore, such metabolic changes of the plant might be related to the absorption of Si through the roots as a result of drift and runoff of foliar-applied Si.

In this study, the plants were irrigated using drip irrigation, avoiding any wash-off of Si from leaves to the soil. However, due to the size of the growing area and other technical limitations, most growers prefer overhead irrigation than drip irrigation. In such cases, Si can be

mixed with the nutrient solution and supplied to the plant as part of the irrigation. Using that technique could improve the efficacy and minimize/avoid the costs of labour that would be involved in spraying of the element three times a week to control the disease. This technique may improve the efficacy of the treatment by increasing the spray coverage, and ensuring a better contact between the spray and the pathogen. If proven economical, the foliar use of Si at appropriate application intervals and concentrations could replace fungicides for the management of PM on zucchini - a welcome development for organic farmers.

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