POTENTIAL OF SILICON AMENDMENT TO REDUCE SEVERITY OF FUSARIUM WILT

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Abstract

Results from silicon (Si) trials support prior research that silicon may be important in reducing the severity of fusarium wilt in cotton seedlings. Si amendments were identified that reduced disease severity under glasshouse conditions when applied to field soil, but were not effective under field conditions. It is possible that inoculum load was too high at the field site for an effect of Si fertilization to be realized. Increasing nutrient availability in an alkaline soil also can be difficult and may have contributed to poor uptake. Another consideration is that placement of fertiliser, which dictates availability of Si for uptake, may not have been optimal.

Introduction

Although Si has not been established as an essential nutrient, it is considered an important constituent of plants (Epstein, 1994). Multiple benefits have been observed where Si is available to growing plants, with reported improvements in plant resistance to disease and pathogenic fungal attack where Si has been applied to numerous crops (Epstein, 1994). Mechanisms suggested for the improvement in disease resistance include a mechanical action whereby Si accumulated in the cell walls acts as a barrier to penetrating fungi. Recent research has also suggested that the production and accumulation of antifungal phenolic compounds and the activation of defence related enzymes may also have a role in this plant defence mechanism (Dann and Muir, 2002).

Fusarium wilt of cotton is a destructive disease caused by the soil-inhabiting fungus, *Fusarium oxysporum* f. sp. *vasinfectum* (*Fov*). Once a field is infested with *Fov* there is no commercially viable way to eliminate the pathogen from the soil. To reduce the impact of fusarium wilt an integrated control strategy is necessary. One strategy investigated is the amendment of infested soil with Si to suppress disease development. Results of glasshouse and field experiments are discussed.

Materials and methods

Glasshouse bioassays

Experiment 1

A glasshouse bioassay was conducted to investigate the effect of Si amendment of a soil mix on *Fov* infection. Naturally infested field soil was collected from a cotton farm near Cecil Plains, Qld and mixed 50:50 with sand. *Gossypium hirsutum* cotton cultivar Siokra V17 (mid-range level of resistance to fusarium wilt) was sown directly into the soil mix in seedling flats. All treatments were applied weekly as a soil/seedling drench. Potassium silicate solution was applied at 0.25, 2 and 5 mL/L. Two control treatments included were water only and potassium sulphate (Sigma-

Aldrich) applied at 5 g/L. After 9 weeks plants were harvested and examined internally for disease severity using a rating for vascular discoloration where 0 = no infection, 1 = stem infected below soil level, 2 = infection up to cotyledons, 3 = infection above cotyledons, 4 = infection up to top node and 5 = dead plant..

Experiment 2

A second glasshouse bioassay was conducted to investigate the effect of various forms of Si amendment, including Silvine (magnesium silicate) and acidified wollastonite (acidified calcium silicate), on *Fov* infection. These powder formulations were applied pre-plant and mixed uniformly into the soil prior to sowing with cotton cultivar Siokra V17. Potassium silicate (5 mL/L) was also applied weekly as a soil drench as described in Experiment 1. At harvest, plants were examined internally for disease severity and rated for vascular discolouration as described in Experiment 1.

Field Trials

Trial 1 (2004-2005)

This experiment consisted of four rows, each row containing six treatment plots that were 10 m in length. All plots were separated by buffers of the same dimensions. Treatments applied were 1= Silvine granules (magnesium silicate) 150 kg/ha; 2= Silvine granules 75 kg/ha + Silvine powder 3.5 g/L; 3= potassium silicate powder 150 kg/ha + potassium silicate liquid 10 mL/L + foliar application potassium silicate liquid 2.5 mL/L fortnightly for 8 weeks; 4= As for treatment 3 + potassium silicate liquid foliar application applied monthly until bolls formed; 5= untreated control; 6= potassium sulphate 8 g/L. Silvine granules and potassium silicate powder were applied by hand to a furrow 10 cm deep in the centre of the raised bed. Silvine powder suspended in water, and potassium silicate and potassium sulphate solutions were applied to the furrow using a watering can (1 L/m row). Hills were then reformed. Seeds were sown (cv Sicot F-1 which has a high tolerance to fusarium wilt) and after germination, potassium silicate liquid was applied to foliage using a watering can. The experiment was irrigated and managed commercially. The fifth terminal leaf was collected for nutrient analysis prior to harvest. After harvest, stems were cut and rated for vascular discolouration.

Trial 2 (2005-2006)

This experiment was conducted as described for Trial 1 with some differences outlined here. All Si treatments were applied pre-plant into the furrow. Two varieties were sown, Sicot F-1 and Sicot 189 (more susceptible to fusarium wilt than Sicot F-1). Treatments included Magnesium silicate granules 450 kg/ha; Magnesium silicate granules 150 kg/ha; Potassium silicate powder 150 kg/ha; Acidified calcium silicate powder 150 kg/ha; and an untreated control.

Results

Glasshouse bioassays

Experiment 1

Potassium silicate applied at 5 mL/L and potassium sulphate significantly reduced the mean rating of vascular discolouration (Figure 1).

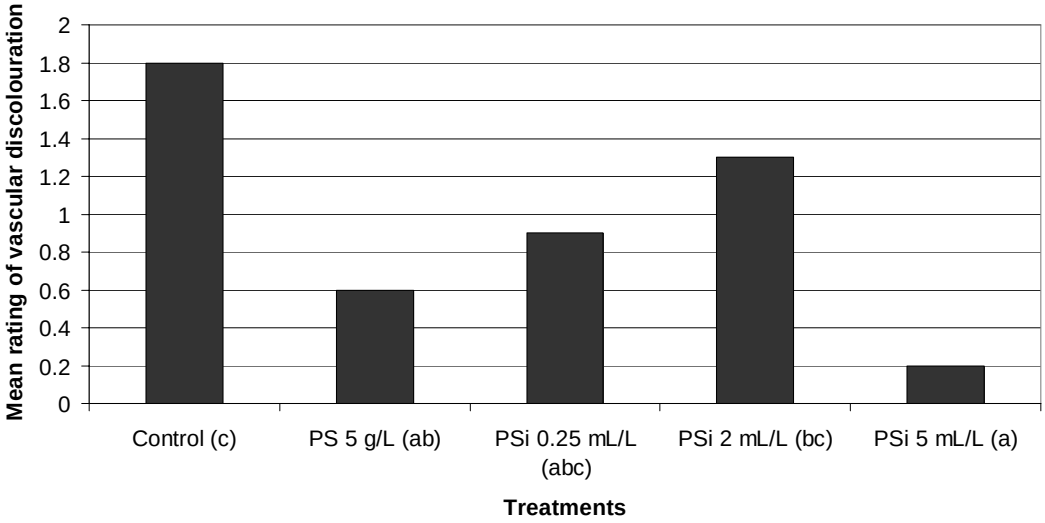

Figure 1. The effect of weekly application of potassium silicate and potassium sulphate solution on mean rating of vascular discolouration when cotton seedlings cv Siokra V17 were grown in soil naturally infested with *Fusarium oxysporum* f. sp. *vasinfectum* mixed 50:50 with sand

Where PS = potassium sulphate (0.45 g K /seedling flat) and PSi = Potassium silicate solution (maximum K applied = 0.1 g/seedling flat).

Rating of vascular discolouration where 0 = no infection, 1 = base of stem infected below soil level, 2 = infection up to node 0, 3 = infection above node 0, 4 = infection up to top node and 5 = dead plant.

Treatments followed by different letters are significantly different from one another (P=0.024) (n=3, 40 seeds per tray).

Statistical analysis

Variances (ANOVA) for replicate and treatment effects were determined using GenStat for Windows Sixth Edition 2002 (Lawes Agricultural Trust, Rothamsted Experimental Station, Herts AL5 2JQ, England).

Figure 2 also describes data from Experiment 1, however it has been displayed differently from that in Figure 1 to show how different treatments effect the distribution or percentage of plants rated 0 to 5 for vascular discolouration. This display allows for a better understanding of how the treatments are affecting disease severity. For the untreated control treatment, approximately 50% of plants displayed no vascular discolouration. When potassium silicate (at 5 mL/L) and potassium sulphate were applied weekly, there was an increase in the percentage of plants that were not diseased, in which approximately 87% of plants were rated 0 for no discolouration for both these treatments. In the control treatment, nearly 20% of plants died (rated 5) however there were no plant deaths when plants were treated with potassium silicate at 5 mL/L or with potassium sulphate.

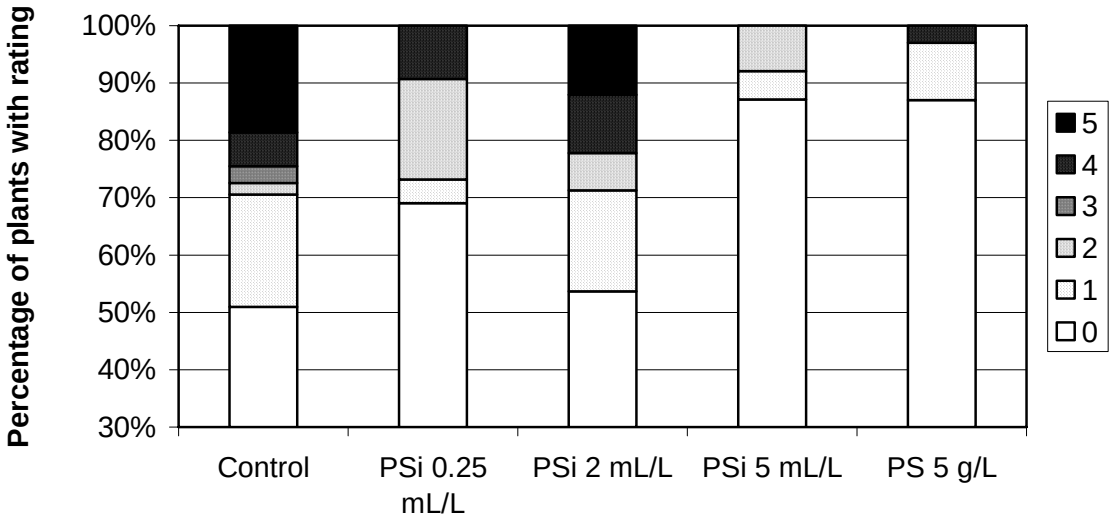

Figure 2. The effect of weekly application of potassium silicate and potassium phosphate on percentage of cotton plants rated 0 to 5 for vascular discolouration when seed was sown into soil naturally infested with *Fusarium oxysporum* f. sp. *vasinfectum* mixed 50:50 with sand.

Where PSi = Potassium silicate solution (maximum K applied = 0.1 g/seedling flat). PS = Potassium sulphate (0.45 g K /seedling flat) n=3, 40 seeds per tray.

Rating of vascular discolouration where 0 = no infection, 1 = base of stem infected below soil level, 2 = infection up to node 0, 3 = infection above node 0, 4 = infection up to top node and 5 = dead plant.

Experiment 2

In addition to potassium silicate, both magnesium silicate and acidified wollastonite, when applied pre-plant and mixed uniformly into the soil mix, significantly reduced disease severity (P=0.027) confirming that disease reduction may be due to Si amendment in addition to potassium (Table 1).

Looking at the distribution of plants rated 0 to 5 (Figure 3) there was a significant increase in the percentage of plants that were rated 0 for no vascular discolouration when Si was applied. There were also no plant deaths in Si treated plants. The percentage of plants rated 0 has significance as this has the potential to translate to an increase in the number of plants surviving to maturity thereby increasing yield.

In these trials, where a significant reduction in disease occurred the pathogen inoculum was not high. The mean rating for discolouration was below 2 meaning that vascular discolouration was below the cotyledons plus 50% or more plants were not showing vascular discolouration.

Table 1. The effect of silicon on infection of *Fusarium oxysporum* f. sp. *vasinfectum* on cultivar Siokra V17 following weekly soil application of Potassium Silicate solution and single pre-plant application of powdered silicon sources mixed uniformly into naturally infested field soil prior to sowing

Treatment	MRVD[A] ± SE
Control	1.64 ± 0.38 a
Silvine (0.7 g/L soil)	0.50 ± 0.32 b
Treated Wollastonite (1 g/L soil)	0.40 ± 0.40 b
Potassium Silicate solution (5 mL/L tap water)	0.15 ± 0.35 b

Means followed by the same letter are not significantly different at the P=0.05 level

[A] Mean of the rating of vascular discolouration where 0 = no infection, 1 = base of stem infected below soil level, 2 = infection up to node 0, 3 = infection above node 0, 4 = infection up to top node and 5 = dead plant.

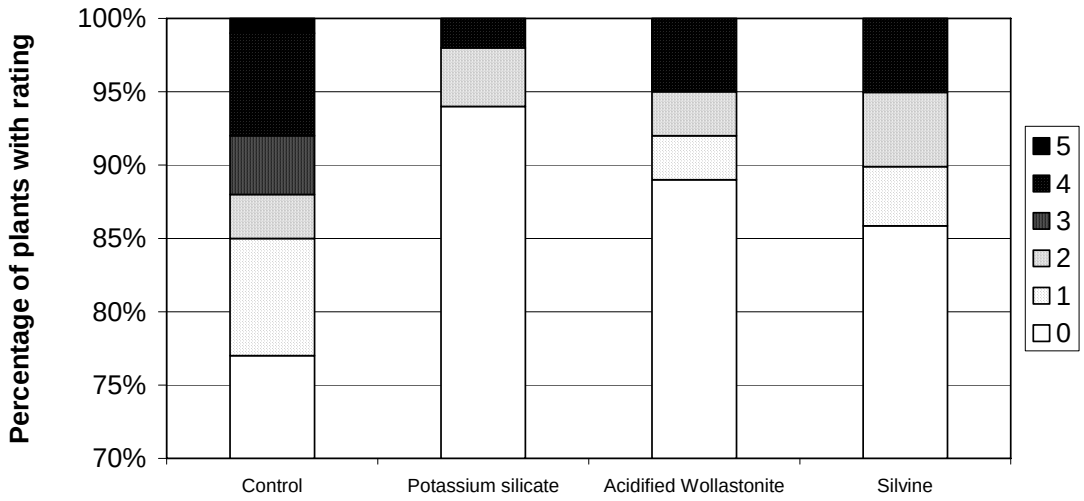

Figure 3. The effect of silicon on infection and disease severity of *Fusarium oxysporum* f. sp. *vasinfectum* on cultivar Siokra V17 following weekly soil application of potassium silicate solution and single pre-plant application of powdered silicon sources mixed uniformly into naturally infested field soil mixed 50:50 with sand prior to sowing.

Rating of vascular discolouration where 0 = no infection, 1 = base of stem infected below soil level, 2 = infection up to node 0, 3 = infection above node 0, 4 = infection up to top node and 5 = dead plant.

Statistical analysis

Data were analysed using a mixed model in REML in GenStat. Random effects were Rep/Plot/Plant, i.e. Rep + Rep*Plot + Rep*Plot*Plant (residual), and fixed effect of Treatment. The full model was fitted initially, then negative and zero variance components removed to arrive at the final random model. Significance of the fixed effect was assessed using a Wald test. If the Wald test indicated a significant Treatment effect, approximate pair-wise comparisons between means were made. Treatment means, their standard errors and differences between them were calculated.

Field Trial Evaluation

Trial 1 (2004-2005)

There was no significant effect of Si treatments on emergence, establishment or 3rd count (data not shown). However, there was considerable variability within each treatment i.e. between replicate plots. Hence, it is very difficult to determine statistically differences among treatments with this level of variability.

Table 2. The effect of silicon based products and potassium sulphate on nutrient content of the 5th terminal leaf, disease severity assessed as % 0 and 1's, and seed cotton yield of cotton plants grown near Cecil Plains in soil naturally infested with *Fusarium oxysporum* f. sp. *vasinfectum*

Treatment[A]	Nutrient analysis of the fifth leaf				% 0 and 1's	Yield
	Si	P	K	Mg		kg/10m plot
	% dm[B]	% dm	% dm	% dm		
1	0.0635 (0.025)	0.040	0.123	0.124	65.9 (0.95)	4.47
2	0.071 (0.027)	0.041	0.124	0.120	72.7 (1.022)	4.29
3	0.0652 (0.026)	0.041	0.132	0.122	67.5 (0.965)	4.13
4	0.066 (0.026)	0.042	0.130	0.120	68.9 (0.995)	4.00
5	0.0385 (0.020)	0.043	0.129	0.121	67.1 (0.96)	3.85
6	0.0362 (0.020)	0.042	0.125	0.120	67.5 (0.96)	4.38
LSD (P=0.05)	(0.002)	ns	ns	ns	ns	ns

[A] = Treatments applied where 1 = Silvine granules (Magnesium silicate) 150 kg/ha; 2 = Silvine granules 75 kg/ha plus Silvine powder 3.5 g/L water; 3 = Potassium silicate powder 150 kg/ha plus Potassium silicate liquid 10 mL/L plus foliar application potassium silicate liquid 2.5 mL/L fortnightly for 8 weeks; 4 = As for treatment 3 however Potassium silicate liquid foliar application also applied monthly until bolls formed; 5 = Untreated control; 6 = Potassium phosphate 8 g/L per metre row.

[B] = Data in parentheses are transformed percentage data (ASIN(SQRT(X/100)).

An important result of this trial was that Si uptake was significantly increased following Si soil amendment in an alkaline soil (Table 2). However, despite an increase in Si uptake there was no effect of Si on disease severity (Table 2). The threshold level of Si required in cotton for disease control is not known and may not have been met in this trial. There was also no significant effect of Si treatments on yield (Table 2 and Figure 4), even though the untreated control treatment had the lowest yield and magnesium silicate the highest. There was however, a high degree of variation between replicate plots in this trial, as discussed, a common problem encountered in field trial assessment.

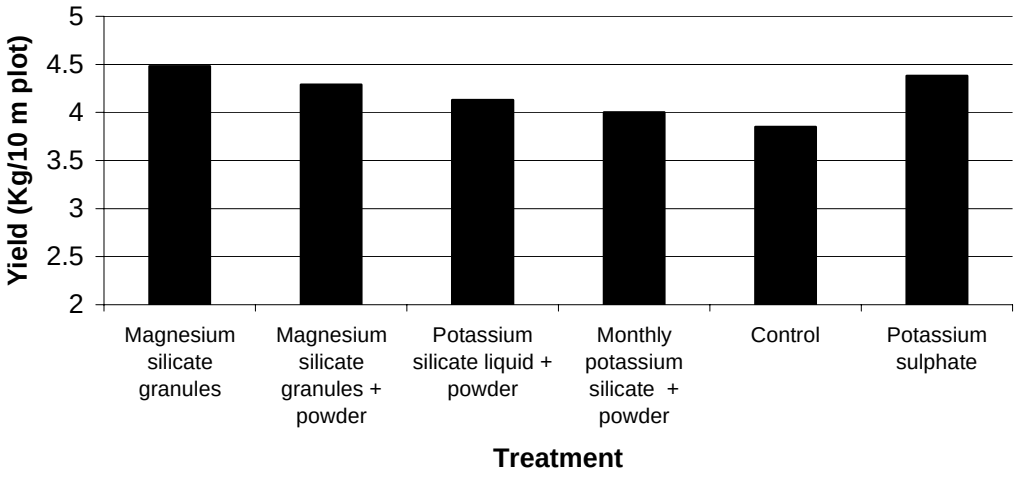

Figure 4. The effect of silicon amendment on seed cotton yield of cultivar Sicot F-1

Trial 2 (2005-2006)

In this trial Si uptake into the plant was not increased by Si amendment of the soil pre-plant. There was no effect of Si amendment on plant establishment or disease severity at maturity. There was no significant effect of Si amendment on seed cotton yield. There was however a significant effect of variety on disease severity as expected, with less disease in Sicot F-1 as determined by percentage of plants with less than 5% vascular discolouration (% 0 and 1's) (data not shown).

Although there was no treatment effect on Si uptake, there was however a varietal effect. There was a significantly higher level of Si in the leaves of the more susceptible variety Sicot 189 than the more tolerant variety Sicot F-1 (data not shown). Other nutrient differences in the leaves were determined between varieties including aluminium, iron and manganese, which were significantly lower in Sicot F-1 than Sicot 189 (data not shown). Nutrient difference may not be important from an agronomic sense when in sufficient concentrations above what is considered deficient, however it is not known if they are important for disease resistance/susceptibility.

Discussion

Results from Si trials support prior research that Si may be important in reducing the severity of fusarium wilt in cotton seedlings. Significant reductions in disease severity were obtained following the application of Si-containing fertilisers in glasshouse trials but were not effective under field conditions. Results of glasshouse trials suggest that *Fov* inoculum level influences effectiveness of Si amendment. The field site used for these trials is known for its high inoculum load, and for this reason has been used routinely for screening germplasm and new lines for wilt resistance. It is possible that inoculum load was too high for an effect of Si to be realized. Increasing nutrient availability in an alkaline soil also can be difficult and may have contributed to poor uptake. However, the same soil was used in both the glasshouse bioassays (although it was diluted with sand) and field trial assessment, therefore a response in the field was expected. Another consideration is that placement of fertiliser, which dictates availability of Si for uptake, may not have been optimal as it was applied to a 10 cm trench dug into the raised bed. Interesting

findings were published by Singh *et. al.* in 2005 regarding deep placement of phosphorus and the effect on various crops including cotton grown at Kununurra in Western Australia. Significant increases in the yield of seed cotton occurred when 50 kg/ha was applied at depth (10-15 and 25-30 cm), compared with the conventional placement at 7-10 cm. The response to deep phosphorus in these layers was attributed to the rapid drying of the soil surface layers, reducing the availability of soil or phosphorus fertiliser in these layers. The deep phosphorus remained available during the growing season and alleviated the phosphorus deficiency that appeared to be a feature of these soils when the surface layers became dry. This may very well be the case for Si. Had the Si fertiliser been applied deeper in the bed, or mixed uniformly throughout the soil profile, different results in Si uptake may have been observed. Further investigation of fertiliser placement and Si uptake is required.

Acknowledgments

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