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Control of sudden wilt in capsicum

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Pty Ltd

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Options for controlling Pythium root rot of capsicum

Graham Stirling and Lois Eden

Final report of HAL project VG02020

(completed 31 December 2004)

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Purpose of the report

Pythium root rot is the most important soil-borne disease problem in the Queensland capsicum industry. It can cause heavy losses to seedlings when temperatures are high or wet weather occurs within a few weeks of planting, and is thought to be involved in a disease of mature plants known as sudden wilt. This report describes research on the control of Pythium root rot on capsicum in tropical and subtropical environments. Evidence is presented which shows that reducing soil temperatures to levels that are unfavourable to heat-tolerant *Pythium* species is the most important control measure. Other useful management practices include improving the biological suppressiveness of soil with amendments of organic matter and reducing pathogen activity with chemicals such as metalaxyl, phosphite and silica.

Acknowledgments

This work would not have been possible without help from others. Ms Rosemary Kopittke advised on statistical analyses, Chrys Akem and Tony Parker helped with field trials in north Queensland, and Neil Halpin kept a watchful eye on trials in Bundaberg. Dale Reibel, Alan Napier and Joe Totorica cooperated with field trials at Bowen and Gumlu, while Don Halpin was an excellent co-operator at Bundaberg. Don managed our trials professionally and was always willing to accommodate experimental requirements that inevitably made his job more difficult. The Australian capsicum industry and HAL provided financial support for the project.

Date: 24 February 2005

MEDIA SUMMARY

Pythium root rot is the most important soil-borne disease in the Queensland capsicum industry. Seedlings suffer from root rot when temperatures are high and soil is wet during the first few weeks after planting, and *Pythium* is a component of a disease of mature plants known as sudden wilt. Growers have previously had relatively few options for reducing losses from root rot, but recent research has provided several potentially useful chemical and non-chemical control measures.

Dr Graham Stirling and Ms Lois Eden from Biological Crop Protection in Brisbane, who did the research on behalf of Horticulture Australia and the vegetable industry, found that reducing soil temperature was the key to controlling *Pythium* in capsicum crops. Species of *Pythium* that cause root rotting in tropical and subtropical areas thrive at soil temperatures of 35-40ºC, so practices such as changing from black plastic to white plastic mulch, planting into plant-based mulch that is grown *in situ*, or using a denser planting configuration to throw more shade on the bed (e.g. double rather than single rows), are all likely to reduce the impact of the disease.

Another useful control option is to use organic matter to enhance the soil's natural mechanisms of biological control. Experiments in vegetable-growing soils from Bundaberg showed that when the soil was amended with organic materials such as sugarcane trash and lucerne hay, losses due to Pythium root rot were reduced. Since the soil organisms that suppress *Pythium* took many months to develop and disease suppression was only obtained when microbial activity was high, it was necessary to incorporate the organic matter about 4-6 months before planting. Once the soil biology is enhanced with an organic amendment, it is important to preserve the new biological community by minimising tillage.

Three chemicals with potential to provide control of Pythium root rot were investigated. Drenches of metalaxyl were effective against the seedling stage of the disease; foliar sprays of phosphite reduced the amount of root rot in capsicum seedlings by about 30%, and silicon applied as a soil drench improved root health in glasshouse experiments. However, Dr Stirling warned that phosphite and EC formulations of metalaxyl are not yet registered for use on capsicums, while field studies on the efficacy and economics of using silicon for root disease control have not commenced.

TECHNICAL SUMMARY

Pythium root rot is the most important soil-borne disease in Australia's tropical and subtropical capsicum industry. *Pythium myriotylum*, *P. aphanidermatum* and other species of *Pythium* with high optimum temperatures kill seedlings when temperatures are high or soil is too wet during the first few weeks after planting, and are involved in a disease of mature plants that is known locally as sudden wilt. The objective of this work was to minimise the impact of Pythium root rot by modifying soil management practices, enhancing biological suppression of *Pythium* and controlling the pathogen with chemical and non-chemical treatments.

Previous work has shown that high soil temperatures and heat stress are important components of the Pythium root rot syndrome. The effects of changing plastic colour, modifying mulching practices and varying planting configurations were therefore investigated. Experiments with plastic colour showed that soil temperatures increased when black rather than white plastic was used as mulch, and this had a major impact on disease severity. In contrast, seedling mortality declined in plant-based mulches that were either laid manually or produced *in situ*, largely because soil temperatures were reduced. This result suggests that minimum till production systems in which seedlings are planted into *in situ* mulch should be further investigated from a soil health and disease minimisation point of view. Dense planting configurations (e.g. double rows) provided more shade than single rows and are therefore likely to improve control of Pythium root rot, particularly if they are used in conjunction with white rather than black plastic.

A series of experiments were done to investigate the role of organic matter in enhancing natural biological mechanisms of disease control. Incorporation of a forage sorghum green manure crop was found to exacerbate Pythium root rot if capsicum seedlings were planted within a month of incorporation, largely because *Pythium* is a good competitive saprophyte and is capable of colonising fresh organic matter. However, when organic matter was introduced 4-6 months before capsicums were planted, losses due to Pythium root rot were reduced. This effect was observed in field plots amended with sugarcane trash plus nitrogen; in bioassay plants grown in undisturbed cores containing soil previously amended with sugarcane trash, nitrogen and compost; and in a pot experiment with soil amended with sugarcane trash or lucerne hay. In all cases, root rotting was less severe as soil microbial activity increased.

Three chemicals with potential to provide control of Pythium root rot were also investigated. Metalaxyl was effective against the seedling stage of the disease, provided it was applied to potting mix just before seedlings were transplanted, or drenched around seedlings as they were planted. Phosphite was not as effective as metalaxyl, but foliar sprays increased phosphite concentrations in roots to levels known to control *Phytophthora*, a closely related root rotting pathogen, and reduced the severity of root rotting in capsicum seedlings by about 30%. Silicon was also a potentially useful control measure, as disease severity was reduced when SiO_2 was drenched around the roots of capsicum seedlings in the glasshouse. However, it may be some time before these products are available for use by vegetable growers. Phosphite and EC formulations of metalaxyl are not registered for use on vegetable crops, while field studies on the efficacy and economics of using silicon for root disease control are yet to commence.

TABLE OF CONTENTS

CHAPTER 1. INTRODUCTION

The Queensland capsicum industry

The tropical and subtropical capsicum industry, which is based around Bowen and Bundaberg in Queensland, is Australia's main capsicum production area during winter and spring. Planting commences in February and continues until September, which means that the Queensland industry has a dominant place in the market from May until December. Most of the capsicums are produced by specialised growers who purchase hybrid varieties from nurseries, transplant seedlings into raised beds covered with plastic, and water crops by trickle irrigation.

Pythium root rot of capsicum

Current methods of establishing capsicum crops are normally very successful, as less than 0.1% of capsicum seedlings are usually lost within a few weeks of planting. However, stunting and death of seedlings due to Pythium root rot is sometimes observed. The disease occurs most frequently at the end of rows, where water tends to accumulate due to inadequate drainage. However, widespread losses of seedlings can also occur, particularly following periods of extremely hot or wet weather. In a crop planted at Bowen in February 2003, for example, hundreds of seedlings in the rows closest to regularly-spaced windbreaks were stunted by Pythium root rot, probably because high temperatures and lack of air movement near the windbreaks exacerbated the disease. In another example, more than 30% of seedlings planted on black plastic in Bundaberg during January 2003 succumbed to Pythium root rot, largely because soil temperatures in beds were greater than 40ºC and more than 280 mm of rain fell in the first five days after planting. *Pythium aphanidermatum* and an unidentified *Pythium* sp. were associated with rotted roots at Bowen, while *P. aphanidermatum* was involved at Bundaberg.

Pythium root rot is also a component of an important disease of mature capsicum plants that is known locally as sudden wilt. Plants appear healthy until fruit set, when they wilt suddenly, drop their leaves and then die or produce small, unmarketable fruit. Outbreaks are unpredictable, with minor losses in some years and serious losses in others. Research done in a previous Horticulture Australia project (Stirling *et al.* 2003; 2004) indicated that severe root rotting always occurs before plants collapse with sudden wilt and several potential fungal pathogens were isolated from rotted roots. However, two species of *Pythium* (*P. myriotylum* and *P. aphanidermatum*) were the only fungi to cause severe root rotting in mature capsicum plants. Root rotting was much more severe at 35-40ºC than at 30ºC. High temperatures were also sub-optimal for capsicum, as root health was poor and shoot growth was markedly reduced at 35 and 40ºC in the absence of pathogens. Since soil temperatures greater than 35ºC may occur at certain times of the year in beds used for capsicum production, it was concluded that sudden wilt is the result of a pathogen × environment interaction in which heat-stressed plants are attacked by *P. myriotylum* or *P. aphanidermatum*.

Control options

Chemical controls are always the first option considered in the vegetable industry when control measures for plant diseases are being developed. However, in the case of Pythium root rot, available evidence suggests that environmental factors are important in exacerbating the disease. It is therefore unlikely that Pythium root rot will be controlled by concentrating only on chemical treatments that target the pathogen. Environmental modification must also be considered, particularly as it pertains to the physical factors (e.g. moisture and temperature) that impact on disease severity, and the biological factors that influence disease suppression.

Excessive soil moisture is recognised as a major factor influencing the severity of diseases caused by *Pythium* (Hendrix and Campbell 1973), and improved irrigation management is therefore one option for reducing losses from Pythium root rot. However, opportunities to change the way crops are watered are relatively limited. Most growers are aware that over-watering exacerbates root rot problems and take particular care to limit the amount of water applied to young seedlings. In mature crops, irrigation frequency is determined using computerised moisture monitoring equipment, which means that in the absence of uncontrollable factors such as heavy rainfall, soil moisture is usually as close as is practically possible to the levels required for optimal plant growth.

Evidence from previous studies (Stirling *et al.* 2004) clearly indicates that when soils are infested with *Pythium* species that have high optimum temperatures, soil temperature has a major effect on the severity of Pythium root rot. Root systems are largely destroyed by *P. aphanidermatum* and *P. myriotylum* at temperatures above 35ºC, whereas they cause little damage at 30ºC. This suggests that reducing soil temperatures by as little as a few degrees will have a major effect on disease severity. Options for achieving this include changing the colour of the plastic mulch, using a non-plastic mulch, avoiding growing crops during excessively hot weather, and changing planting configurations to provide more shade on the surface of the bed.

Although the effects of the biological environment on diseases caused by Pythium are poorly understood, Hoitink and Boehm (1999) showed that some *Pythium* species cause severe damage in relatively sterile potting mixes, whereas root rotting is suppressed when the potting mixes are microbially active. Since soils used for capsicum production in Queensland are low in microbial activity (Pung *et al*. 2003), it is therefore possible that losses from *Pythium* root rot could be reduced by improving their microbial status. Reducing tillage and increasing the amount of organic matter returned to soil are management options that should therefore be considered.

Project objectives

The objective of the work described in this report was to find control measures for Pythium root rot of capsicum that were effective against the seedling stage of the disease and also reduced the severity of sudden wilt in mature plants. The research had three main components: 1, Identifying chemical and non-chemical treatments that are effective against the *Pythium* species that cause root rot in tropical and subtropical environments; 2, Minimising the impact of Pythium root rot by reducing soil temperatures and 3, Enhancing biological suppression of *Pythium* by minimising tillage and increasing organic inputs into capsicum-growing soils.

CHAPTER 2. A BIOASSAY TO ASSESS THE POTENTIAL FOR PYTHIUM ROOT ROT IN CAPSICUM-GROWING SOILS

Introduction

Previous work in soils infested with *Pythium aphanidermatum* and *P. myriotylum* showed that these pathogens cause severe root rotting in capsicum at 35ºC but virtually no symptoms at 30ºC (Stirling *et al.* 2004). The fact that inoculum density is not necessarily related to disease severity means that there is little point in estimating levels of Pythium inoculum when predicting whether root rotting will occur in a particular soil, or determining whether an experimental treatment has been effective. This chapter describes a bioassay that can be used to assess the level of Pythium root rot likely to occur in field soil. Its main value is that it accounts for the fact that environment × host × pathogen interactions are important in the etiology of Pythium root rot in tropical and subtropical environments.

Methods

All experiments were done using capsicum seedlings (cv. Target) transplanted into 15 cm high, 6.5 cm diameter watertight pots containing 400 mL of soil. Pots were placed into temperature-controlled waterbaths and the soil was maintained at different temperatures.

Temperature required to eliminate pathogen activity

A clay loam soil from Ayr with a history of vegetable growing was either heated at 50, 55, 60 or 65°C for 35 minutes, autoclaved at 121°C for 20 minutes or left unheated. Three replicate pots were then filled with each soil, 6-week-old capsicum seedlings were planted in the pots and plants were grown for 3 days at a soil temperature of 28°C followed by 10 days at 36°C. Roots were then washed and rated for root rotting using a 1 - 6 scale, where 1 = 0-5%, 2 = 6-24%, 3 = 25-49%, 4 = 50-74%, 5 = 75-99% and 6 = 100% of the roots were rotted. If root lesions were present, tissue was taken from the margin of the lesions and either surfaced sterilised in NaOCl and plated on P + S (potato dextrose agar amended with 0.12 g/L streptomycin) or washed in sterile distilled water and plated on 3P (cornmeal agar amended with 0.05 g/L penicillin; 0.05 g/L polymixin and 0.025 g/L pimaricin, Eckert and Tsao, 1962).

Bioassay of field soils for Pythium root rot

In July 2003, nine soils were collected from fields in the Bundaberg area. All fields were to be planted to capsicum in spring 2003 and eight of the soils had a recent history of vegetable production. Soil no. 9 had been fallow for nine months after sugarcane.

Each soil was sieved through a 3.2 mm sieve and any organic matter present was cut into pieces (less than about 1 cm^2) and returned to the soil. The soil and organic matter were then mixed thoroughly, five replicate pots were filled with each soil and a capsicum seedling was planted in each pot. Five replicate control pots were prepared by heating each soil at 65°C for 1 hour. Plants were evaluated for root rotting using methods described in the previous experiment.

Pathogenicity under different soil temperature regimes

Seedlings were planted in two soils with a history of vegetable production and grown for 3 days at a soil temperature of 28°C. Five plants in each soil were then transferred to a water bath at 36°C, while another five were grown for 8 hours each day at 36°C and for 16 hours at the ambient temperature in the glasshouse (about 25°C). After 10 days, roots were rated for root rotting as described above.

Results

Temperature required to eliminate pathogen activity

Roots of plants were severely rotted when grown in unheated soil and in soil heated to 50°C. Root rotting was not as severe at 55°C and was not evident at 60°C (Figure 2.1). *P. aphanidermatum*, *P. deliense* and another unidentified *Pythium* sp. were isolated from affected plants on both 3P and P+S.

Bioassay of field soils for Pythium root rot

Plants from eight of nine soils showed severe root rotting (Figure 2.2), with symptoms extending to the crown in some replicates of some soils. Root rotting was not observed in the soil previously used for sugarcane. *P. aphanidermatum* was isolated from all soils (except soil no. 9 from sugarcane) and an unidentified *Pythium* sp. with an optimal growth temperature of about 35°C was isolated from four vegetable-growing soils.

Figure 2.1: The effect of heating soil at different temperatures for 35 min, autoclaving for 20 min (A) or leaving soil untreated (U), on the severity of capsicum root rot.

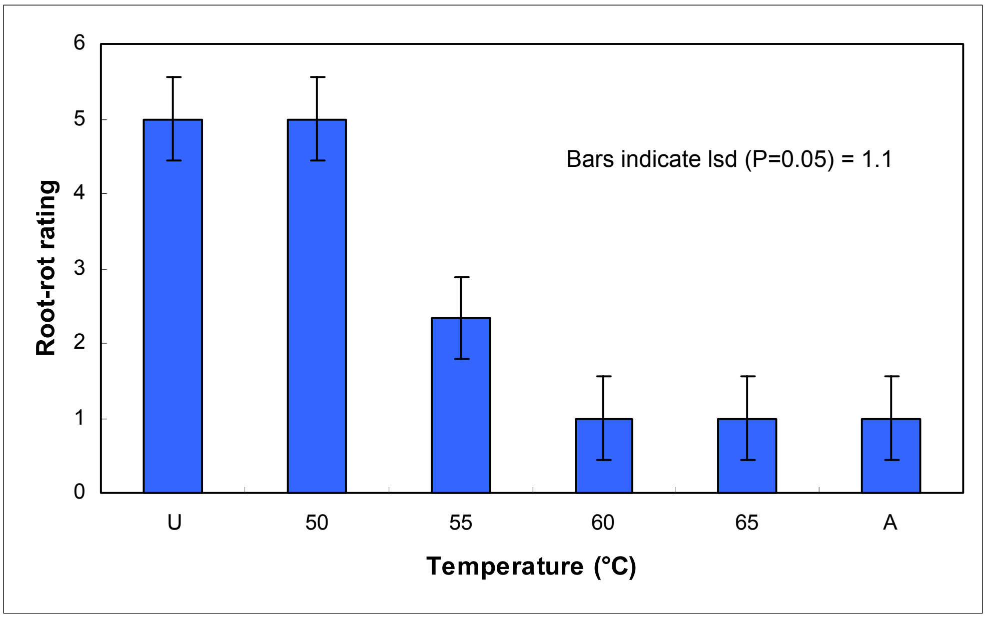
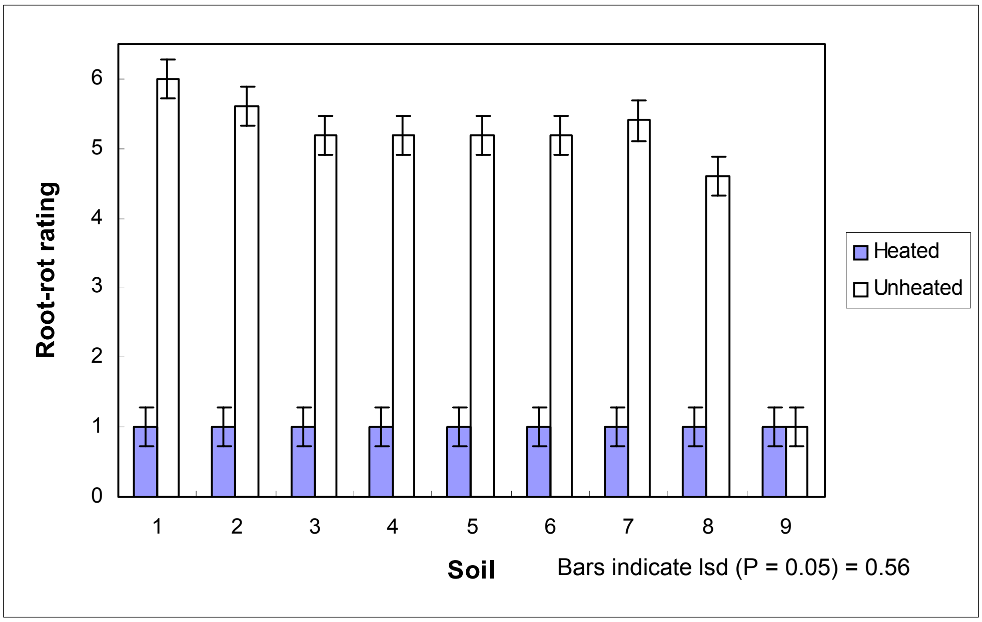


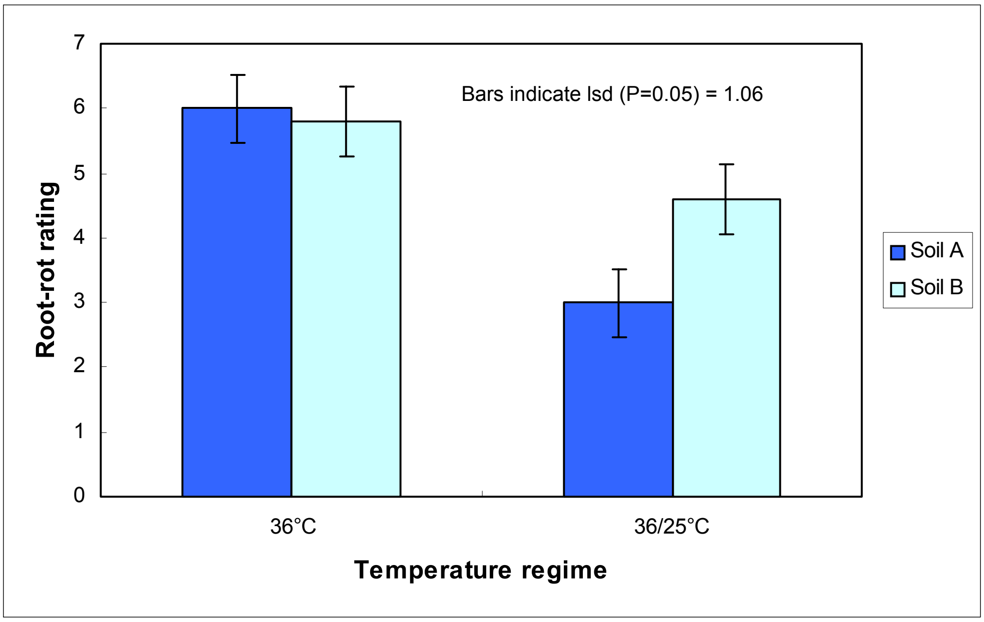
Figure 2.2: The severity of root rotting in heated and non-heated vegetable-growing soils and one sugarcane soil (soil 9) when bioassay plants were grown at a constant soil temperature of 36ºC.



Pathogenicity under different soil temperature regimes

Roots of plants grown at a constant 36°C were severely rotted in both soils. However, when plants were grown at 36°C for only 8 hours a day, roots were more severely rotted in one soil than the other (Figure 2.3).

Figure 2.3: Effect of two soil temperature regimes, 36°C for 24 hours/day or 36°C for 8 hours/day and 25°C for 16 hours/day, on root rot in capsicum bioassay plants grown in two soils from Bundaberg.



Discussion

When capsicum plants were grown in field soil under temperature conditions that stressed the plant, severe root rotting was observed. However, when soil was heated to above 55°C and then bioassayed, symptoms were no longer evident, indicating that the root rotting had a biological cause. Interestingly, heating to 50°C for 35 minutes did not reduce the severity of root rot, indicating that the pathogens involved are relatively resilient to heat.

Pythium species with high optimal growth temperatures appeared to be the primary root rotting pathogens, as they were isolated from capsicum roots in all eight vegetable-growing soils. The soil from which *Pythium* was not recovered had a history of sugarcane rather than vegetables. These results show that *Pythium* is widespread in vegetable-growing soils, and that the fungus can destroy capsicum root systems when environmental conditions are favourable.

This bioassay is useful for assessing the root-rotting potential of soils to be used for capsicum production. However, the use of a constant temperature of 36°C created an environment that favoured the disease and probably overestimated the degree of root rotting that would occur in a field situation. A constant high temperature also meant that it was difficult to separate soils that differed in their root-rotting potential (see Figure 2.2). The severity of root rotting decreased when the temperature was reduced for part of the day, and such a temperature regime probably gives a better indication of what is likely to occur in the field. It is certainly more useful for comparing the root rot potential of different soils (see Figure 2.3).

On the basis of the above results, a high/low temperature regime was used for all the bioassays done later in this report (unless otherwise indicated). Assay soils were either collected from the field, or potting mix was assayed after it was inoculated with *Pythium* or left uninoculated. Capsicum seedlings were planted in 400 mL of soil and seedlings were grown for 3 days at 28°C to allow them to establish without damage from root pathogens. They were then transferred to a day/night soil temperature regime of 36°C for 8 hours followed by 22-25°C for 16 hours. Aboveground temperatures were maintained at 22-28°C throughout the day. After 10 days, the severity of root rot was rated on a 1 to 6 scale where 1 = 0-5%, 2 = 6-24%, 3 = 25-49%, 4 = 50-74%, 5 = 75-99% and 6 = 100% rotted roots.

CHAPTER 3. CONTROL OF PYTHIUM ROOT ROT BY REDUCING SOIL TEMPERATURE

Introduction

In soils that are infested with *Pythium* species that grow well at 35-40ºC, soil temperature has a major effect on the severity of Pythium root rot (Stirling *et al.* 2004). Root systems are largely destroyed by *P. aphanidermatum* and *P. myriotylum* at temperatures above 35ºC, whereas there is little damage at 30ºC. This suggests that reducing soil temperature by as little as a few degrees will have a major effect on disease severity. The following work aimed to verify this by comparing the level of root rot in experiments where soil temperature was altered by changing plastic colour or the type of mulch. An experiment was also done to see whether changing the planting configuration affected soil temperatures in capsicum beds.

The effect of mulch colour and mulch type on Pythium root rot (Ayr 2003)

Methods

In late March 2003, an experiment was established at Ayr Research Station using three different mulches: white plastic, white plastic painted black and a plant-based mulch. The experimental site was initially prepared by laying white plastic on beds with a single irrigation line down the centre. Plots 10 m long were then marked out and the plastic was either left unaltered, painted black (using acrylic low sheen paint) or removed completely. Capsicum seedlings (c.v. Aries) were then planted in double rows on either side of the irrigation line at a spacing of one plant per 27.5 cm of bed. The plant-based mulch was formed by spreading about 2.3 bales of a 1:2:2 mixture of sugarcane trash, grass hay and kenaf stems onto plots from which plastic had been removed. Initially, the mulch was 10-15 cm deep and was laid so that it was not in contact with the seedlings. Later, the mulch subsided to a depth of 5-10 cm. Temperature probes (Tiny Talk®) were placed in the centre of beds and data loggers were used to record hourly temperatures in each treatment at depths of 5 and 15 cm.

The effects of compost and Superzyme (a commercial biological product supplied by Zadco Pty. Ltd. and purported to contain *Trichoderma*, *Bacillus* and *Pseudomonas* spp.) were also examined in this experiment. Five replicate plots involving +/- compost and +/- Superzyme were therefore included within each mulch treatment, so that the final experimental design was 3 mulch treatments × 2 compost treatments × 2 Superzyme treatments × 5 replicates. The compost was prepared by a local grower and was incorporated into soil prior to planting at an application rate of 10 t/ha Superzyme (1 kg/ha) was applied through the irrigation water when the capsicums were planted and the application was repeated 4 and 8 weeks after planting.

Fifteen days after planting, the number of dead and dying plants in each treatment was assessed. Roots from healthy and dying plants were examined and pathogens in infected root tissue were isolated by plating pieces of root tissue on P+S and 3P media as described previously (Chapter 2).

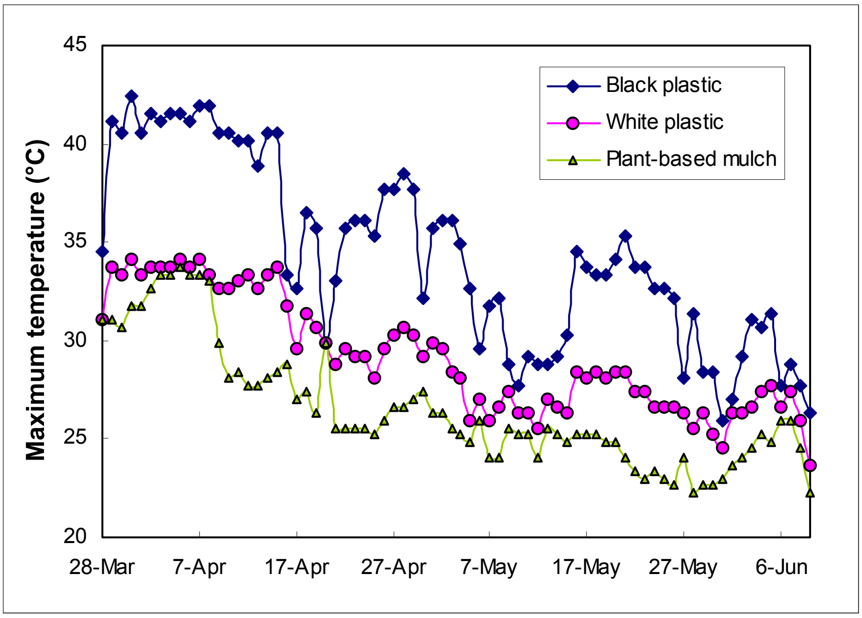
Six weeks after planting, plants in each plot were given a growth rating of 1-4 where 1 = a poor plant less than 15 cm high, 2 = a plant 15-25 cm high with a few flowers and occasional fruit, 3 = a plant 25-35 cm high with some flowers and a few small fruit and 4 = a plant 35-50 cm high with more than 8 flowers and a few small fruit. The ratings for each plant were then averaged to produce a growth rating for each plot.

After 11 weeks, the plants surviving in each plot were counted and plant health was assessed using a scale of 1-6 where: 1= plants small, with occasional flowers and small fruit and some chlorosis, 2 = plants less than 30 cm with 1-2 fruit and sometimes mild chlorosis, 3 = plants 30-45 cm with 2-3 mature fruit, 4 = plants 45-60 cm with 3-4 mature fruit, 5 = plants 45-60 cm with 5 or more mature fruit, 6 = plants over 60 cm with 5 or more mature fruit. The ratings for each plant were then averaged to calculate a growth rating for each plot. Shoots of two plants from each plot were sampled and fresh weights of the ripe fruit were measured. Roots of four plants per plot were washed, fresh weights of roots were measured and small roots were rated for rotting as described previously (Chapter 2).

Results

There were marked differences in soil temperatures under the three mulch treatments (Figures 3.1 and 3.2). In the first few weeks after planting, average soil temperatures at a depth of 5 cm under the black, white and plant-based mulches were 32.0, 28.7 and 27.1°C, respectively, and the average maximum temperatures at the same depth were 40.9, 33.4 and 30.9°C. Thus, the plant-based mulch reduced average maximum temperatures by 10 and 2.5°C compared with black and white plastic respectively. On a daily basis, the maximum soil temperature under the plant-based mulch was often 12ºC lower than under black plastic and 5°C lower than under white plastic (Figure 3.1).

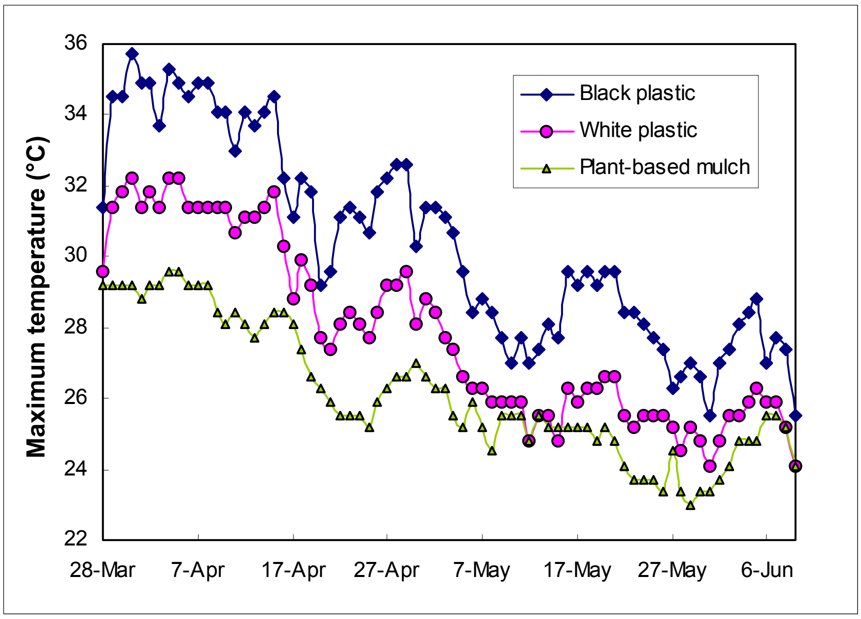
Figure 3.1: Daily maximum soil temperatures at 5 cm from March to June 2003 under a capsicum crop at Ayr growing in beds mulched with black plastic, white plastic or plant-based mulch.



Differences in soil temperature due to treatment, while not as extreme as those at 5cm, also occurred at a depth of 15 cm. During the first few weeks, average soil temperatures under black, white and plant-based mulches were 31.5, 29.3 and 27.8°C respectively, and the average maximum temperatures were 34.3, 31.4 and 28.8°C. On a daily basis, the plant-based mulch reduced maximum soil temperature at 15 cm by about 3ºC compared with white plastic and 6ºC compared with black plastic (Figure 3.2).

Soil temperatures decreased as the season progressed, and although differences between mulch treatments were always present, they were much smaller late in the growing season (Figures 3.1 and 3.2). By early June, temperatures under the plant-based mulch and white plastic were possibly low enough to limit plant growth, as maximum temperatures at 5 and 15 cm were less than 25°C and minimum temperatures were a few degrees lower.

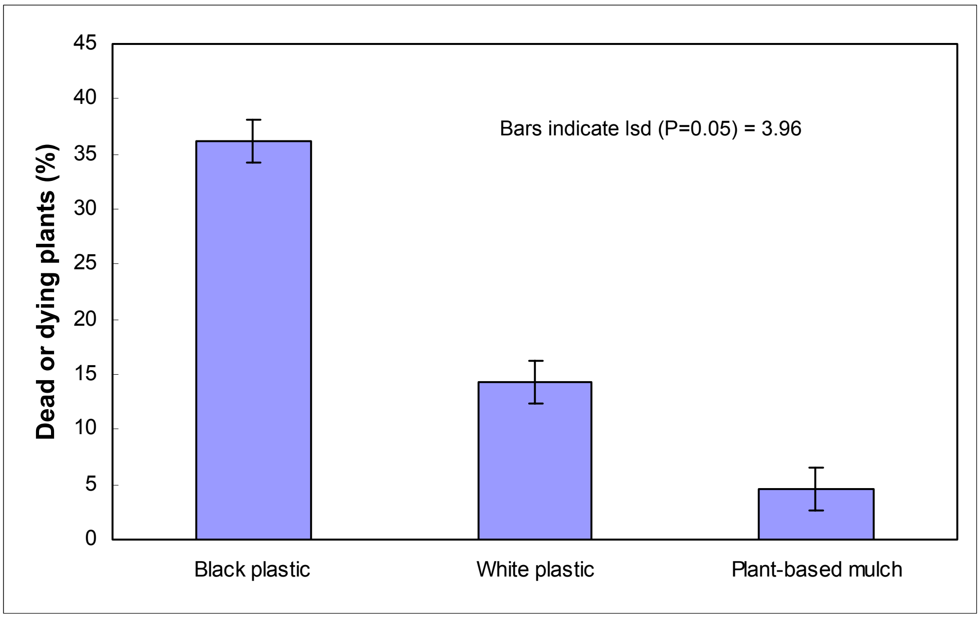
Figure 3.2: Daily maximum soil temperatures at 15 cm from March to June 2003 under a capsicum crop at Ayr growing in beds mulched with black plastic, white plastic or plant-based mulch.



When plant health and fruit yield data were analysed as a factorial using analysis of variance, compost and the type of mulch both had significant effects whereas Superzyme had no effect. Results for compost are presented in Chapter 4, while the main effects of the mulch treatments are presented below.

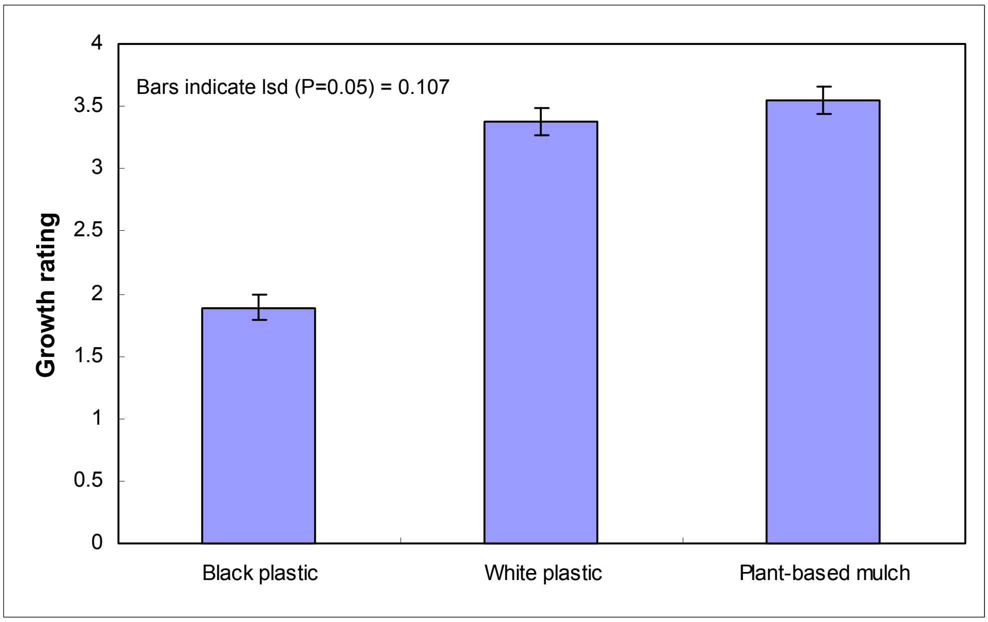
Fifteen days after seedlings were planted, some seedlings were wilting during the heat of the day and others had died. Seedling mortality under black plastic, white plastic and plant-based mulch treatments was 36.2%, 14.3% and 4.6% respectively (Figure 3.3). Roots of dead and dying plants were severely rotted and stem lesions were observed under the soil line. *P aphanidermatum*, *P. deliense* and an unidentified *Pythium* species, were isolated from diseased roots. All three of these species had optimal growth temperatures of 35-36°C.

Figure 3.3: Severity of Pythium root rot in April 2003, 15 days after capsicums were planted on different types of mulch at Ayr.



Surviving seedlings that were affected by root rot recovered when soil temperatures started to decline in late April. However, growth ratings at 6 weeks reflected earlier disease severity (Figure 3.4), with plants grown under black plastic significantly smaller than those grown under white plastic or plant-based mulch.

Figure 3.4: Growth rating of capsicum plants in May 2003, 6 weeks after seedlings were planted into different types of mulches at Ayr.



At maturity, plants grown on plant-based mulch had a higher survival rate, plant growth rating and fresh root weight than those grown under black or white plastic (Table 3.1). Plants on white plastic were better in all respects than those on black plastic. Root rot ratings were highest in plants grown on black plastic, but the level of damage in all treatments was relatively low, with rotting confined mainly to small roots.

Table 3.1: Effect of mulch type on capsicum plants in June 2003, 11 weeks after planting at Ayr.

Mulch type	Plant survival (%)	Plant growth rating	Weight of fruit per plant (g)	Root-rot rating	Fresh weight of roots per plant (g)
Black plastic	60.6	3.8	586	1.6	5.2
White plastic	85.5	4.6	1098	1.2	7.6
Plant-based mulch	90.7	4.9	983	1.3	8.7
l.s.d. (P=0.05)	5.01	0.22	188.1	0.14	0.70

The effect of mulch colour on Pythium root rot (Ayr 2004)

Methods

This experiment was established on the Ayr Research Station in early March 2004. Beds were covered with white plastic and a single irrigation line was laid down the centre of each bed. Plots 7.5 m long were marked out and the plastic was either left unaltered or painted black (with acrylic low sheen paint), except that an 18 cm strip of white was left down the centre of the bed. Capsicum seedlings (cv. Aries) were planted 21 or 27.5 cm apart in a single row in the centre of the bed. Temperature probes (Tiny Talk®) were placed approximately 10 cm from the western edge of beds covered with each type of mulch and data loggers were used to record hourly temperatures at 5 and 15 cm.

The effect of two chemicals (metalaxyl and phosphite) was also examined in this experiment. Five replicate plots of four treatments (nil, metalaxyl, phosphite and metalaxyl + phosphite) were therefore included within each mulch treatment, so that the final experimental design was 2 mulch treatments × 2 planting distances × 4 chemical treatments × 5 replicates. Details of the chemical treatments can be found in Chapter 5.

One week after planting, the number of dead and dying plants was counted and diseased plants were sampled. Roots were examined and then pathogens were isolated from pieces of infected root and stem tissue using isolation methods described previously. Fruit was picked three and four months after planting, fresh weight was measured and yield per plant was calculated.

Results

Analyses of the data showed that planting distance did not affect any of the parameters measured. Results for the chemical treatments are presented in Chapter 5, while the following results are the main effects of the mulch treatments on temperature, seedling mortality and yield.

The temperature data collected during the experiment were considered unreliable because one of the probes was disturbed after it was set in position. Nevertheless, manual measurements taken during the first week after planting indicated that maximum temperatures at 5 cm were about 40ºC under black plastic and 33ºC under white plastic. The soil was also relatively wet, with 15 mm of rain being recorded 2 days after planting. This resulted in high seedling mortality, particularly in seedlings on black plastic. One week after planting, 29.2% of the seedlings on black plastic had died, compared to only 6.4% on white plastic (Table 3.2). Roots of diseased plants displayed little or no rotting but stems had severe lesions near the soil line. *Pythium* was occasionally isolated from rotted roots, while all the fungi obtained from stem lesions were saprophytes.

When the crop was harvested, fruit yield per metre of bed was greater for white plastic than for black plastic, largely because more plants remained. However, the average yield of plants that reached maturity was not affected by plastic colour (Table 3.2).

Table 3.2: Effect of plastic colour on a capsicum crop planted on black or white plastic at Ayr in March 2004.

Plastic colour	Mortality rate (%)	Fruit yield per m (kg)	Fruit yield per plant (kg)
Black	29.2	3.62	1.31
White	6.4	4.90	1.32
l.s.d. (P=0.05)	6.42	0.367	n.s.

The effect of mulch colour and plant position on Pythium root rot (Bundaberg 2004)

Methods

This experiment was established on a grower's property at Bundaberg and aimed to determine whether soil temperatures could be lowered by changing the positioning of plants on the bed, thereby reducing the severity of Pythium root rot. Beds 0.75 wide were covered with black plastic and had two irrigation lines, both about 12 cm from the centre of the bed. The four treatments consisted of black plastic with a single row of plants in the centre of the bed, white plastic (achieved using acrylic low sheen paint) with a single row of plants in the centre of the bed, black plastic with two rows of plants 22 cm apart and black plastic with a single row of plants on the northern side of the bed, about half way between the centre and the edge.

Five replicate 12 m plots of each treatment were established and seedlings of capsicum (cv. Tycoon) were planted 23 cm apart on 24 September 2004. Plants in the centre of the bed were machine-planted, while the other two treatments were hand-planted. Temperature probes (Tiny

Talk®) were placed approximately 10 cm from the edge of the north side of bed and data loggers recorded hourly temperatures at 5 cm.

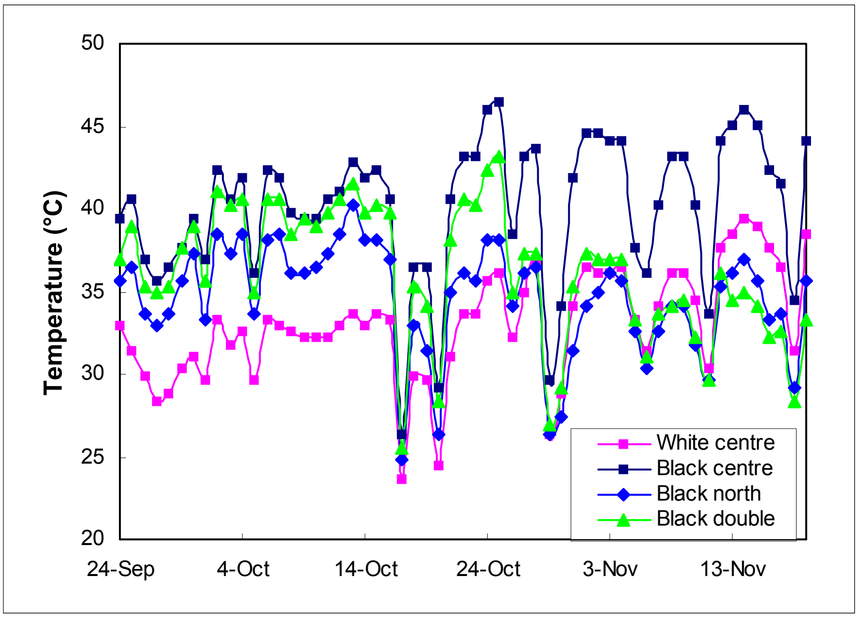
After capsicums were planted, plots were split to accommodate foliar sprays of +/- phosphite. Thus on 13 December, when the experiment was harvested, there were two datum plots each 5 m long within each plot. Four plants from each datum plot were chosen at random, the weight of marketable, sunburned and immature fruit on these plants was recorded and roots were examined for Pythium root rot.

Results

Because the crop was not staked, plants tended to fall over as the crop matured. However, plants in double rows and in the single row on the north side of the bed were more prone to collapse than the standard single row in the centre of the bed. Since affected plants tended to fall outwards (i.e. towards the inter-row), the centre of the beds in these treatments was left exposed from about mid November. The effects of planting position on soil temperatures were therefore only considered for the first 8 weeks after planting.

Maximum soil temperatures under black plastic were 7-10°C higher than under white plastic throughout most of the first 8 weeks (Figure 3.5). During the first 4 weeks, temperatures on the northern side of the bed in plots with double rows or a northern row on black plastic were usually 1-2°C lower than plots with a single central row on black plastic. However, as the crop matured and started to provide shade, temperatures in these treatments declined to levels that were lower than the white plastic. In plots with only a single row on the northern side, soil temperatures on the exposed southern side remained high whereas this did not occur when there was a double row of plants (data not shown).

Figure 3.5: Daily maximum soil temperatures 10 cm from northern edge of beds at a depth of 5 cm under a capsicum crop at Bundaberg growing in beds mulched with black plastic and planted in different patterns (centre row, northern row, double row), or mulched with white plastic.



When fruit from the experiment was harvested, some was not marketable due to sunburn, with losses being more severe on black plastic than white plastic (Table 3.3). Plants growing in a single centre row on white plastic produced significantly more marketable fruit per plant than the same planting pattern on black plastic (Table 3.3). Individual plants in a double row on black plastic yielded less marketable fruit than other treatments, but the higher planting density in this treatment compensated for the low yields per plant. Plants in both the high-yielding treatments were more chlorotic than other plants at the end of the season, almost certainly because of high nutrient demand due to the greater fruit load.

Roots were healthy at the end of the experiment and showed no signs of Pythium root rot. Symptoms of sudden wilt were not observed, and so it was not possible to draw conclusions about the effect of temperature on this component of the Pythium root rot syndrome.

Table 3.3: Effect of plastic colour and planting pattern on marketable and sunburnt fruit produced by a capsicum crop planted on black and white plastic at Bundaberg in September 2004.

Plastic colour	Planting pattern	Fruit yield (kg/plant)			Fruit yield (kg/metre of bed)		
		Marketable	Sunburnt	Total	Marketable	Sunburnt	Total
White	Single centre row	1.91	0.06	1.97	8.43	0.24	8.67
Black	Single centre row	1.41	0.18	1.59	6.17	0.80	6.97
Black	Single northern row	1.25	0.19	1.44	5.43	0.85	6.28
Black	Double rows	0.89	0.11	1.00	7.83	0.99	8.82
l.s.d. (P = 0.05)		0.179	0.079	0.198	1.428	0.557	1.199

Discussion

Data from the experiment at Ayr in 2003 indicated that in the upper 15 cm of the bed, soil temperatures under black plastic in the first few weeks after planting were 3-7ºC higher than temperatures under the white plastic normally used in the tropical capsicum industry. This increase was high enough to markedly increase seedling mortality due to Pythium root rot, and clearly shows the importance of high soil temperatures in exacerbating this disease. Although black plastic is never likely to be used commercially for capsicums in the tropics, our results demonstrate that relatively small changes in temperature can have a major impact on disease severity in an environment where temperatures under white plastic in summer and early autumn often reach levels that are sub-optimal to capsicum. The results also suggest that even under white plastic, a week or two of extremely hot weather is likely to create environmental conditions that are conducive to Pythium root rot.

Although black plastic had a similar effect on soil temperatures and seedling mortality at Ayr in our trial the following year, *Pythium* was not readily isolated from affected plants. This suggests that at least some of the deaths in this experiment were due to heat stress, and again demonstrates the vulnerability of capsicum seedlings to soil temperatures above about 35ºC.

The plant-based mulch used in the 2003 field trial had the biggest impact on soil temperatures, and this resulted in seedling mortalities that were lower than under white plastic. Manually-laid mulches are never likely to be economically viable in the capsicum industry, but this result suggests that production systems in which seedlings are planted into *in situ* mulch should be further investigated from a soil health and disease minimisation point of view. Initial studies in Bundaberg (see Chapter 4 and Stirling 2005) have already demonstrated that the severity of Pythium root rot can be reduced by planting seedlings into undisturbed beds mulched with forage sorghum. Minimum till and *in situ* mulch cropping systems therefore have potential, but are likely to be most useful for crops planted in summer and early autumn, when temperatures in the tropics and sub-tropics are often sub-optimal for capsicums. Plant-based mulches may be detrimental to later plantings, as soil temperatures may be reduced to levels that affect plant growth.

Although planting position was varied in an experiment at Bundaberg in 2004, Pythium root rot did not occur because the previous crop was sugarcane and the field had been fallowed for 18 months prior to planting capsicum. The likely effect of planting position on the disease was therefore determined by examining the effects of treatments on soil temperature. A single row of capsicums on the northern side of the bed shaded the area where excessive heating often occurs, and from late October this resulted in soil temperatures that were similar to or even lower than those under white plastic. However, a larger area on the southern side of the bed was left unshaded, which meant that there was no net benefit from this approach. The double row of plants on black plastic was more useful, as soil temperatures on both sides of the bed were similar to those under white plastic. Such a planting arrangement is therefore likely to result in the best control of Pythium root rot, particularly if it is used in conjunction with white plastic. Double row planting is used extensively in north Queensland, possibly because growers have inadvertently found that it reduces heat stress and the severity of Pythium root rot. Although many plants in double rows toppled over in our trial, this is overcome in practice by selecting an appropriate variety, or by trellising.

Sudden wilt was not observed in the two years of this study, either in our experiments or in commercial plantings at both Bowen and Bundaberg. It is therefore not possible to conclude with certainty that practices that reduce soil temperature (e.g. shading, white rather than black plastic, plant-based mulches, double row planting) will have an impact on sudden wilt. However, our understanding of the pathogen × environment interactions involved in sudden wilt (Stirling *et al.* 2004) suggests that such practices should reduce disease severity.

CHAPTER 4. ENHANCING BIOLOGICAL SUPPRESSION OF *PYTHIUM* WITH ORGANIC MATTER

Introduction

Advances in modern agriculture, particularly the introduction of synthetic fertilisers and pesticides and the development of disease-resistant varieties, have allowed farmers to break the well-established link between organic matter and soil fertility (Hoitink and Boehm 1999). Crop rotation has largely been abandoned, green manure crops are rarely included in the farming system and by-products such as manures are seen as solid wastes rather than valued resources. As levels of soil organic matter have declined due to mineralisation over time, soil structure has deteriorated and diseases caused by soil-borne pathogens have reached epidemic proportions. This decline in soil health has reached its zenith in the vegetable industry, which is faced with more soil-related problems than any other agricultural industry in Australia. Soils used for vegetable production are almost devoid of microbial activity (Pung *et al.* 2003) and soil is often fumigated to control soil-borne diseases.

One way of reducing losses from soil-borne pathogens such as *Pythium* is to use organic matter to restore the soil's natural mechanisms of biological control. When green manures, animal manures and composts are added to soil, edaphic microorganisms recolonise the soil and help suppress pathogens through mechanisms such as competition, antibiosis, parasitism and predation (Hoitink and Boehm 1999).

One problem with using organic matter amendments to control competitive saprophytes such as *Pythium* is that they sometimes increase disease severity. Fresh organic matter and immature or inadequately-stabilised composts are colonised by the pathogen, and so there is an initial period when populations of *Pythium* in amended soil may increase. Later, when the incorporated organic matter has been colonised by soil microorganisms, the disease is suppressed, even though populations of the pathogen may remain elevated (Hoitink and Boehm 1999).

In the work reported in this chapter, the capacity of *Pythium* to grow saprophytically on various green manures, amendments and composts was confirmed, and experiments were done to see whether such materials could be used to enhance suppressiveness to the pathogen. Since forage sorghum is particularly suited for use as a green manure crop in the tropics and subtropics, it was often used as a source of organic matter.

Effect of compost on Pythium root rot (Ayr 2003)

Methods

This field trial was established at Ayr in 2003 and consisted of 3 mulch treatments × 2 compost treatments × 2 Superzyme treatments × 5 replicates. Details of the experiment and some of the results were presented previously (Chapter 3), but the effect of compost is covered in this chapter. The compost was prepared by a local grower and was incorporated to a depth of 15 cm prior to planting capsicums. The application rate was 10 t/ha.

Results

The main effects of adding compost are presented in Table 4.1. Compost did not affect the survival of plants, but it improved plant growth and root biomass. However, root rotting was more severe in soil amended with compost than it was in non-amended soil.

Table 4.1: Effect of compost on growth, yield and root rotting in an 11-week-old crop of capsicums grown at Ayr from March-June 2003.

Treatment	Plant survival (%)	Plant growth rating	Weight of fruit (g/plant)	Root-rot rating	Fresh weight of roots (g/plant)
Compost	79.0	4.5	960	1.41	7.9
No compost	78.8	4.3	817	1.26	6.4
l.s.d. (P=0.05)	n.s.	0.18	n.s.	0.118	0.57

Effect of green manured forage sorghum on Pythium root rot (Bundaberg 2003)

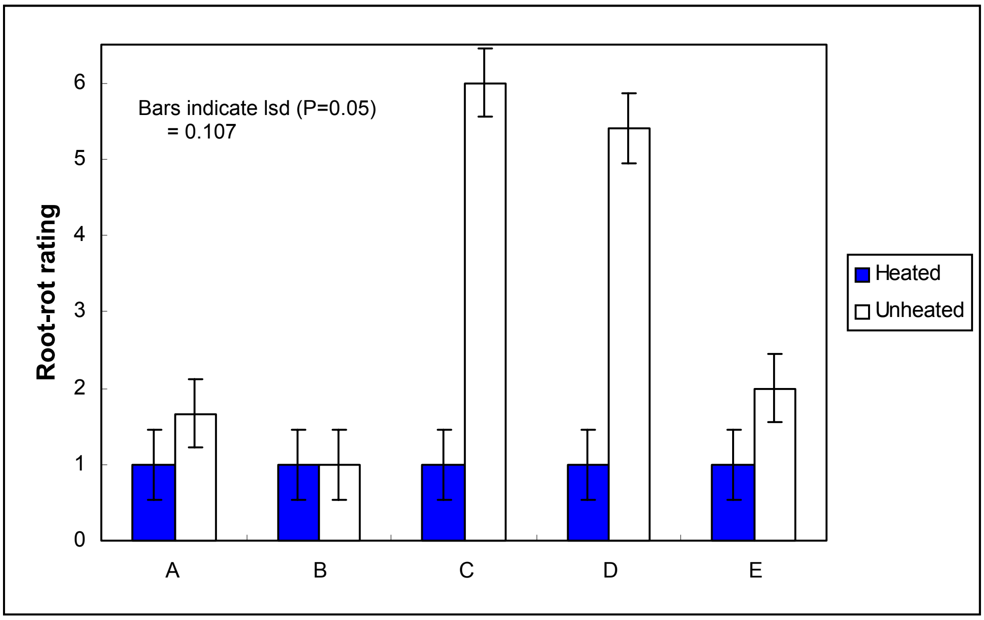
Methods

Soil samples were collected from a field at Bundaberg in April 2003, soon after a forage sorghum crop had been planted. Further samples were collected in June, immediately after the forage sorghum was incorporated as a green manure, and in July, one month after incorporation. Each batch of soil was bioassayed at the time of collection using methods described in Chapter 2, with soil heated to 65ºC for 1 hour used as a control. Additional soil collected in June was sieved and organic matter was either returned to the soil or removed. These samples were stored moist at ambient temperatures until the last sample was collected in July, and then were bioassayed using methods described in Chapter 2.

Results

Plants growing in soil collected 2 months prior to incorporation of the forage sorghum crop or at the time of incorporation displayed little or no root rotting. However, roots in soil collected 1 month after forage sorghum was incorporated were severely rotted (Figure 4.1). Soil stored for 1 month with organic matter removed displayed much less root rotting than soil with organic matter retained (Figure 4.1). Root rotting was not observed in heated soil, indicating that the symptoms were caused by a pathogen. *P. aphanidermatum* was isolated from lesions in rotted roots.

Figure 4.1: Effect of organic matter on root rotting when soil was bioassayed 2 months prior to incorporating forage sorghum (A), at the time of incorporation (B) and 1 month later (C). Results for soil collected at the time of incorporation and stored for 1 month with its organic matter intact (D) or removed (E) are also given.



Effect of green manured forage sorghum on Pythium root rot (Bundaberg 2004)

Methods

The objective of this experiment was to determine whether the severity of Pythium root rot was affected by the time of incorporation of a forage sorghum green manure crop (relative to the date capsicum seedlings were planted). A site near Bundaberg that had been fallowed for 18 months following sugarcane was selected for the experiment and in March 2004 forage sorghum was planted in all plots except those that were to remain fallow. Later, the following treatments were established in five replicate plots each 15 m long: 1. Bare fallow, 2. Forage sorghum ploughed out on 10 June 2004, 3. Forage sorghum ploughed out on 13 July 2004 and 4. Forage sorghum ploughed out on 13 July 2004 after above-ground biomass had been removed. Immediately after the forage sorghum in treatments 3 and 4 was incorporated, soil was collected from treatments 1 and 3 and bioassayed at 36ºC, or at day/night temperatures of 25/36ºC (see Chapter 2). In the field, the soil was formed into beds, black plastic was laid and then the plastic in half the plots was painted white. The field experiment therefore consisted of 2 mulch colours × 4 forage sorghum treatments × 5 replicates. Capsicum seedlings (cv. Tycoon) were planted on 24 September 2004 and the crop was harvested on 13 December 2004.

Results

Root rotting was minimal in standard bioassays of bare fallowed soil and soil amended with forage sorghum. However, when bioassay plants were incubated for 24 hours a day at 36ºC, moderate levels of root rotting caused by *P. aphanidermatum* were observed in both treatments.

In the field, symptoms caused by Pythium root rot were not observed on seedlings growing in any of the mulch colour or green manure treatments. Roots examined after harvest were healthy, and symptoms of root rotting were only seen on a few small roots. Green manure treatments did not affect yield, but plants on white plastic significantly out-yielded plants on black plastic (1.75 kg fruit/plant for white plastic *vs*. 1.50 kg fruit/plant for black plastic).

The impact of sugarcane trash and compost amendments on Pythium root rot

Methods

Field experiment. This experiment was done at Bundaberg in a related project on suppressive soils for root-knot nematode (see Stirling 2005). Treatments included a factorial combination of three cultivation/mulch treatments (1, Beds cultivated and mulched with plastic; 2, Beds not cultivated, forage sorghum removed and mulched with plastic, and 3, Beds not cultivated and mulched with forage sorghum) × two amendment treatments (amended with sugarcane trash plus nitrogen or non-amended).

Details of experimental procedures can be found in Stirling (2005), but briefly, a rotary hoe was used to incorporate 12.5 t/ha of sugarcane trash and 100 kg N/ha (as ammonium nitrate) into soil on 17 September 2003. Non-amended plots were also rotary hoed, and then beds were formed and seed of forage sorghum (cv. Zulu) was drilled into each bed. The forage sorghum crop (13 t of dry matter/ha) was incorporated into cultivated plots on 25 November 2003, whereas it was left standing in non-cultivated plots until it was sprayed with glyphosate on either 22 December 2003 or 6 January 2004. On 8 January 2004, the cultivated plots were rotary hoed, beds were formed and plastic mulch was laid, while on 19 January 2004, the dead forage sorghum in non-cultivated plots was laid on the surface of the bed to form mulch. Capsicum seedlings (cv. Raptor) were planted on 30 January 2004. Soil from amended and non-amended plots was collected on 19 January 2004 and tested for microbial activity using fluorescein diacetate (FDA) hydrolysis (Schnűrer and Rosswall 1982).

Symptoms of Pythium root rot were apparent within a week of planting and so the opportunity was taken to collect data on the effect of treatments on this disease. Disease severity was assessed on 17 February by giving each seedling a plant health rating of 1-3 where 3 = healthy plants showing no symptoms, 2 = smaller plants with no other symptoms, 1 = small wilting, plants with some chlorosis and 0 = plants with almost no leaves, or dead. The number of poor plants was determined by summing the number of plants with ratings of 0 and 1. Pathogens were isolated from the roots of unhealthy plants as previously described.

Glasshouse experiment. An additional treatment (soil amended with compost, sugarcane trash and nitrogen) was available in plots adjacent to the above experiment, and so an experiment was established to determine whether this amendment had increased the soil's suppressiveness to *Pythium*. The treatments included in the experiment were a factorial combination of two cultivation/mulch treatments (beds cultivated and mulched with plastic or not-cultivated and mulched with forage sorghum) × two amendment treatments (amended and non-amended). Experimental procedures were the same as for the previous experiment, except that 2.5 t/ha of compost was incorporated into plots amended with sugarcane trash and nitrogen on 17 September 2003. On 8 January 2004, another 5 t of compost/ha was applied to these plots and then the cultivated plots were rotary hoed, beds were formed and plastic mulch was laid as described above. In non-cultivated plots, the compost remained on the soil surface until 19 January 2004, when dead forage sorghum was laid on the surface of the bed to form mulch as described above.

On 20 January 2004, undisturbed soil cores (10 cm diameter x 21 cm deep) were collected from each of the four treatments. Microbial activity was assessed using the fluorescein diacetate (FDA) hydrolysis technique of Schnűrer and Rosswall (1982). A capsicum seedling (cv. Target) was planted in each core and pairs of cores were either inoculated with 5.5 x 10^5 *P. myriotylum* oospores or left uninoculated. Inoculum was prepared by growing the fungus in a liquid medium for 3 weeks at 25°C (Schmitthenner 1972). Plants were then placed in a closed glass chamber in a glasshouse and after 3 months, roots were rated for root rotting on a scale of 1-6 as previously described.

Results

Field experiment. Pythium root rot was observed in the field within a few days of planting. Roots of dead and dying plants were severely rotted and had stem lesions under the soil line, whereas healthy plants showed little or no root rotting. *P. aphanidermatum* was isolated consistently from diseased roots. Amending soil with sugarcane trash and nitrogen improved plant health ratings and reduced the percentage of plants with severe root rot (Table 4.2). Pythium root rot was particularly severe when black plastic was laid onto non-amended soil without cultivation but much less severe when the same cultivation/mulch treatment was amended. Plots mulched with forage sorghum had fewer poor plants, almost certainly because of lower soil temperatures (Figure 4.2 and Table 4.2). At planting, microbial activity was greater in soil amended with sugarcane trash and nitrogen than it was in non-amended soil (Table 4.2).

Figure 4.2: An example of the difference in the severity of Pythium root rot in capsicum seedlings planted on black plastic (front right) and into an *in situ* mulch of forage sorghum (centre left).

Table 4.2: Effect of organic amendments and cultivation/mulching practice on microbial activity and root-rotting caused by *Pythium aphanidermatum*.

Treatment	Plant health rating (0-3)	Poor plants (%)	Microbial activity at planting (µg FDA/g/min)
Plastic mulch	1.96	34.3	1.10
Forage sorghum mulch	2.31	3.6	0.94
l.s.d. (P=0.05)	n.s.	27.13	n.s.
No amendment	1.8	38.3	0.40
Sugarcane trash + N amendment	2.3	9.9	0.73
l.s.d. (P=0.05)	0.30	15.48	0.064

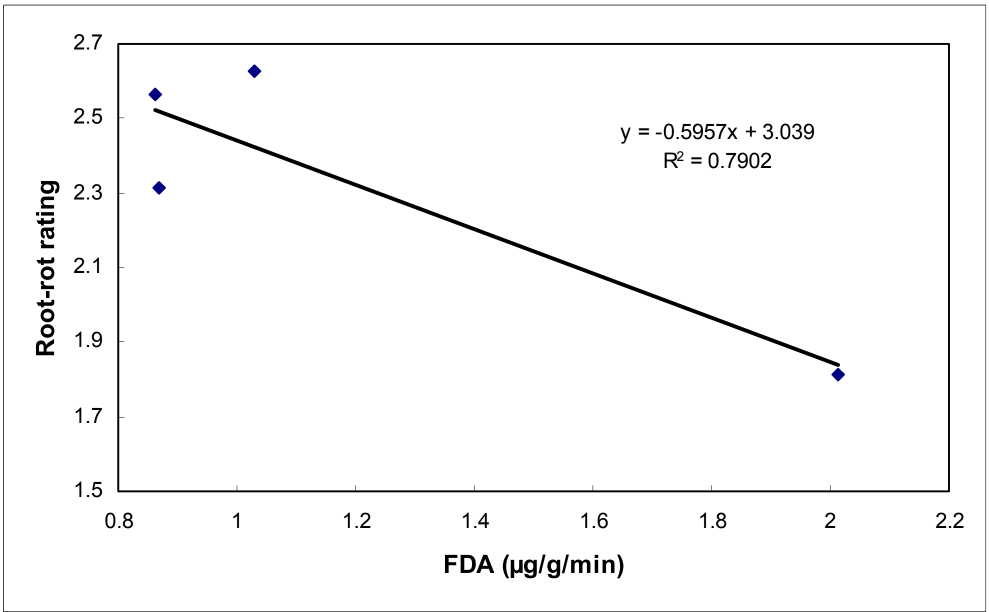
Glasshouse experiment. The average maximum temperature in the plant growth chamber over the course of the experiment was 33.1°C, but temperatures exceeded 35°C for 17 days in the latter part of the growth period. When roots were examined at the end of the experiment, galling caused by root knot nematode was observed, but the level of nematode damage was lower in soil amended with sugarcane trash, nitrogen and compost than in non-amended soil (data not shown). Moderate to high levels of root-rotting were also observed, whether cores were inoculated with *Pythium* or not. *P. aphanidermatum* was isolated from roots in both inoculated and non-inoculated cores, indicating that the pathogen was present in soil at the commencement of the experiment. *P. myriotylum* was isolated only from roots growing in soil into which it had been inoculated.

When data were analysed by analysis of variance, inoculation with *P. myriotylum* did not affect either of the parameters measured. The results presented are therefore the main effects of treatments. Amended but non-disturbed soil stood out from the other three treatments, as root rotting was less severe and microbial activity was much higher than the other treatments (Table 4.3). Regression analysis indicated that the level of root rotting declined as microbial activity increased (Figure 4.3).

Table 4.3. The effect of organic amendments and cultivation/mulching practice on soil microbial activity and root-rotting caused by *Pythium aphanidermatum* and *P. myriotylum*.

Treatment	Root-rot rating		Microbial activity (µg FDA/g/min)	
	Cultivated/ plastic mulch	No till /forage sorghum mulch	Cultivated/ plastic mulch	No till /forage sorghum mulch
No amendment	2.31	2.56	0.87	0.86
Sugarcane trash + N + compost	2.69	1.81	1.03	2.01
l.s.d. (P=0.05)	0.494		0.445	

Figure 4.3: The relationship between the severity of root rotting and microbial activity (measured by FDA hydrolysis) in a bioassay of amended and non-amended soil collected from a field site at Bundaberg.



Development of suppression in an organically-amended soil

Methods

Soil was collected from a field used for vegetable production at Bundaberg and in August 2004, sugarcane trash or lucerne hay was chopped into pieces less than 2 cm long and mixed into the soil at an application rate of 5.36 g/L soil (equivalent to 20 t dry matter/ha incorporated to a depth of 15 cm). Pots were filled with 400 mL of amended or non-amended soil and were then covered with newspaper to minimize drying and incubated at ambient temperatures for 25 weeks. Pots were checked periodically and soil was kept moist by adding 50 mL of water every 10-15 days.

After the incubation period was complete, 5 g soil was taken from three replicate pots of each treatment and microbial activity was measured by FDA hydrolysis. A factorial experiment consisting of three amended soils (nil, sugarcane trash or lucerne) × two heat treatments (heated at 65°C for 1 hour or unheated) × two *Pythium* treatments (non-inoculated or inoculated with *P. myriotylum*) was then established. Inoculum of *P. myriotylum* was grown on corn meal sand

(CMS) for 3 weeks and pots were inoculated by adding 10 g CMS/L soil to four holes in each pot. The inoculum was then washed into the soil with a gentle stream of water. There were five replicate pots of each treatment. Capsicum seedlings were planted in each pot with minimal soil disturbance, and pots were placed in a glass chamber inside a glasshouse, where maximum temperatures reached 36-41°C. Pots were watered every day and the severity of root rotting was assessed after 10 days using the 1-6 scale described previously.

Results

Analysis of variance indicated that all experimental variables significantly affected root rotting. Heating the soil or inoculating it with *P. myriotylum* increased root rot severity, while amending soil with organic matter reduced root rotting (Table 4.4). There were also two significant second-order interactions (*Pythium* × Heat, and Amendment × Heat), and the latter interaction table is presented as Table 4.5. Both amendments reduced the severity of root rotting, while root rotting was more severe when soil was heated, indicating that the suppression was caused by biological factors (Figure 4.4).

Table 4.4: Significant main effects on Pythium root rot of capsicum* in a factorial experiment with three amendments (nil, sugarcane trash or lucerne), two heat treatments (heated at 65°C for 1 hour or unheated) and two *Pythium myriotylum* treatments (non-inoculated or inoculated with *P. myriotylum*).

Amendment	Nil	Sugarcane trash	Lucerne hay	l.s.d. (P=0.05)
	3.2	2.9	2.4	0.51

Pythium	Non-inoculated	Inoculated	l.s.d. (P=0.05)
	1.4	4.2	0.42

Heat	Heated	Unheated	l.s.d. (P=0.05)
	3.3	2.3	0.42

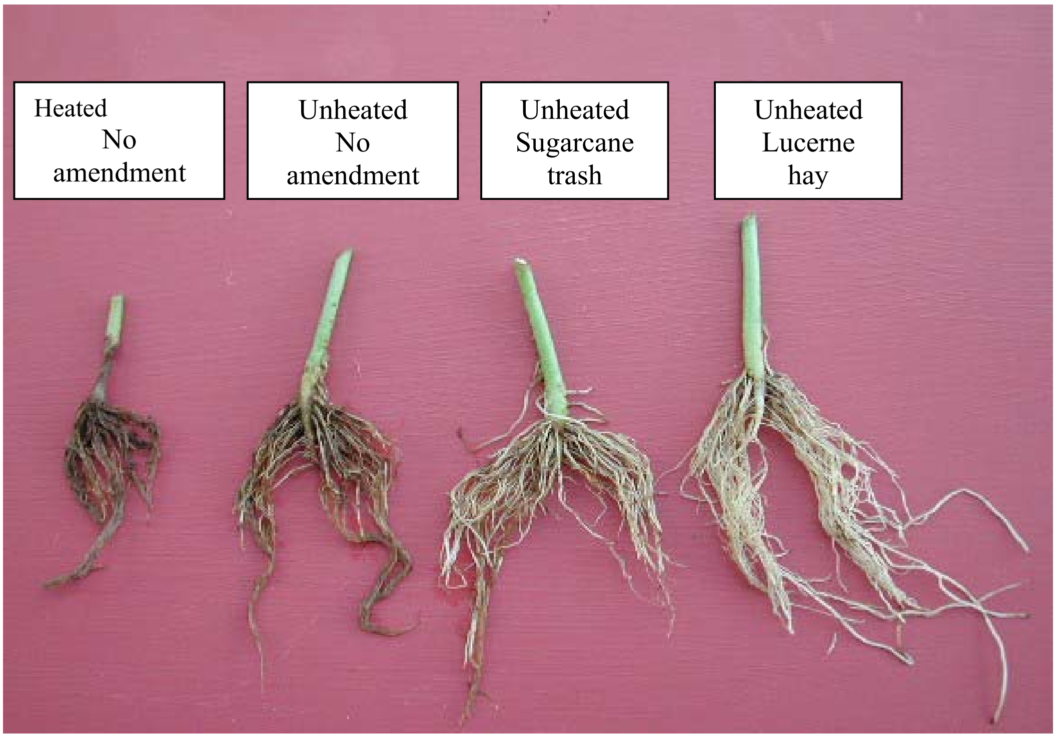
* Values are root rot ratings on bioassay plants after 10 days

Table 4.5: Effects of organic amendments on microbial activity six months after organic matter was incorporated into soil, and effects of organic amendments and heat on Pythium root rot of capsicum* in a factorial experiment with three amendments (nil, sugarcane trash or lucerne), two heat treatments (heated at 65°C for 1 hour or unheated) and two *Pythium myriotylum* treatments (un-inoculated or inoculated with *P. myriotylum*).

Amendment	Root rot rating		Microbial activity (µg FDA/g/min)
	Heated soil	Unheated soil	
Nil	3.2	3.1	0.81
Sugarcane trash	3.5	2.2	1.37
Lucerne hay	3.2	1.6	1.65
l.s.d. (P=0.05)	0.72		0.205

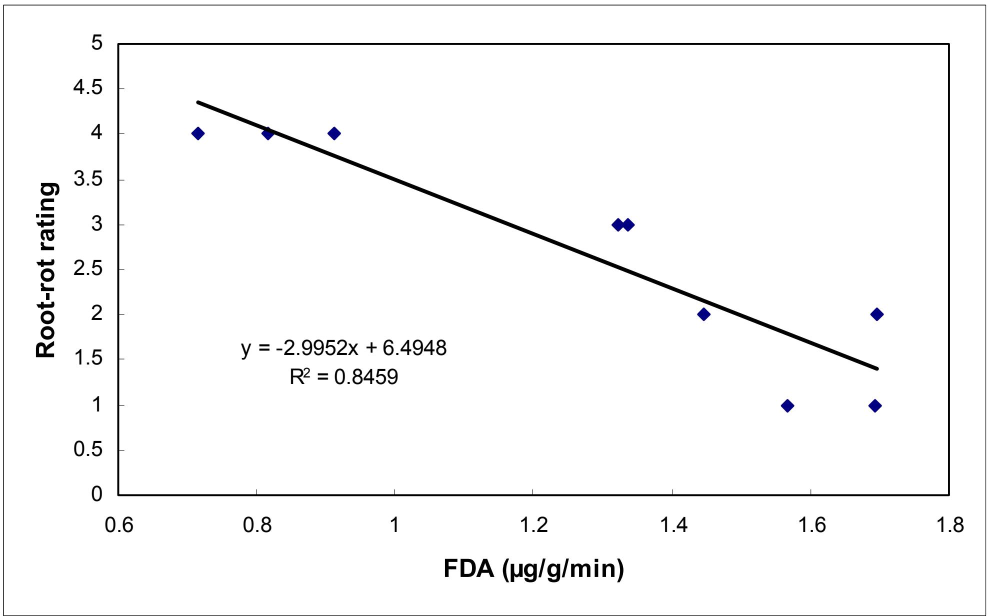
* Values are root rot ratings on bioassay plants after 10 days

Figure 4.4: Roots of capsicum bioassay plants from heated and unheated soil inoculated with *Pythium myriotylum* six months after soil was amended with sugarcane trash or lucerne hay or left unamended.



Addition of either sugarcane trash or lucerne hay increased microbial activity (Table 4.5) and regression analysis showed that root rotting decreased as microbial activity increased (Figure 4.5).

Figure 4.5: Relationship between microbial activity and root rotting in capsicum plants grown for 10 days in amended and non-amended soil.



Discussion

Our results indicate that the addition of fresh forage sorghum tissue to soil can exacerbate Pythium root rot (Figure 4.1). One month after fresh plant material was incorporated into soil, the severity of root rotting in bioassay plants was much greater in amended than non-amended soil. Such an effect was not unexpected, as *Pythium* is known to multiply in soils where an appropriate organic source of nutrients is available. For example, when vetch is incorporated into soil as a green manure or free glucose is present in compost-amended potting mixes, populations of *Pythium* increase and root rotting becomes more severe (Watson 1973, cited by Hoitink and Boehm 1999; Chen *et al.* 1988).

Although green manured plant tissue can enhance Pythium root rot, the practice of green manuring is unlikely to cause major problems in the vegetable industry. *Pythium* is unable to multiply once plant tissue has begun to decompose and is therefore colonised by other microorganisms (Hoitink and Boehm 1999), which means that root rot is only likely to be enhanced for a relatively short period after plant tissue is ploughed in. Most vegetable growers incorporate green manure crops at least one month before laying plastic and do not plant until one or two months later, so there is usually ample time for suppressive microorganisms rather than *Pythium* to become dominant. The failure of forage sorghum to stimulate Pythium root rot on bioassay plants or to affect capsicums grown on black plastic in our experiment at Bundaberg in 2004, confirms that in practice, a green manure crop will normally suppress Pythium root rot or have no effect. Disease enhancement is most likely to occur when crops are planted within a few weeks of fresh organic matter being incorporated.

Amendments of sugarcane trash reduced the severity of Pythium root rot in the field (Table 4.2 and Stirling 2005), while our results with non-disturbed soil cores from the field and with amended soils that were incubated for 6 months in pots also demonstrated the potential of using organic amendments to enhance suppression. Root rotting in bioassay plants grown at temperatures capable of exacerbating Pythium root rot was less severe in soil amended previously with compost and sugarcane trash than in non-amended soil (Table 4.3), while similar effects were observed with lucerne hay and sugarcane trash in another experiment (Table 4.5). Since the bioassays in both experiments were done about 6 months after the amendments were first incorporated into soil, these results show that organically-amended soil remains suppressive to *Pythium* for a relatively long period.

Hoitink and Boehm (1999) noted that the rate of hydrolysis of FDA mimics the level of organic matter decomposition and is therefore related to the potential of potting mixes and organically-amended field soils to suppress root pathogens. It is therefore not surprising that suppression in our experiments was related to microbial activity. However, the level of microbial activity in the suppressive treatments (e.g. non-tilled soil amended with compost and sugarcane trash in one experiment and the lucerne hay and sugarcane trash treatments in another experiment) was much higher than is typical for Bundaberg vegetable-growing soils. This suggests that if suppression is to be achieved in the vegetable industry, major changes will be required in the way soils are managed.

Although compost was a component of a treatment that suppressed Pythium root rot in one experiment at Bundaberg, it had no effect when it was used on its own in a field trial at Ayr in 2003. Since the application rate of compost was approximately the same in both experiments, the dissimilar results may have been due to differences in the quality of the composts. The compost used at Ayr was relatively immature, and *Pythium* may have been able to use it as a food source. It is also possible that the biological status of Queensland's capsicum-growing soils is so poor that additional organic matter (e.g. sugarcane trash) may be needed with compost to raise soil microbial activity to the levels required to achieve suppression.

One unexpected result of this work was the finding that minimum tillage was an important component of the suppression achieved with organic amendments. In one experiment, amended soil was not suppressive to Pythium root rot when it was cultivated twice prior to planting, whereas its non-tilled counterpart was suppressive. Tillage is known to be detrimental to many components of the detritus food web (Wardle 1995), and so it is possible that tillage affected some of the organisms responsible for suppressing the pathogen in our experiment. Further work is needed to confirm this, but in the meantime, the role of tillage in exacerbating Pythium root rot should be further investigated.

CHAPTER 5. THE EFFICACY OF CHEMICALS AGAINST PYTHIUM ROOT ROT

Introduction

This chapter reports the results of experiments on the efficacy of three chemicals (metalaxyl, phosphite and silicon) that have the potential to provide some control of Pythium root rot.

Metalaxyl is mainly used to control root rots caused by *Phytophthora*, but it was chosen for use against *Pythium* because *Phytophthora* and *Pythium* are closely related, and because metalaxyl was effective against *Pythium* in several recent studies (e.g. Hoy and Schneider 1988; Hwang *et al.* 2001; Taylor *et al.* 2002). In Australia, granular formulations of metalaxyl can be applied to soil prior to planting vegetable crops, but post-plant treatments with EC formulations are only registered for use on pineapples.

Phosphites are a group of fungicides that act against both *Pythium* and *Phytophthora* by directly inhibiting the pathogen and eliciting a defence response in the host (Guest and Bompeix 1990). Phosphonic (phosphorous) acid is the phosphite most widely used for root rot control in Australia, and *Phytophthora cinnamomi* is the most common target (Pegg *et al.* 1990). Phosphite was examined in this study because it is sometimes effective against *Pythium* (Utkhede and Smith 1991; Walker 1991).

Silicon was investigated because of its suppressive effects on root rots caused by *Pythium ultimum* and *P. aphanidermatum* in hydroponic vegetable production systems (Chérif and Bélanger 1992; Chérif *et al.* 1994b). Its mechanisms of action are not completely understood, but silicon is thought to induce biochemical changes in the plant that initiate a defence response against the invading pathogen (Chérif *et al*. 1994a).

A. GLASSHOUSE EXPERIMENTS WITH METALAXYL

Effectiveness of pre-plant applications of metalaxyl in controlling Pythium root rot

Methods

Experiment 1. *Pythium*-infested field soil from Ayr was treated with granular or EC formulations of metalaxyl (Ridomil 50G™, containing 50 g metalaxyl/kg, or Ridomil 250EC™, containing 250 g metalaxyl/L, respectively). Both formulations were used at the same application rate (1, 2 and 4 kg of metalaxyl/ha) by applying Ridomil 50G™ at 5.4, 10.7 and 21.4 mg/400 mL soil and Ridomil 250EC™ at 1.1, 2.2 and 4.4 µL/400 mL soil. Assuming, even incorporation to a depth of 15 cm this is equivalent to 1, 2 and 4 kg of metalaxyl/ha. Treated and untreated soil was then added to 400 mL pots without drainage holes and capsicum seedlings were planted 0, 2 and 4 weeks after the metalaxyl was applied. Root rotting was assessed using procedures described in Chapter 2.

Experiment 2. The metalaxyl treatments in this experiment were the same as in experiment 1, but potting mix inoculated with *P. myriotylum* or left un-inoculated was used instead of field soil. The inoculum was grown in Schmitthenner's liquid medium (Schmitthenner 1972) and oospores (10^3 spores/g soil) were mixed into the potting mix one day before capsicum seedlings were planted. Plants were bioassayed in the same way as in experiment 1.

Results

Experiment 1. Both formulations of metalaxyl decreased the severity of root rotting in field soil (Table 5.1). Efficacy improved marginally as the rate of metalaxyl increased and the fungicide remained effective for at least 4 weeks after it was applied.

Table 5.1. The severity of root-rotting in capsicum seedlings planted 0, 2 or 4 weeks after different formulations of metalaxyl were incorporated into *Pythium*-infested field soil from Ayr.

Treatment	Root rot rating		
	0 weeks	2 weeks	4 weeks
Nil	4.8	4.0	4.3
Metalaxyl granules 1 kg/ha	2.3	1.3	2.3
Metalaxyl granules 2 kg/ha	1.5	1.3	1.7
Metalaxyl granules 4 kg/ha	1.3	1.0	1.3
Metalaxyl EC 1 kg/ha	2.0	1.7	2.0
Metalaxyl EC 2 kg/ha	1.8	1.3	2.0
Metalaxyl EC 4kg/ha	1.3	1.3	1.7
l.s.d. ($P = 0.05$)	0.95	1.08	0.86

Experiment 2. Metalaxyl was effective against Pythium root rot in potting mix, but the granular formulation gave better results than the EC formulation at 0 and 4 weeks (Table 5.2). However, efficacy declined more quickly than it did in field soil, with the chemical being much less efficacious in potting mix at 4 weeks than it was after 0 and 2 weeks. An application rate of 1 kg/ha gave good control, but sometimes efficacy was slightly better at higher application rates (Table 5.2).

Table 5.2: The severity of root rotting in capsicum seedlings planted 0, 2 or 4 weeks after different formulations of metalaxyl were incorporated into potting mix that was either inoculated with *Pythium myriotylum* (10^3 oospores/g of soil) or left un-inoculated.

Treatment	Root-rot rating		
	0 weeks	2 weeks	4 weeks
Control (no *Pythium* or fungicide)	1.0	1.0	1.0
Control (no *Pythium* + metalaxyl [4 kg/ha])	1.0	1.0	1.0
Control (*Pythium*, no fungicide)	4.3	4.3	4.3
Metalaxyl granules 1 kg/ha	1.5	1.0	2.3
Metalaxyl granules 2 kg/ha	1.0	1.0	2.7
Metalaxyl granules 4 kg/ha	1.0	1.0	1.7
Metalaxyl 250 EC 1 kg/ha	2.8	1.0	3.3
Metalaxyl 250 EC 2 kg/ha	2.0	1.7	3.0
Metalaxyl 250 EC 4 kg/ha	1.0	1.0	2.3
l.s.d. ($P = 0.05$)	0.64	0.44	0.76

Application of metalaxyl to seedling trays prior to transplanting seedlings

Methods

A soil drench of Metalaxyl 250EC™ (250 g metalaxyl/L) was applied to eight-week-old capsicum seedlings growing in potting mix. On the basis of estimates of the amount of drench absorbed by the potting mix, it was assumed that 0.0125 g of metalaxyl remained in every litre of potting mix. The following day, treated and untreated seedlings were planted into potting mix inoculated with *P. myriotylum* on corn meal sand (10 g inoculum/L potting mix). Seedlings were transplanted into five replicates of three different types of container: 400 mL closed cups, 400 mL pots and 1.5 L pots. Untreated seedlings planted into either inoculated or un-inoculated potting mix served as controls. Three days after planting, pots were transferred to a chamber where the temperature reached 36-38°C for about 8 hours during the day and then declined to the mid 20's at night. Plants were harvested after 2 weeks and checked for root rotting as previously described.

Results

Drenching metalaxyl into seedling potting mix immediately before transplanting reduced root rotting after seedlings were transplanted (Table 5.3). The effect was apparent in all types of containers, suggesting that the leaching of irrigation water through pots with drainage holes did not reduce efficacy.

Table 5.3: The severity of root rotting in capsicum seedlings pre-treated with an EC formulation of metalaxyl and then transplanted into different types of container filled with potting mix previously inoculated or not inoculated with *Pythium myriotylum*.

	Root rot rating		
Type of container	Metalaxyl-treated seedlings in non-inoculated potting mix	Untreated seedlings in *P. myriotylum*-infested potting mix	Metalaxyl-treated seedlings in *P. myriotylum*-infested potting mix
Closed 400 mL cup	1.0	5.2	4.2
400 mL pot	1.0	4.6	2.4
1.5 L pot	1.0	4.6	3.4
l.s.d. (P=0.05)		0.63	

B. EXPERIMENTS WITH PHOSPHITE

The effect of phosphite as a drench or spray

Methods

<u>Experiment 1.</u> The efficacy of phosphite as a post-plant drench or spray was examined by assessing its capacity to reduce root rot in 400 mL of potting mix inoculated with oospores of *P. myriotylum* (10^3 spores/g soil). Capsicum seedlings were planted in inoculated potting mix, pots were transferred to a water bath maintained at 28°C and then phosphite (Anti Rot™ [Yates Pty. Ltd] containing 200 g phosphite/L) was drenched onto soil or sprayed onto the foliage of five replicate pots. One set of plants was drenched or sprayed at planting, another set 2 weeks later, and a third set was treated at both times. Drench application rates were 0, 0.27, 0.54 and 1.08 µL phosphite/400 mL soil (equivalent to 0, 1, 2 and 4 kg phosphite/ha, assuming even incorporation

to a depth of 15 cm). Plants were sprayed with a solution containing 1 g of phosphite and 0.2 mL Tween per L of water. Three days after the last application of phosphite, bioassay plants were treated as described in Chapter 2.

Experiment 2. Phosphite (1, 2 and 4 mL/L water) was drenched onto capsicum seedlings growing in potting mix in seedling trays prior to transplanting. One set of plants was drenched 2 weeks prior to transplanting, another set 1 week prior to transplanting and a third set was treated at both times. Seedlings were transplanted into potting mix inoculated with *P. myriotylum* (10^3 oospores/g of soil). There were five replicates of each treatment and untreated plants were used as controls. Bioassay conditions were the same as in the previous experiment.

Results

Experiment 1. Phosphite did not reduce root rotting when it was drenched onto plants growing in soil that had previously been inoculated with *Pythium*. However, a post-plant spray at 1 g/L was partially effective (Table 5.4).

Table 5.4: Effect of phosphite on root rot of capsicums when the chemical was drenched or sprayed onto plants growing in soil infested by *Pythium myriotylum*.

Amount of phosphite and application method	Root-rot rating		
	One application (at planting)	One application (2 weeks after planting)	Two applications (0 and 2 weeks)
1 kg/ha (drench)	4.5	4.3	3.9
2 kg/ha (drench)	4.3	4.8	4.8
4 kg/ha (drench)	4.1	4.2	4.0
1 g/L (spray)	2.8	3.1	2.8
No phosphite (+ *Pythium)*		4.3	
No phosphite (- *Pythium*)		1.0	
l.s.d. (P=0.05)		1.04	

Experiment 2. The severity of Pythium root rot was reduced when seedlings were treated with phosphite (1 g/L) prior to transplanting into soil inoculated with *P. myriotylum* (Table 5.5). However, higher concentrations of phosphite (2 and 4 g/L) were phytotoxic.

Table 5.5. Effect of one or two drenches of phosphite (1 g/L) applied prior to inoculation with *Pythium myriotylum*, on Pythium root rot of capsicum.

Treatment	Root-rot rating
One application of phosphite (1 week before inoculation)	2.2
One application of phosphite (2 weeks before inoculation)	2.2
Two applications of phosphite (1 and 2 weeks before inoculation)	1.6
No phosphite	3.6
No phosphite in non-inoculated soil	1.0
l.s.d. (P=0.05)	0.59

Phosphite concentrations in plant roots

Methods

Capsicum seedlings (cv. Raptor) were planted in a field trial at Bundaberg on 12 September 2003 into 9 m beds covered with black or white plastic. A solution of phosphite (1 g/L) and Tween (0.2 g/L) was sprayed on capsicum foliage to the point of run-off 1, 3, 5, 7 and 9 weeks after planting. Five replicate plots were sprayed and a similar number were left unsprayed for each plastic colour. Two weeks after each application (except week 1), one randomly selected control plant and two phosphite-treated plants were harvested from each treatment. Roots were washed, blotted dry and frozen, and once all samples had been collected they were air-dried at 36°C. Dried roots were forwarded to the Chemistry Centre, Western Australian Government, Perth, WA, and phosphite concentrations in roots were determined by flame photometric detection of the methyl ester with gas-liquid chromatography.

Results

The concentration of phosphite in unsprayed plants was less than 1.2 μg/g. Regardless of the number of times plants were sprayed, levels of phosphite in the roots of unsprayed plants ranged from 27 to 33 μg/g. Disease pressure was low and differences in root rotting or fruit yield between sprayed and unsprayed plants were not observed.

C. GLASSHOUSE EXPERIMENTS WITH SILICON

Effect of silicon as a soil drench

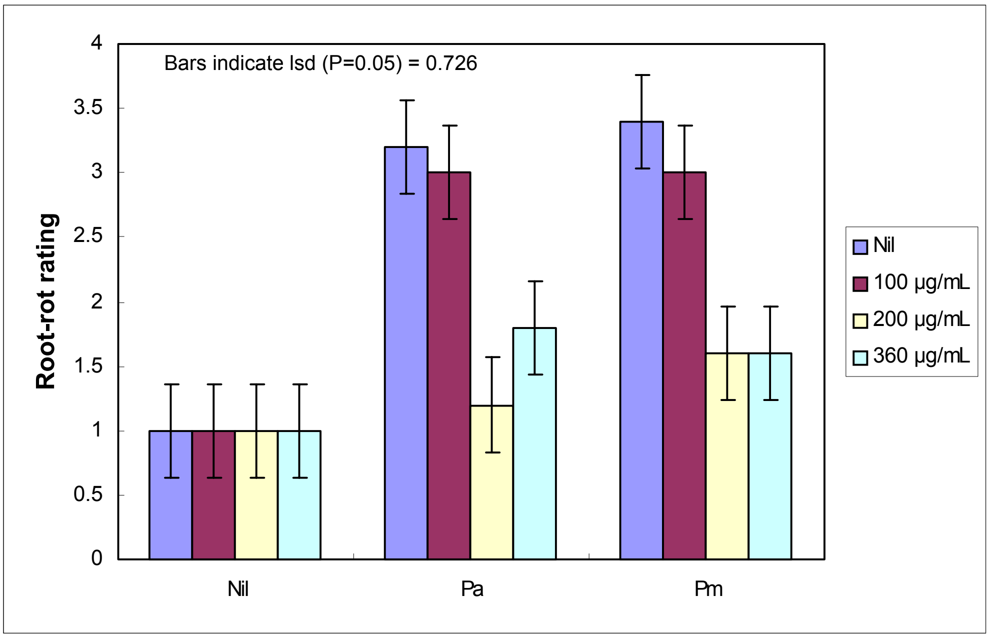
Methods

Kasil 2040™ (~6 M SiO_2) supplied by PQ Australia Pty. Ltd. was diluted with water (100, 200, 360 and 1800 μg SiO_2/mL water) and drenched onto capsicums in seedling trays. One and 2 weeks after seedlings were treated, they were transplanted into potting mix inoculated with either *P. myriotylum* or *P. aphanidermatum* (10^3 oospores/g soil). Pots were then transferred to a water bath and plants were irrigated as required by adding the SiO_2 solution previously applied to seedlings. The effects of treatments on root rot were assessed using the bioassay procedure described in Chapter 2.

Results

SiO_2 did not affect root rotting at 100 μg/mL but reduced the impact of both *P. aphanidermatum* and *P. myriotylum* at 200 and 360 μg/mL (Figure 5.1). Plants treated with silicon at 1800 μg/mL appeared normal, but roots were brown and distorted, indicating that the chemical was phytotoxic at this application rate.

Figure 5.1: Effect of SiO_2 at three application rates on the severity of root rotting caused by *Pythium aphanidermatum* (Pa) and *P. myriotylum* (Pm) on capsicum seedlings.



Comparison of silicon drenches and sprays

Methods

Capsicum seedlings grown in seedling trays for 8 weeks were transplanted into 400 mL pots filled with either pasteurised potting mix or potting mix previously inoculated with *P. myriotylum* (4 g of colonised CMS/pot). SiO_2 treatments were applied by diluting Kasil 2040™ with water and either drenching the solution onto potting mix or spraying it on foliage. The pre-plant drench consisted of 360 µg SiO_2 per mL water and was applied to seedling trays 2 and 1 weeks before seedlings were transplanted. The post-plant drench was applied by adding 80 mL of a solution containing 360 µg SiO_2 per mL water to pots 2 days after seedlings were transplanted. The foliar spray was also applied 2 days after seedlings were transplanted by spraying foliage to run-off with a solution of 1000 µg SiO_2 per mL water. Each treatment was replicated five times, with all treatments included individually and in combination. There was also an untreated control and a control consisting of plants growing in non-inoculated soil. Soil temperature was maintained at

28ºC until the treatment schedule was completed, and then treatments were compared using the bioassay described in Chapter 2.

Results

SiO_2 reduced the severity of root rotting when drenched onto soil after seedlings were planted, whether the drench was applied on its own or with a pre-plant drench or foliar spray. The pre-plant drench and foliar spray had no effect (Table 5.6).

Table 5.6: The effect of foliar sprays and pre- and post-plant drenches of SiO_2 on the severity of root rotting caused by *Pythium myriotylum* on capsicum seedlings.

Treatment	Root rot rating	
	Control	*P. myriotylum*
Control	1.0	4.6
Pre-plant drench	1.0	4.4
Post- plant drench	1.0	2.6
Post-plant spray	1.0	4.6
Pre- & post- plant drenches	1.0	2.4
Pre-plant drench & post–plant spray	1.0	4.0
Post-plant drench & spray	1.0	2.6
l.s.d. (P=0.05)	0.45	

Silicon levels in roots

Methods

The effect of various methods of applying SiO_2 on Si concentrations in roots and foliage was determined by diluting Kasil 2040™ with water and applying the solution as a drench or foliar spray to 8-week-old capsicum plants growing in water-tight pots filled with 400 mL of pasteurised potting mix. The drench consisted of 80 mL of a solution containing 360 µg SiO_2/mL water, while foliage was sprayed with 1000 µg SiO_2/mL water. The eight silicon treatments consisted of a drench or spray at either 20, 10 and 5 days (A); 10 and 5 days (B); 5 days (C) or 2 days (D) prior to harvest. Each treatment was replicated five times (three plants/replicate), and there was also an untreated control. When plants were harvested, roots were washed free of potting mix and the organic silicon content in both shoots and roots was determined by Crop Tech Laboratories, Bundaberg, Queensland.

Results

Application of silicon as a foliar spray did not increase concentrations of organic silicon in root or shoot tissue (Table 5.7). Drenching was far more effective, with silicon concentrations in roots and shoots increasing significantly compared with the untreated control. Drenches within 2-5 days of harvest had the greatest impact on silicon concentrations in shoots.

Table 5.7: Silicon concentrations in sap extracted from capsicum shoots and roots after plants were treated with silicon by drenching or spraying.

Treatment	No. days prior to harvest when treatment applied	Silicon (SiO_2) extracted (µg/mL)	
		Shoots	Roots
Control	-	8.4	3.8
Drench (3 times)	20, 10 and 5	11.5	7.0
Drench (2 times)	10 and 5	10.5	7.4
Drench (once)	5	13.1	7.3
Drench (once)	2	16.7	7.2
Spray (3 times)	20, 10 and 5	8.5	3.6
Spray (2 times)	10 and 5	7.2	5.1
Spray (once)	5	8.7	5.6
Spray (once)	2	10.2	4.7
l.s.d. (P = 0.05)		3.85	2.24

D. FIELD EXPERIMENTS WITH CHEMICALS

Effect of metalaxyl and phosphite on root rot of seedlings at Ayr in 2004

Methods

Details of a factorial experiment consisting of 2 mulch colours (black and white) × 2 planting distances (21 and 27.5 cm) × 4 chemicals were presented in Chapter 3. The effect of mulch colour and planting distance components were discussed previously, but details of the chemical component are presented here.

The two chemicals applied in the experiment were metalaxyl (Ridomil 50G), which was incorporated into soil before the crop was planted at a rate of 2 kg/treated ha, and phosphite (Anti Rot™ [Yates Pty. Ltd] containing 200 g phosphite/L), which was sprayed onto plants 1, 3, 5 and 7 weeks after planting at 1 g/L. Each chemical was applied alone and in combination with the other, while the fourth treatment was an untreated control. Effects of treatments were compared by counting the number of dead and dying plants after one week, and by measuring yield three and four months after planting.

Results

When the seedling mortality and yield data obtained from the experiment were analysed as a factorial consisting of 2 plastic colours × 2 planting distances × 4 chemical treatments, the chemical effect was not significant. However, re-analysis using 2 plastic colours × 2 planting distances × +/- metalaxyl × +/- phosphite resulted in a significant effect of metalaxyl (P =0.05). One week after planting, 21.6% of plants had died in the plots that were not treated with metalaxyl compared with only 14.1% of plants in metalaxyl-treated plots (Table 5.8). When the crop was harvested, metalaxyl-treated plots also yielded more fruit than non-treated plots. Phosphite treatment did not affect seedling mortality or fruit yield.

Table 5.8: Effect of metalaxyl on seedling mortality and yield of a capsicum crop planted at Ayr in March 2004.

Treatment	Seedling mortality (%)	Fruit yield (kg/metre of row)
No metalaxyl	21.6	4.01
Metalaxyl	14.1	4.51
l.s.d. (P = 0.05)	6.42	0.367

Efficacy of metalaxyl, phosphite and silicon at Bundaberg in 2004

Methods

This experiment was done on a grower's property at Bundaberg during the spring of 2004. Black plastic was laid on beds in July and a single row of capsicum seedlings was planted on 24 September 2004. Five chemical treatments were then applied to five replicate plots 12 m long. Metalaxyl (2 kg/treated ha) was applied through the irrigation system 5 or 7 weeks after planting, with Ridomil 250EC™ used at 5 weeks and Metalaxyl 250EC™ used at 7 weeks. SiO_2 (108 kg/treated ha) was applied through the irrigation system (as Kasil 2040™) at 5, 7 and 9 weeks after planting or SiO_2 (1000 µg/mL) was sprayed on foliage 5, 7 and 9 weeks after planting. The fifth treatment was and an untreated control. Phosphite (1 g/L) was applied in a split-plot design to half of each plot by spraying Anti Rot™ (200 g phosphite/L) onto plants at 5, 7 and 9 weeks.

Results

Pythium root rot was not observed in this trial and differences in yield due to treatment were not obtained. Metalaxyl applied 5 weeks after planting caused a leaf burn that was apparent for about 4 weeks. However, these leaf symptoms were no longer apparent at 12 weeks and the treatment did not have a detrimental effect on yield. Phytotoxicity was not observed when metalaxyl was applied at 7 weeks.

Discussion

The results of our studies in pots and the field indicated that metalaxyl reduced losses from Pythium root rot in capsicum seedlings. Granular or EC formulations applied prior to planting gave satisfactory control for at least four weeks. However, the problem with using pre-plant treatments in the vegetable industry is that they can only be applied at the time beds are prepared, and this often occurs 2-3 months before planting. Since metalaxyl is subject to biological degradation in soil (Droby and Coffey 1991), there is therefore potential for it to lose effectiveness by the time seedlings are planted. Also, efficacy may be lost due to enhanced microbial degradation if it is routinely used for root rot control.

The strategy most likely to work against the seedling stage of the disease is to apply metalaxyl to potting mix just before seedlings are planted, or to drench the chemical around seedlings as they are planted. Our results indicate that both these strategies are effective. Nurseries would have to apply metalaxyl to potting media, but growers could easily apply metalaxyl to transplants by adding it to the water that is drenched around seedlings when they are planted. However, the only metalaxyl formulations currently registered for use on vegetable crops are granules that must be applied to soil as a pre-plant treatment. Registration of an EC formulation that can be drenched onto seedlings before or at planting or applied via the irrigation system soon after planting would provide growers with a range of options for controlling Pythium root rot in vegetable seedlings.

When metalaxyl is applied before or at planting, it is unlikely that its residual activity would be good enough to prevent sudden wilt of capsicum, as the root rotting associated with sudden wilt occurs at the fruit-fill stage, and this is usually 6-10 weeks after planting. We therefore tested an EC formulation that could be added to irrigation water and applied through the trickle irrigation system prior to fruit-fill. Unfortunately, sudden wilt did not occur in our trials and so further work on the effectiveness of this approach will be required. When this work is done, some adjustments to our application rates and application schedule may have to be made, as slight phytotoxicity was observed when metalaxyl was applied to 5 week-old capsicums. Residue data will also have to be collected to confirm that the current 7-day withholding period for pre-plant applications is satisfactory for post-plant treatments.

Our results with phosphite were encouraging, as phosphite reduced root rotting caused by high-temperature *Pythium* species that are widespread on vegetable crops in tropical and subtropical environments. However, the effects of phosphite were relatively subtle, as root rotting was not reduced to the same extent as for metalaxyl. Root health improved when soil was drenched with phosphite, but the chemical had to be applied early so that roots could take it up before they were attacked by the pathogen. Foliar sprays will probably be more practical in the vegetable industry, and our results suggest that they are likely to provide useful control. In pots, for example, a single phosphite spray reduced the number of rotted roots on capsicum seedlings by about a third.

It is not possible to predict from our results whether phosphite will prevent the root rotting that is associated with sudden wilt. However, data from our field experiment at Bundaberg indicate that phosphite is translocated to the roots of mature plants when it is sprayed on foliage, as root phosphite concentrations ranged from 27-33 µg/g, depending on the number of sprays applied. Since previous work with *P. cinnamomi* on avocado has shown that root rotting is reduced at phosphite concentrations of 20-30 µg/g root (Whiley *et al.* 1995), it is possible that phosphite sprays will have a similar effect on *Pythium*, which is closely related to *Phytophthora*. However, it is likely that efficacy will vary, as environmental conditions and the level of root activity affect phosphite concentrations in roots (K. Pegg, pers. comm.). Given the unpredictable nature of sudden wilt and the strong environment × pathogen interactions involved in the disease, it will also be difficult to obtain the data required to confirm that phosphite is a useful control measure.

We obtained evidence from two pot experiments to show that the severity of Pythium root rot is reduced when SiO_2 is drenched around the roots of capsicum seedlings. However, these results should be treated with caution because limited data are available on the effects of silicon on root diseases in the field. One reason for caution is that silicon is relatively mobile in soil, and may be easily leached below the root zone under field conditions. Forms of silicon capable of affecting *Pythium* are also relatively expensive, and so application rates and the number of times that silicon can be applied will be limited by economics.

We conclude that strategic applications of the three chemicals used in this study are likely to be useful against both the seedling and sudden wilt components of the Pythium root rot syndrome. Metalaxyl will probably have the greatest impact, but registration is required for EC formulations that can be applied to seedling trays in the nursery, drenched around seedlings at the time they are transplanted or applied via irrigation water within a few weeks of planting. Phosphite is also likely to be useful, as the chemical is relatively non-toxic and is easy to apply as a foliar spray. When used in this manner on capsicums, phosphite is translocated to the root system at concentrations that should be sufficient to reduce root rotting. Drenches of SiO_2 are also worthy of further investigation, as the chemical is safe to use and initial results show that it reduces the severity of Pythium root rot in pots. Future research should aim to define the application rates and treatment schedule required to achieve satisfactory root rot control with silicon, and confirm that it is economically feasible to apply it to vegetable crops.

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