Project title:	**Screening candidate fungicides for the control of the *Pythium* spp. which cause cavity spot disease**
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PRACTICAL SECTION FOR GROWERS

Objectives and background

Cavity spot is the most important disease of carrots in the UK. *Pythium violae* is the cause of most outbreaks, while *Pythium sulcatum* is responsible for localised outbreaks. *P. violae* can be controlled by metalaxyl, whereas *P. sulcatum* is less sensitive to this fungicide. Since the early 1980's, the control of cavity spot has been possible using Fubol 58 WP, where the causal organism was *P. violae*. The active ingredients of Fubol 58 WP are metalaxyl (10% w/w) and mancozeb (48% w/w). The cost of this treatment is considered by growers to be expensive, especially as the formulation contains mancozeb, which contributes little to the control of cavity spot. The long-term environmental impact of mancozeb is not known. Furthermore, there have been reports of reduction in the efficacy of metalaxyl. It is therefore important to look for alternatives to metalaxyl which could either replace or be used alongside it. Secondly, as *P. sulcatum* is not very sensitive to metalaxyl, the availability of an effective fungicide against *P. sulcatum* would be valuable in situations where this pathogen predominates.

The objective for the first year of the project was to assess the fungicides currently commercially available, or likely to become available, for activity against the *Pythium* species which cause cavity spot of carrot. The tests made comprised those in the laboratory in which *P. violae* and *P. sulcatum* were included in plate assays with the fungicides. Glasshouse tests were also carried out in which carrots were grown in pots of compost inoculated with the pathogens or field soil and then sprayed with the range of fungicides and compared with the commercial standard metalaxyl.

In the second year the efficacies of those products that gave promising results in the first year were further evaluated in pot experiments using field soils.

Summary of results

Results from the screening of fungicides in plate tests confirmed earlier findings with metalaxyl for *Pythium violae* and *Pythium sulcatum*. The former was highly sensitive (ED_{50} 0.48 μg/ml) to the fungicide, whilst the latter was relatively resistant (ED_{50} 4.53 μg/ml). The fungicide coded A9408B was more active against *P. violae* than metalaxyl, giving an ED_{50} of 0.06 μg/ml. The ED_{50} for *P. sulcatum* was 1.54, whereas with metalaxyl it was 4.53. A9180A, which is known to be inactive *in vitro* showed no sign of activity against either of the species. Hymexazole gave variable, but poor results for *P. violae*, but the ED_{50} for *P sulcatum*, at 0.08 μg/ml, was particularly low. It appears unlikely that this observation correctly indicates the likely efficacy of hymexazole against *P. sulcatum*. ICIA5504 also gave an ED_{50} of 0.08 μg/ml for *P. sulcatum* and in this case the curve produced (Appendix 5, Figure 6) does indicate a high level of activity against the fungus. Of the other fungicides tested, ED_{50}'s obtained indicated moderate, or no activity against the two fungi, and significant beneficial effects in soil tests would not be expected.

In the tests in which carrots were grown in pots containing field soil infested with *P. violae*, 65.6% of roots from the untreated controls had cavity spot at the first harvest, and 83.3% had lesions at the second harvest. At the first harvest the commercial standard metalaxyl reduced percentage infection to 21.1, and three other treatments (A9180A, dimethomorph and ICIA5504, all at half rate) gave reductions in disease which were not significantly different from that value. By the second harvest, the percentage of roots with lesions in the metalaxyl treatment had risen to 34.2%. With all other treatments there was significantly more disease ($P < 0.05$).

At the first harvest the distribution of small and large cavities (< 5 mm or > 5 mm) was relatively even. By the second harvest, in addition to a general increase in number of roots with lesions, the size distribution was heavily skewed towards the large size grade.

None of the candidate fungicides appear to have activity in soil with high inoculum pressure from *P. violae* equivalent to that of metalaxyl. Some effects *in vitro* appear to indicate high level activity against *P. violae* or *P. sulcatum*. These observations indicated the need for further study with the fungicides concerned.

The second year work comprised a series of pot experiments examining different aspects of control of the disease. Two experiments were with fields soils known to produce cavity spot caused by *P. violae*. In soil 1, over four harvests Fubol 58 WP applied at the commercial rate and A9408B at 1 X, 2 X and 4 X that rate reduced cavity spot as measured by three parameters. However, most reductions were at levels which would not be considered commercially significant. It was only with the highest rate of A9408B that disease was reduced to 40 % of that in the untreated control pots.

In contrast to findings in year 1, there was no evidence that disease control from A9408B reduced with successive harvests.

These trials were carried out in conditions favouring the pathogen, and with a highly susceptible carrot cultivar. Disease control would predictably have been better with a cultivar with a higher level of field resistance.

In two other pots experiments the Zeneca product ICIA5504 was confirmed as active against *P. sulcatum*, but the fungicide had no effect where *P. violae* was the causal agent.

Action points for growers

- Be aware of the likely risk of cavity spot of fields in which you intend to grow carrots. Use the cavity spot diagnostic test.

- Where fungicide is appropriate use Fubol 58 WP in accordance with the label recommendation.

- Where disease risk is classified as high do not grow carrot in the coming year. Re-test those high risk fields in subsequent years because levels may have fallen.

- Read the review of World cavity spot literature (Final Report for HDC Project FV 5e) and consider adopting some of the agronomic practices which have been shown to reduce disease levels. It is increasingly important to think in terms of managing disease levels down.

Practical and financial benefits from study

The work has shown that there are currently no fungicides in the market place which would be expected to be more effective in controlling cavity spot than Fubol 58 WP. The laboratory studies in which fungi were grown on agar plates amended with fungicides confirmed that these findings have a rational basis. Metalaxyl is a more effective molecule in controlling *P. violae* than are the other available fungicides. In comparison with the earliest studies on control of cavity spot with metalaxyl fungicides, it would appear that degree of control is falling, and this is a concern for future years.

A new formulation of metalaxyl is to be released for use on carrots. This study does not indicate that the new product will improve disease control. In the first year of the project there was evidence of control falling off as compared with standard metalaxyl as from the first to last harvest. This was not seen in the second year trial with one soil, but in the second experiment in year 2 the fungicide appeared to have no effect even at rates well in excess of the projected commercial application rate. Because it was confirmed that the *Pythium violae* isolates from the field soil were highly sensitive to metalaxyl, we now have first-hand evidence backing up commercial observations of failure of the fungicide to control the disease.

With the Zeneca fungicide ICIA5504 there is a prospect for controlling outbreaks of cavity spot where the causal agent is *Pythium sulcatum*. This will depend on the company progressing the chemical for this use.

The major benefit of the study is to remove uncertainty of growers on the possibility that fungicides other than Fubol 58 WP might give better control of cavity spot. For the researchers, the work gives very clear guidelines for the work needed in the prospective Horticulture Link programme (see Final Report to HDC Project FV 5e; A review of literature on cavity spot disease).

EXPERIMENTAL SECTION

A. GENERAL INTRODUCTION

Cavity spot is the major disease of carrots in temperate countries (White, 1986, 1988). It was first noted in Eastern England in the 1960's (Baker, 1972) and has since then been reported from all the main carrot growing areas in the UK. Currently it is the major disease problem for UK carrot growers. Cavity spot of carrot is caused by slow-growing members of the genus *Pythium*. *P. violae* is the most common cause, while *P. sulcatum* is responsible for localised outbreaks.

In the beginning of the 1980's, metalaxyl was found to reduce cavity spot in glasshouse tests in Norway (Lyshol, Semb & Taksdal, 1984), and these results were later confirmed for field-grown carrots (Gladders & Crompton, 1984; Perry & Groom, 1984). Since then metalaxyl has been routinely used for control of cavity spot in several countries. Currently Fubol 58 WP the active ingredients of which are metalaxyl and mancozeb is widely used for control of cavity spot in the UK. However, the mancozeb component of the product plays little or no part in the control of *P. violae* (Gladders & McPherson, 1986; White, Wakeham & Petch, 1992; McPherson, 1995), and has been demonstrated to adversely affect beneficial organisms in the soil (White *et al.*, 1992). Over the past ten years there have been reports of metalaxyl failing to control cavity spot (Gladders & McPherson, 1986; McPherson, 1995). This reduction in the efficacy of metalaxyl could be due to a number of reasons including high inoculum levels, the development of resistance to the fungicide by the *Pythium* spp. responsible for causing cavity spot, enhanced biodegradation of the fungicide with repeated use in fields where cavity spot occurs or other fates (binding to organic matter, leaching etc.) in different soil types.

P. sulcatum is less sensitive to metalaxyl than *P. violae* (White, Stanghellini & Ayoubi, 1988), and there has been a report of metalaxyl failing to control cavity spot in a field where *P. sulcatum* was identified as the primary pathogen involved in cavity spot development (White, 1988).

B. YEAR 1 (1995-1996) - SCREENING OF FUNGICIDES AGAINST THE PATHOGENS WHICH CAUSE CAVITY SPOT OF CARROT

Introduction

Control of cavity spot at present relies on the application of Fubol 58 WP which is expensive and has for some time been considered by carrot growers to be giving less than adequate control. Fubol 58WP contains metalaxyl (10% w/w), which is active against *P. violae*, but the major component is mancozeb (48% w/w). Mancozeb does not affect the pathogens causing cavity spot, is damaging to the environment and presumably contributes significantly to the cost of the product. The availability of an alternative product containing only metalaxyl could give considerable cost savings to growers and would be environmentally more desirable. As *P. sulcatum* is not controlled by metalaxyl, availability of an effective fungicide against *P. sulcatum* would be important for sites where this pathogen predominates.

In the first year of the project a number of fungicides and compounds which were considered to have potential for control of cavity spot were tested in laboratory and glasshouse experiments.

Materials and methods

1. Agar plate tests

Ten fungicides obtained as commercial formulations or provided as samples by the manufacturers (Table 1) were screened in agar plate tests against *P. violae* and *Pythium sulcatum* and their ED_{50} values (the concentration of fungicide which reduced colony growth to half of that in the untreated control) was determined.

Isolates of *P. violae* and *P. sulcatum* were maintained on Campbells vegetable juice agar (V8) (Appendix 1) at 20±2°C.

Stock solutions of fungicides containing 1000 µg/ml (ppm) active ingredient were prepared in sterile distilled water (SDW). Because 2-(thiocyanomethylthio)-benzothiazole has a low solubility in water, it was first dissolved in 3 ml of methanol and then diluted in SDW. These stock solutions were diluted 2- and 10-fold in SDW. Each dilution (20 ml) was mixed with 180 ml of autoclaved and cooled corn meal agar (CMA) (Difco) to achieve the required concentrations of the fungicide active ingredients (100, 50, 10, 5, 1, 0.1, 0.01 µg/ml). The media were mixed thoroughly, poured into 9 cm Petri dishes and dried on a laminar flow bench for 20 min. A control treatment was prepared for each test by adding SDW to the medium instead of fungicide. Each dish was inoculated at one side with a 5 mm diameter mycelial disc of *P. violae*, isolates 2, 6 or 8, or an isolate of *P. sulcatum* cut from 7 day old V8 agar cultures. The dishes were then incubated at 20±2°C for 7 days. Colony growth was measured after 3, 5 and 7 days. There were four replicate dishes for each fungicide/isolate combination.

Table 1. Fungicides used in the agar plate tests for determination of ED_{50} values in year 1.

Active ingredient (a.i.)	Product name	Concentration of a.i.	Manufacturer
Metalaxyl *	Ridomil §	25% (w/w)	Ciba Agriculture
Metalaxyl derivative	A9408B †	48% (w/v)	Ciba Agriculture
Compound found to have activity against Oomycetes	A9180A †	50% (w/w)	Ciba Agriculture
Hymexazole	Tachigaren #	30% (w/v)	Sumitomo Corporation plc
Propamocarb hydrochloride	Filex #	72.2% (w/v)	Levington Horticulture
Fosetyl-Al	Aliette #	80% (w/w)	Rhone Poulenc Agriculture Ltd.
Dimethomorph	Acrobat §	50% (w/w)	Cyanamid Agriculture
Dimethomorph (+ mancozeb)	Invader #	7.5% (w/w)	Cyanamid Agriculture
ß-methoxyacrylate derivative	ICIA5504 †	23.6% (w/w)	Zeneca Agrochemicals
2-(thiocyanomethylthio)-benzothiazole	Not formulated †	99% (w/w)	Buckman Laboratories, Belgium

* approved and available for control of cavity spot on carrots in combination with mancozeb (Fubol 58WP)

§ not available (active ingredient only available in combinations)

† not registered

registered but not approved for use on carrots

Results from measurements after 5 days, by which time the growth in the untreated control plates had almost reached the furthest edge of the plates, were used for statistical analysis. The data was analysed using a Genstat 5 program which fitted logistic curves and calculated the ED_{50} values for each fungicide/isolate combination.

2. Pot experiments

In the first year of trials pot experiments were carried out using naturally infested soil and compost artificially inoculated with the two pathogens. The same fungicides that were screened in the agar plate tests were evaluated with the following exceptions (Table 2). A sample of 2-(thiocyanomethylthio)-benzothiazole was not supplied in time to be used in these pot experiments. Of the two products containing dimethomorph, the one having only dimethomorph as active ingredient (Acrobat) was chosen for the pot experiments.

2.1 Pot experiment with naturally infested field soil

Field soil was recovered from fields where the cavity spot diagnostic assay had indicated high risk of disease, and when the field was visited the crop had c. 45% of carrots with severe cavity spot. Isolation work had shown that the causal organism was *P. violae*. The soil was moistened with tap water and stored in polythene bags in a shaded building (July-August) for four weeks. Gravel (1.5 kg) was placed in the bottom of 25 cm pots and the pots were then filled to the rim with the moistened field soil. Carrot seeds of a susceptible cultivar Nanco F1 were sown in the pots (40 per pot) and covered with a thin layer of the same field soil. The pots were placed in saucers and transferred to a cool glasshouse under natural daylight conditions with frost protection at 5°C and venting at 10°C. The pots were covered with paper until seedling emergence was under way and water was added via the saucers in which they stood. Once emergence was complete the number of seedlings in each pot was reduced to 20. During the growing season the carrots were routinely treated for whitefly (with pyrethrin) as necessary.

The pots were sprayed with the fungicides using the recommended rates given in Table 2 or half of these rates. Dilutions of fungicides were prepared in SDW. Metalaxyl was included in the experiment as a standard and used only at one rate, that being equivalent to the amount of the fungicide in the current standard commercial application. Untreated control pots were sprayed with SDW. Spraying was done with DeVilbiss atomisers using an electric pump. There were 9 replicate pots per fungicide treatment and 12 untreated control pots.

The first harvest of roots was made 140 days after sowing (Appendix 2). Ten roots from each pot were harvested, weighed, scored for numbers of lesions less than 5 mm diameter and those larger than 5 mm diameter. A second harvest from the same pots was made 185 days after sowing, when the same parameters were recorded.

For data from the experiment, analysis of variance was carried out using Genstat 5 to evaluate the effect of the fungicides on the percentage of carrots with cavities (after angular transformation), the total number of cavities, the number of large cavities per carrot, and root weight.

Table 2. The rates of fungicides used in the pot experiments in year 1.*

Active ingredient (a.i.)	Product name (concentration of a.i.)	Rate of a.i.	Equivalent rate of product
Metalaxyl	Ridomil (25% w/w)	1.2 kg/ha in 1000 l	4.8 kg/ha in 1000 l
Metalaxyl derivative	A9408B (48% w/v)	0.6 kg/ha in 1000 l	1.25 l/ha in 1000 l
Compound found to have activity against Oomycetes	A9180A (50% w/w)	0.1 kg/ha in 1000 l	0.2 kg/ha in 1000 l
Hymexazole	Tachigaren (30% w/v)	0.36 g/m^2 in 2 l	1.2 ml/m^2 in 2 l
Propamocarb hydrochloride	Filex (72.2% w/v)	3.61 g/m^2 in 2 l	5.0 ml/m^2 in 2 l
Fosetyl-Al	Aliette (80% w/w)	3.0 kg/ha in 1000 l	3.75 kg/ha in 1000 l
Dimethomorph	Acrobat (50% w/w)	1.2 kg/ha in 1000 l	2.4 kg/ha in 1000 l
ß-methoxyacrylate derivative	ICIA5504 (23.6% w/w)	1.6 kg/ha in 1000 l	6.78 kg/ha in 1000 l

* The above rates were used in the experiments on *P. violae* and *P. sulcatum* in compost. In the experiment on naturally infested soil, the above rates and half rates were used, with the exception of metalaxyl where only the above rate was used.

2.2 Pot experiments with artificially inoculated compost

Two experiments were carried out using compost artificially inoculated with *P. violae* or *P. sulcatum.*

Inocula for these experiments were produced by growing *P. violae* (isolate 6) and *P. sulcatum* in sand/maize meal mixture in 1l Duran bottles for 17 days at 20±2°C. The resulting inoculum was mixed with the moistened compost (Levington M2) to give a concentration of 3.3% (w/w).

Carrot seeds, cv. Nanco F1, were sown in pots (25 cm diam) filled with the moistened inoculated compost (40 seeds per pot) and covered with a thin layer of the same compost. The pots were then treated as described for the experiment with naturally infested soil.

The pots were sprayed with the fungicides using the recommended rates given in Table 2 and dilutions of fungicides were prepared in SDW. Metalaxyl was included in the experiments as a standard. Untreated control pots were sprayed with SDW. Spraying was done with DeVilbiss atomisers using an electric pump. There were 5 replicate pots for each fungicide/pathogen combination, 10 untreated inoculated control pots and 10 untreated uninoculated control pots in each experiment.

The carrots were grown, harvested (135 days after sowing) and scored as for the field soil experiment. Because attack by the fungi was extreme, the data obtained was not suitable for analysis and is presented only in Appendix 3. Figures 1 and 2 in Appendix 4 show respectively the effects of attack by *P. violae* and *P. sulcatum.* Fanging (Appendix 4, Figure 1) has been attributed to many causes including nematodes. In the current system, the only pathogen present would be *P violae*, and it must be assumed that the damage has resulted from attack by that fungus at an early growth stage. The symptom seen in Appendix 4, Figure 2 closely resembles that of 'cobby' carrots frequently seen in field grown crops, and for which no causal agent has yet been identified.

Results and discussion

1. Agar plate tests

Data obtained from screening fungicides in plate tests with *P. violae* and *P. sulcatum* are shown in Table 3. Because the experiment generated numerous similar curves it is inappropriate to present each curve for each fungus/fungicide combination. Some examples of the curves obtained are shown in Appendix 5.

The results in Table 3 are consistent with expectations for metalaxyl (White *et al.*, 1988). *P. violae* was highly sensitive to the fungicide (Appendix 5, Figure 1), giving a mean ED_{50} of 0.48 µg/ml, while the equivalent value for *P. sulcatum* was an order of magnitude higher at 4.53 µg/ml (Appendix 5, Figure 2). For the other fungicides, equivalent work had never been done so they are considered in detail below.

The Ciba-Geigy products were known to comprise one fungicide which is generally more active than metalaxyl, and one which lacks activity *in vitro* but is effective *in vivo.* A9408B was found to have a mean ED_{50} for *P. violae* of 0.06 μg/ml (almost an order of magnitude more active than metalaxyl, Appendix 5, Figure 3), while the equivalent value for *P. sulcatum* was 1.54 μg/ml. A9180A showed no sign of activity against either species of *Pythium* (Appendix 5, Figure 4).

Hymexazole is primarily active against *Aphanomyces* spp. Whilst having little or no effect against *P. violae*, the fungicide was indicated as highly effective against *P. sulcatum*. On closer inspection of the curve derived for hymexazole with *P. sulcatum* (Appendix 5, Figure 5), it is clear that at the lower concentrations the response is very steep. However, at all the higher concentrations the fungus produced significant growth, and it must be considered that the calculated ED_{50} is spurious.

Results with propamocarb reflect the relatively poor effect of the fungicide seen in early cavity spot field trials, while the lack of effect of fosetyl-Al on either species reflects the known absence of effect *in vitro* of that fungicide.

While dimethomorph alone was ineffective against either species of *Pythium*, when used as part of the formulated product with mancozeb, reasonable ED_{50} values were obtained. As it is known that mancozeb is not active against either of the species (White *et al.*, 1992) these results must indicate some synergistic action of the components of the product which could warrant further investigation.

Both ICIA5504 and 2-(thiocyanomethylthio)-benzothiazole have been the subject of few studies with *Pythium* species, although activity against members of the genus has been reported. The former was found to have moderate activity against *P. violae*, but gave a low ED_{50} for *P sulcatum* (0.08 μg/ml) the latter gave moderate ED_{50}'s for both species. The result with ICIA5504 appears valid, and if so is worthy of further investigation.

2. Pot experiments

The results of the test with field soil are shown in Table 4 (Harvest 1), and Table 5 (Harvest 2). For statistical reasons, analysis is done using percentage data transformed to Angles, but for accessibility we consider effects shown in untransformed data. At the first harvest 65.6% of all roots in the untreated control pots had cavity spot. Treatment with the commercial standard fungicide metalaxyl gave a highly significant ($P < 0.001$) reduction of that value to 21.1%. Results with A9408B at full rate, A9180A, dimethomorph and ICIA5504, all at half rate, were not significantly different from that with metalaxyl. Differences between the fungicide rates were generally not significant. Only for A9408B did the full rate appear to be more effective than the half rate. For A9180A, propamocarb and dimethomorph there was no apparent difference, and for hymexazole and ICIA5504 the full rate application appeared to give more cavities than the half rate.

By the second harvest, the percentage of roots with cavities in the untreated control had increased to 83.3%, and metalaxyl remained the most effective treatment with 34.2% disease. All other treatments showed a considerable loss of effectiveness and gave significantly more carrots with cavities than metalaxyl ($P < 0.05$). Effects of rate of application were largely absent, and although there are apparent differences between some of the fungicide treatments, these are below the level of statistical significance.

For size of cavity, at the first harvest the < 5 mm / > 5 mm ratio for carrots from the untreated control was 1.3, while that value in the metalaxyl treatment was 1.0. For the other treatments, the number of small and large cavities were generally of the same order. By the second harvest, the number of large cavities had increased in all treatments. The ratio of cavities < 5 mm to those > 5 mm in the untreated controls was 0.5, while that value in the metalaxyl treatment was 0.2 (little increase in number of cavities, but considerable increase in size). The trend was similar for all the other fungicide treatments.

Although root weight is not an important parameter in cavity spot work, treatment effects in this system can indicate control of the *Pythium* root dieback which results from maintaining moist soil conditions. At the first harvest, few treatments significantly increased root weight as compared with the untreated control. The commercial standard metalaxyl showed a 15.4% increase, A9180A at the full rate increased root weight by 16.0%, while the equivalent value for dimethomorph was 34.7%. Between the first and second harvests root weight across all treatments increased by 83.4%. The difference between roots from the metalaxyl treatment and the untreated controls increased to 24.7%. Several other treatments (full rate A9408B, half rate of fosetyl-Al and ICIA5504) gave similar increases, whilst the increase resulting from the use of dimethomorph at the full rate was 41.2%.

Conclusions

1. In plate tests, metalaxyl was confirmed as highly active against *P.violae* the primary cause of cavity spot of carrot in the UK. Of the other fungicides tested, A9408B gave an ED_{50} value an order of magnitude lower than that of metalaxyl. Of the fungicides already in commercial usage and others still in product development, none had equivalent activity against *P. violae.*

2. For *P. sulcatum*, most fungicides gave relatively high ED_{50} values. That with hymexazole was particularly low, but probably does not reflect the true efficacy of hymexazole with the fungus. In contrast, the low ED_{50} with ICIA5504 for *P. sulcatum* appears a true result, and is worthy of further investigation.

3. In pot experiments the commercial standard fungicide metalaxyl was most effective at each of two harvests. At the first harvest several other treatments had reduced the disease to a similar level, but as disease progressed in the period up to the second harvest, the benefits from all treatments other than metalaxyl were reduced.

4. In the current pot experiments conditions favoured the pathogens for the whole growing period. This meant that inoculum pressure was uniformly high. For future work, some of the materials showing activity close to that of metalaxyl should be tested in experiments where conditions are less biased towards the pathogens

Table 3. ED_{50} values (ppm) of ten fungicide active ingredients on the isolates of *P. violae* and *P. sulcatum*.

	P. violae, isolate 2		*P. violae*, isolate 6		*P. violae*, isolate 8		*P. sulcatum*	
Active ingredient	ED_{50}	SE	ED_{50}	SE	ED_{50}	SE	ED_{50}	SE
Metalaxyl	0.47	0.01	0.40	0.08	0.57	0.01	4.53	2.84
A9408B	0.06	0.00	0.06	0.01	0.07	0.00	1.54	0.50
A9180A	>100	-	>100	-	>100	-	>100	-
Hymexazole	97.00	1.69	96.25	-	33.51	1.27	0.08	0.00
Propamocarb hydrochloride	4.68	0.30	15.53	2.07	13.31	4.45	3.78	0.11
Fosetyl-Al	>100	-	>100	-	>100	-	>100	-
Dimethomorph	>100	-	>100	-	>100	-	>100	-
Dimethomorph + mancozeb	1.10	0.14	3.03	0.19	0.14	0.02	2.01	0.60
ICIA5504	3.16	0.15	1.32	0.20	3.49	0.05	0.08	0.00
2-(thiocyanomethylthio)-benzothiazole	2.98	0.03	2.00	0.07	2.53	0.09	0.54	0.04

ED_{50} - amount of fungicide required to reduce growth of fungus by 50 %; SE - standard error

Table 4. Results of the first harvest of carrots from field soil naturally infested with *Pythium violae* in year 1.

Fungicide a.i.	Rate	Percentage of carrots with cavities	Angle % of carrots with cavities	Number of cavities/carrot	Root weight expressed as % of untreated control
Untreated control		65.6	54.2	1.29	100.0
Metalaxyl		21.1	27.1	0.27	115.4
A9408B	A	38.9	38.4	0.63	100.1
	B	27.8	31.1	0.37	103.1
A9180A	A	31.1	31.7	0.54	80.5
	B	32.2	33.9	0.57	116.0
Hymexazole	A	36.7	36.9	0.58	114.1
	B	43.3	40.7	0.69	108.3
Propamocarb	A	41.1	39.6	0.66	94.5
	B	38.9	38.4	0.68	100.4
Fosetyl-Al	A	38.9	38.1	0.60	103.3
	B	31.1	33.2	0.62	106.0
Dimethomorph	A	31.1	31.9	0.44	109.5
	B	31.1	33.4	0.48	134.7
ICIA5504	A	28.9	32.1	0.49	83.4
	B	45.6	42.4	0.64	92.1
LSD p 0.05			8.74	0.27	21.03
0.01			11.55	0.35	27.64
0.001			14.89	0.45	35.31

Rate A - half the standard commercial rate of application; Rate B - the standard commercial rate of application.

Table 5. Results of the second harvest of carrots from field soil naturally infested with *Pythium violae* in year 1.

Fungicide a.i.	Rate	Percentage of carrots with cavities	Angle % of carrots with cavities	Number of cavities/carrot	Root weight expressed as % of untreated control
Untreated control		83.3	69.1	1.72	100.0
Metalaxyl		34.2	35.6	0.55	124.7
A9408B	A	53.3	47.1	1.19	116.1
	B	48.1	43.9	0.93	131.2
A9180A	A	58.9	50.3	1.37	102.9
	B	54.0	47.6	1.20	105.4
Hymexazole	A	51.1	45.7	0.94	117.9
	B	57.7	49.5	1.05	114.1
Propamocarb	A	65.4	54.5	1.45	112.2
	B	60.8	51.4	1.39	114.7
Fosetyl-Al	A	49.2	44.5	0.94	124.5
	B	59.4	50.8	1.33	110.0
Dimethomorph	A	47.2	43.3	0.77	127.2
	B	50.0	45.0	0.79	141.2
ICIA5504	A	54.4	47.6	1.07	124.4
	B	58.9	50.3	1.18	117.9
LSD p 0.05			9.04	0.38	19.40
0.01			11.95	0.49	25.50
0.001			15.40	0.63	32.58

Rate A - half the standard commercial rate of application; rate B - the standard commercial rate of application.

YEAR 2 (1996-1997) - EFFICACY OF A9408B AND ICIA5504 IN CONTROLLING CAVITY SPOT OF CARROT

Introduction

In the first year of the project nine potential fungicides were screened for control of cavity spot. Two of these were considered to be worthy of further evaluation. The new formulation of metalaxyl, A9408B, gave a lower ED_{50} value for *P. violae* than metalaxyl in laboratory tests in the first year. In a pot experiment, A9408B was initially as effective as the metalaxyl standard, but at the last harvest it did not give as good control as metalaxyl. As registration is being sought for this new formulation on carrots, and as it may shortly replace Fubol 58 WP, it was considered essential to determine its efficacy in controlling cavity spot particularly in the latest stages of development. Also in the first year experiments, the product ICIA5504 gave a lower ED_{50} value for *P. sulcatum* than metalaxyl in laboratory tests, but its efficacy was not tested in a pot experiment. Consequently in the second year, its efficacy was further evaluated in pot experiments using field soil.

Materials and methods

1. Pot experiments

In the second year of trials the efficacy of the new formulation of metalaxyl (A9408B) was further evaluated in two field soils. In another experiment the efficacy of ICIA5504 that had given promising results in the agar plate tests in the first year was tested in field soil inoculated with *P. sulcatum.*

1.1 Efficacy of A9408B

Two pot experiments were carried out using naturally infested soils from two different locations (Soil 1 and 2) (Table 7). Soil 1 was from the same field used in the 1st year while the incidence of cavity spot in field-grown carrots was 25% in 1996 in soil 2 and the causal organism was found to be *P. violae*. The efficacy of different rates of A9408B was compared with that of Fubol 58 WP in pot experiments using these two field soils.

The soils were collected and stored and the pots were set up as in year 1. Two weeks after sowing the seedlings were sprayed with fungicides by the same method as described for year 1. The fungicides and spray rates are given in Table 6. There were 8 replicate pots per treatment for each soil and enough pots for four harvests. The experiment was carried out in a cool glasshouse as for year 1 and the carrots were routinely treated for aphids (with heptenophos and pirimicarb) and powdery mildew (with triadimefon) as necessary.

The roots were harvested four times at monthly intervals from 130 days after sowing onwards (Appendix 6). At each of the four harvests, ten roots from each treatment were lifted, washed, weighed and scored for numbers of lesions less than 5 mm diameter and those larger than 5 mm diameter.

Table 6. The rates of A9408B and Fubol 58WP used in pot experiments on two different field soils (soils 1 and 2) naturally infested by *Pythium violae* in year 2.

Active ingredient (a.i.)	Product name (concentration of a.i.)	Equivalent rate of a.i./ ha in 1000 l	Equivalent rate of product/ ha in 1000 l
None (Untreated control)	-	-	-
Metalaxyl	Fubol 58 WP (10% w/w) †	1.2 kg	12 kg
Metalaxyl *	A9408B (48% w/v) #	0.6 kg	1.25 l
Metalaxyl *	A9408B (48% w/v) #	1.2 kg	2.5 l
Metalaxyl *	A9408B (48% w/v) #	2.4 kg	5.0 l

* new formulation

† approved for control of cavity spot on carrots

not registered

Isolations were made from cavities on corn meal agar amended with rifamycin (30 ppm) and pimaricin (100 ppm). Several isolates of *P. violae* and fast growing species of *Pythium* were recovered. Five *P. violae* isolates from soil 1 and four from soil 2 were tested for their sensitivity to metalaxyl. ED_{50} values of metalaxyl for these isolates were determined in agar plate tests in a similar manner to that described for the fungicides in year 1.

1.2 Efficacy of ICIA5504

A pot experiment was carried out to test the efficacy of ICIA5504 against *P. sulcatum* in field soil 3 (Table 7) which had been disinfected prior to inoculation with *P. sulcatum*.

The soil was disinfected by heat treating (+80°C) for 48 hours and then distilled water was added to give the same moisture content (approx. 10% of dry weight) as in the field. Inoculum for this experiment was produced by growing *P. sulcatum* in sand/maize meal mixture in autoclave bags for 4 weeks. The resulting inoculum was mixed with the moist soil to give a concentration of 2% (w/w).

The pots were set up as in year 1 and the seedlings were sprayed with fungicides by the same method as described for year 1. The fungicides and spray rates are given in Table 8. There were 8 replicate pots per treatment. Eight replicate untreated control pots were also included using uninoculated (heat treated) soil. The plants were grown in a cool glasshouse as for year 1. During the growing season, the carrots were routinely treated for aphids (with heptehophos and pirimicarb) and powdery mildew (with triadimefon) as necessary.

The roots were harvested twice at a monthly interval starting 140 days after sowing (Appendix 6). At each harvest ten roots from each treatment were lifted, washed, weighed and scored for numbers of lesions less than 5 mm diameter and those larger than 5 mm diameter.

It was decided to carry out an extra pot experiment (Table 9) to clarify the activity of ICIA5504 using field soil 3 without heat treatment (natural infestation with *P. violae*). The experiment was performed as described above for the heat treated field soil 3.

Table 7. Origin, pH and causal organism of cavity spot of the soils used in the glasshouse experiments in year 2.

Soil number	Origin of soil	pH	Causal organism of cavity spot present in soil
1	Gooderstone, Norfolk	7.9	*P. violae*
2	Gooderstone, Norfolk	7.8	*P. violae*
3	HRI, Wellesbourne	7.1	*P. violae*

1.3 Analysis of data

For data from all experiments, analysis of variance was carried out using Genstat 5 to evaluate the effect of A9408B and ICIA5504 on the percentage of carrots with cavities (after angular transformation), the total number of cavities, the number of large cavities per carrot, and root weight.

Table 8. The rates of ICIA5504 and Fubol 58 WP used in the pot experiment on field soil 3 following disinfection and subsequent inoculation with *Pythium sulcatum* in year 2.

Active ingredient (a.i.)	Product name (concentration of a.i.)	Equivalent rate of a.i./ ha in 1000 l	Equivalent rate of product/ha in 1000 l
None (Uninoculated control)	-	-	-
None (Inoculated control)	-	-	-
Metalaxyl	Fubol 58 WP (10% w/w) *	1.2 kg	12 kg
ß-methoxyacrylate derivative	ICIA5504 (23.6% w/w) †	1.6 kg	6.78 kg

* approved for control of cavity spot on carrots

† not registered

Table 9. The rate of ICIA5504 used in the pot experiment on field soil 3 naturally infested by *Pythium violae* in year 2.

Active ingredient (a.i.)	Product name (concentration of a.i.)	Equivalent rate of a.i./ha in 1000 l	Equivalent rate of product/ha in 1000 l
None (Untreated control)	-	-	-
ß-methoxyacrylate derivative	ICIA5504 (23.6% w/w) †	1.6 kg	6.78 kg

† not registered

Results and discussion

1. Pot experiments

1.1 Efficacy of A9408B

Results of the four harvests from two pot tests with field soils both naturally infested with *P. violae* are shown in Tables 10 - 13 (Soil 1) and Tables 15 - 18 (Soil 2). Summaries meaning the data over all harvests are given in Table 14 and Table 19. *P. violae* isolates that were tested from both field soils were highly sensitive to metalaxyl.

For Soil 1, across the four harvests, the major effect was that only A9408B at the highest rate consistently reduced the percentage of carrots with cavities, the number and size of cavities, and gave an increase in root weight. Because of the size of each harvest, statistical significance was variable or non-existent. The data were therefore meaned over the four harvest to give an appraisal of the findings (Table 14). Reduction in percentage infection from Fubol 58 WP was just significant ($P < 0.05$) and was equivalent to that from the two lower rates of A9408B. Reduction in percentage infection due to the highest rate of A9408B was highly significant ($P < 0.001$). The same pattern was seen for both the number of cavities per carrot and the number of cavities > 5 mm/carrot. Over the experiment, Fubol 58 WP and the high rate of A9408B both significantly increased root weight ($P < 0.001$; $P < 0.01$ respectively).

In the experiment with soil 2 levels of cavity spot were low and erratic (Tables 15 - 18). The summary Table (Table 19) shows no significant differences across the data set for percentage of carrots with cavities. Number of cavities per carrot was highly variable, and this was mirrored in the number of large cavities per carrot. These results, in combination with the complete absence of root weight benefits suggest that with this soil, the different metalaxyl treatments have had no effect at all.

1.2 Efficacy of ICIA5504

Tables 20 and 21 show the results from two harvests of the experiment with ICIA5504 and soil artificially infested with *P. sulcatum*. Results of the data meaned across the harvests is given in Table 22. In both harvests the uninoculated controls gave a low percentage of carrots with cavities, indicating either incomplete sterilisation of the soil, or transfer by insects of *Pythium* from other pots in the compartment. The inoculated control gave a high percentage of carrots with cavities, total number of cavities and number of large cavities compared with either Fubol 58 WP or ICIA5504. While the reduction in percentage infection was not statistically significant, reductions in total cavities and number of large cavities were highly significant. Both fungicides gave significant increases in root weight. While the results with ICIA5504 were as expected, that Fubol 58 WP should give apparently equivalent results in an experiment with *P. sulcatum* is not consistent with our knowledge of that fungus/fungicide combination.

To clarify the information on the range of activity of ICIA5504, an extra experiment was conducted using field soil 3 (natural infestation with *P. violae*). The experiment comprised an untreated soil control and one treatment in which ICIA5504 was applied as in the work above. There were two harvests at 141 and 168 days from sowing (Tables 23 and 24) and data were meaned across those harvests (Table 25). As was expected from it's lack of *in vitro* activity against *P. violae*, there was no significant benefit from the use of ICIA5504 for any parameter measured.

Conclusions

1. Pot experiment results for soil 1 generally confirmed findings for year 1. Different formulations with metalaxyl reduced cavity spot as measured by percentage of carrots with cavities, number of cavities and number of large cavities per carrot. The reductions were less than would have been expected from similar treatments made shortly after the introduction of that fungicide in the mid 1980's.

2. In soil 2, for practical purposes there was no evidence of effect of any of the treatments.

3. ICIA5504 was shown to be effective in controlling cavity spot in soil artificially infested with *P. sulcatum*. However, in line with expectations from work in year 1 the fungicide had no effect on cavity spot where *P. violae* was causal.

Table 11. The efficacy of A9408B in controlling cavity spot in the field soil 1 naturally infested with *Pythium violae* in year 2. Second harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	40.0	(38.9)	0.66	0.39	100
Fubol 58 WP (1.2)	31.3	(31.6)	0.48	0.29	120
A9408B (0.6)	37.5	(33.4)	0.71	0.34	124
A9408B (1.2)	18.8	(22.2)	0.31	0.14	109
A9408B (2.4)	16.3	(20.1)	0.27	0.18	112
LSD p 0.05		17.67 (NS)	0.25	0.16	14.05
0.01			0.33		18.47
0.001			0.42		

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 10. The efficacy of A9408B in controlling cavity spot in the field soil 1 naturally infested with *Pythium violae* in year 2. First harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	42.5	(39.8)	0.76	0.34	100
Fubol 58 WP (1.2)	36.3	(35.8)	0.54	0.28	115
A9408B (0.6)	40.0	(38.4)	0.73	0.43	104
A9408B (1.2)	35.0	(35.7)	0.55	0.26	104
A9408B (2.4)	21.3	(21.9)	0.29	0.08	123
LSD p 0.05		15.61 (NS)	0.25	0.16	12.11
0.01			0.33	0.21	15.92
0.001				0.27	20.34

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 12. The efficacy of A9408B in controlling cavity spot in the field soil 1 naturally infested with *Pythium violae* in year 2. Third harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	36.2	(36.2)	0.65	0.49	100
Fubol 58 WP (1.2)	21.2	(26.8)	0.34	0.25	99
A9408B (0.6)	25.0	(27.7)	0.42	0.23	94
A9408B (1.2)	22.5	(25.9)	0.38	0.30	112
A9408B (2.4)	10.0	(17.1)	0.15	0.11	108
LSD p 0.05		13.60 (NS)	0.25	0.20	11.70
0.01			0.33	0.26	

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 13. The efficacy of A9408B in controlling cavity spot in the field soil 1 naturally infested with *Pythium violae* in year 2. Fourth harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	37.5	(37.2)	0.66	0.42	100
Fubol 58 WP (1.2)	25.0	(27.8)	0.48	0.26	126
A9408B (0.6)	26.3	(28.6)	0.50	0.24	96
A9408B (1.2)	23.8	(26.8)	0.45	0.23	106
A9408B (2.4)	18.8	(21.9)	0.34	0.28	104
LSD p 0.05		13.46 (NS)	0.29 (NS)	0.21 (NS)	12.76
0.01					16.77
0.001					21.42

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 14. The efficacy of A9408B in controlling cavity spot in the field soil 1 naturally infested with *Pythium violae* in year 2. Combined results of four harvests.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	39.1	(38.0)	0.68	0.40	100
Fubol 58 WP (1.2)	28.4	(30.5)	0.46	0.27	114
A9408B (0.6)	32.2	(32.0)	0.59	0.31	103
A9408B (1.2)	25.0	(27.6)	0.42	0.23	108
A9408B (2.4)	16.6	(20.3)	0.26	0.16	111
LSD p 0.05		7.05	0.14	0.10	6.72
0.01		9.32	0.18	0.13	8.84
0.001		12.01	0.23	0.16	11.29

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 15. The efficacy of A9408B in controlling cavity spot in the field soil 2 naturally infested with *Pythium violae* in year 2. First harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	10.0	(12.9)	0.14	0.04	100
Fubol 58 WP (1.2)	8.8	(12.1)	0.10	0.03	104
A9408B (0.6)	13.8	(13.6)	0.21	0.06	107
A9408B (1.2)	17.5	(17.6)	0.24	0.06	105
A9408B (2.4)	7.5	(11.3)	0.13	0.03	107
LSD p 0.05		16.42 (NS)	0.14 (NS)	0.06 (NS)	11.07 (NS)

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 16. The efficacy of A9408B in controlling cavity spot in the field soil 2 naturally infested with *Pythium violae* in year 2. Second harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	10.0	(14.4)	0.13	0.06	100
Fubol 58 WP (1.2)	8.8	(13.6)	0.13	0.05	116
A9408B (0.6)	8.8	(10.5)	0.14	0.03	107
A9408B (1.2)	13.8	(18.3)	0.20	0.10	111
A9408B (2.4)	1.2	(2.3)	0.01	0.00	104
LSD p 0.05		13.82 (NS)	0.13 (NS)	0.07	14.23 (NS)

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 17. The efficacy of A9408B in controlling cavity spot in the field soil 2 naturally infested with *Pythium violae* in year 2.Third harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	16.3	(19.9)	0.28	0.11	100
Fubol 58 WP (1.2)	28.8	(28.1)	0.93	0.55	92
A9408B (0.6)	21.3	(21.8)	0.40	0.18	92
A9408B (1.2)	7.5	(11.3)	0.13	0.08	90
A9408B (2.4)	15.0	(14.6)	0.33	0.20	84
LSD p 0.05		21.75 (NS)	0.29	0.18	10.90 (NS)
0.01			0.38	0.24	
0.001			0.49	0.31	

Angle = arcsine transformation of percentages to which LSD applies

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 18. The efficacy of A9408B in controlling cavity spot in the field soil 2 naturally infested with *Pythium violae* in year 2. Fourth harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	10.0	(14.6)	0.24	0.23	100
Fubol 58 WP (1.2)	28.7	(29.4)	0.69	0.29	104
A9408B (0.6)	35.0	(33.9)	0.90	0.25	95
A9408B (1.2)	11.3	(15.1)	0.15	0.10	99
A9408B (2.4)	23.7	(27.1)	0.51	0.24	92
LSD p 0.05		16.92 (NS)	0.33	0.19 (NS)	12.21 (NS)
0.01			0.44		
0.001			0.56		

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 19. The efficacy of A9408B in controlling cavity spot in the field soil 2 naturally infested with *Pythium violae* in year 2. Combined results of four harvests.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	11.6	(15.4)	0.19	0.11	100
Fubol 58 WP (1.2)	18.8	(20.8)	0.46	0.23	103
A9408B (0.6)	19.7	(19.9)	0.41	0.13	99
A9408B (1.2)	12.5	(15.6)	0.18	0.09	100
A9408B (2.4)	11.9	(13.8)	0.24	0.12	95
LSD p 0.05		8.32 (NS)	0.14	0.08	6.21 (NS)
0.01			0.18	0.10	
0.001			0.23		

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 20.The efficacy of ICIA5504 in controlling cavity spot in the field soil 3 artificially inoculated with *Pythium sulcatum* in year 2. First harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Uninoculated + Untreated	5.0	(4.9)	0.11	0.00	146
Untreated	62.5	(53.2)	2.52	1.42	100
Fubol 58 WP (1.2)	32.5	(28.6)	1.11	0.59	133
ICIA5504 (1.6)	36.2	(33.0)	1.31	0.36	128
LSD* p 0.05		29.41 (NS)	0.58	0.40	17.01
0.01			0.77	0.53	22.48
0.001			1.00	0.69	28.97

Angle = arcsine transformation of percentages to which LSD applies.

* LSD between treatments excluding Uninoculated + Untreated control .

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 21.The efficacy of ICIA5504 in controlling cavity spot in the field soil 3 artificially inoculated with *Pythium sulcatum* in year 2. Second harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Uninoculated + Untreated	7.5	(11.3)	0.13	0.09	140
Untreated	55.0	(47.9)	1.99	1.21	100
Fubol 58 WP (1.2)	38.7	(36.1)	0.90	0.55	143
ICIA5504 (1.6)	28.7	(29.6)	0.76	0.53	127
LSD* p 0.05		26.40 (NS)	0.45	0.35	18.49
0.01			0.59	0.47	24.44
0.001			0.76	0.60	31.50

Angle = arcsine transformation of percentages to which LSD applies.

* LSD between treatments excluding Uninoculated + Untreated control .

NS = not significant difference at the 5% level of probability (p<0.05).

Table 22. The efficacy of ICIA5504 in controlling cavity spot in the field soil 3 artificially inoculated with *Pythium sulcatum* in year 2. Combined results of the first and second harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Uninoculated + Untreated	6.3	(8.1)	0.14	0.04	144
Untreated	58.8	(50.6)	2.22	1.29	100
Fubol 58 WP (1.2)	35.6	(32.3)	1.01	0.57	138
ICIA5504 (1.6)	32.5	(31.3)	1.04	0.44	128
LSD * p 0.05		17.12	0.46	0.31	12.92
0.01			0.61	0.40	16.98
0.001			0.78	0.51	21.69

Angle = arcsine transformation of percentages to which LSD applies.

* LSD between treatments excluding Uninoculated + Untreated control.

Table 23.The efficacy of ICIA5504 in controlling cavity spot in the field soil 3 naturally infested with *Pythium violae* in year 2. First harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	72.5	(58.9)	1.53	0.69	100
ICIA5504 (1.6)	53.8	(47.3)	1.11	0.43	104
LSD p 0.05		14.24 (NS)	0.40	0.26	13.17 (NS)

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 24.The efficacy of ICIA5504 in controlling cavity spot in the field soil 3 naturally infested with *Pythium violae* in year 2. Second harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	50.0	(45.0)	0.98	0.52	100
ICIA5504 (1.6)	52.5	(48.4)	1.06	0.72	88
LSD p 0.05		17.03 (NS)	0.35 (NS)	0.28 (NS)	13.11 (NS)

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

Table 25. The efficacy of ICIA5504 in controlling cavity spot in the field soil 3 naturally infested with *Pythium violae* in year 2. Combined results of the first and second harvest.

Fungicide (a.i. kg/ha)	Percentage of carrots with cavities	(Angle)	Total number of cavities per carrot	Number of large (>5mm) cavities per carrot	Root weight expressed as % of untreated control
Untreated	61.3	(52.0)	1.25	0.60	100
ICIA5504 (1.6)	53.1	(47.9)	1.09	0.57	96
LSD p 0.05		9.44 (NS)	0.27 (NS)	0.20 (NS)	10.00 (NS)

Angle = arcsine transformation of percentages to which LSD applies.

NS = not significant difference at the 5% level of probability ($p<0.05$).

C. OVERALL DISCUSSION

For some years there has been no work on control of cavity spot, although it was recognised that there were considerable gaps in our knowledge in that area. In that time there has been an increasing feeling, without factual information, that the current commercial application of metalaxyl fungicide in the form of Fubol 58 WP becomes progressively less effective. Because of the precise way metalaxyl controls *Pythium*, and from evidence in other areas, this might be expected. In the UK protected ornamentals sector resistance to metalaxyl/furalaxyl is widespread. However, no sign of resistance has been detected in any isolates of *P. violae* assessed in this or other work. *P. sulcatum* isolates have generally shown a natural tolerance of metalaxyl, and tests done here confirm that.

Early experiments in the mid-1980's with metalaxyl showed reductions of cavity spot to one tenth of the value seen in untreated controls with soils which had not previously been exposed to the fungicide. This was not seen in any of the present experiments with soil from carrot fields which have probably grown three or four metalaxyl treated crops in the meantime. Without further evidence, it is not possible to do more than state that the feelings that the treatment is becoming less effective over the years could have substance. This aspect will be thoroughly examined in collaboration with Novartis should the prospective Horticulture Link project be funded.

Fungicides other than metalaxyl were generally found to be less, or non-effective against cavity spot pathogens. Because questions have continued to be asked by growers on whether they might benefit from switching to alternative products, we have looked at the alternatives and confirmed earlier findings. Further, with the exception of the Zeneca product ICIA5504 which has activity against *P. sulcatum*, there appears little in the new molecules area which will help the carrot grower.

Although the search for new fungicides for cavity spot control will continue, it is likely that growers will necessarily be forced to concentrate more on management of the disease than on chemical control. There is a finite limit on the number of approaches which might be used. These are listed in the Final Report of HDC Project FV 5e which reviews the World literature on the disease.

D. OVERALL CONCLUSIONS

1. Plate tests confirmed that metalaxyl was the most active of the currently available relevant fungicides against *P. violae*.

2. At present there is no fungicide on the market which would improve on cavity spot control obtained from an application of Fubol 58 WP in accordance with the label recommendation.

3. The new formulation of metalaxyl (A9408B) is considerably more active than metalaxyl *in vitro* but appears less so in tests with soil. Comparative field tests are required to confirm or reject this finding.

4. There are no fungicides currently in development which in the present tests appeared likely to improve on metalaxyl.

5. For *P. sulcatum*, the Zeneca product ICIA5504 shows considerable promise, but it has been confirmed to have no effect on *P. violae*. Future availability of the fungicide will depend on Zeneca.

E. RECOMMENDATIONS

1. It is increasingly important that growers are aware of the likely risk of cavity spot before they decide to grow the crop in a particular field. Use the cavity spot diagnostic test.

2. Fubol 58 WP is confirmed as the present most effective fungicide for the control of cavity spot. Use it in accordance with the label recommendation.

3. Where results from the cavity spot diagnostic test show that a field has a high level of inoculum do not grow carrots in the coming year. Because pathogen levels fluctuate from year to year, re-test the field in subsequent years.

4. Read the review of World cavity spot literature (Final Report for HDC Project FV 5e) because it covers all aspects of crop management to avoid the disease.

F. GLOSSARY

Cavity spot	the major disease of carrot in temperate countires which is caused by slow-growing *Pythium* species. The disease is initially seen as pale sunken lesions which over a short period discolour and are enlarged by secondary pathogens, bacteria and nematodes.
ED_{50}-value	the concentration of fungicide which reduces colony growth to half of that in the untreated control
Genstat	the computing language used to process all data in this project.
ppm	parts per million = μg/ml

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APPENDICES

Appendix 1

Campbells vegetable juice agar (V8)

$CaCO_3$	0.8g
V8 juice	40ml
Distilled water	360ml
Agar	6g

Appendix 2

Sowing, spraying and harvest dates in year 1.

Growing medium	Sowing date	Spraying date	Harvest dates	Number of days to the harvest
Field soil	22.8.1995	5.9.1995	8.1.1996 22.2.1996	139 185
Compost	30.8.1995	12.9.1995	11.1.1996	134

Appendix 3

Results of fungicide screening for the control of cavity spot in compost inoculated with either *Pythium violae* or *Pythium sulcatum*

Fungicide a.i.	Percentage of roots with cavities	Mean number of cavities/root
	Pythium violae	
Untreated uninoculated	0	0
Untreated inoculated	58.0	2.3
Metalaxyl	52.0	2.4
A9408B	56.0	2.7
A9180A	54.0	2.4
Hymexazole	44.0	3.1
Propamocarb	70.0	2.4
Fosetyl-Al	56.0	2.4
Dimethomorph	36.0	2.2
ICIA5504	38.0	2.5
	Pythium sulcatum	
Untreated uninoculated	0	0
Untreated inoculated	14.0	0.9
Metalaxyl	2.0	1.0
A9408B	12.0	1.5
A9180A	10.0	1.1
Hymexazole	16.0	0.9
Propamocarb	16.0	1.1
Fosetyl-Al	10.0	0.9
Dimethomorph	16.0	1.6
ICIA5504	10.0	0.7

Appendix 4

Figure 1. Fanging effect from extreme attack of *Pythium violae* on carrots growing in compost artificially infested with the fungus

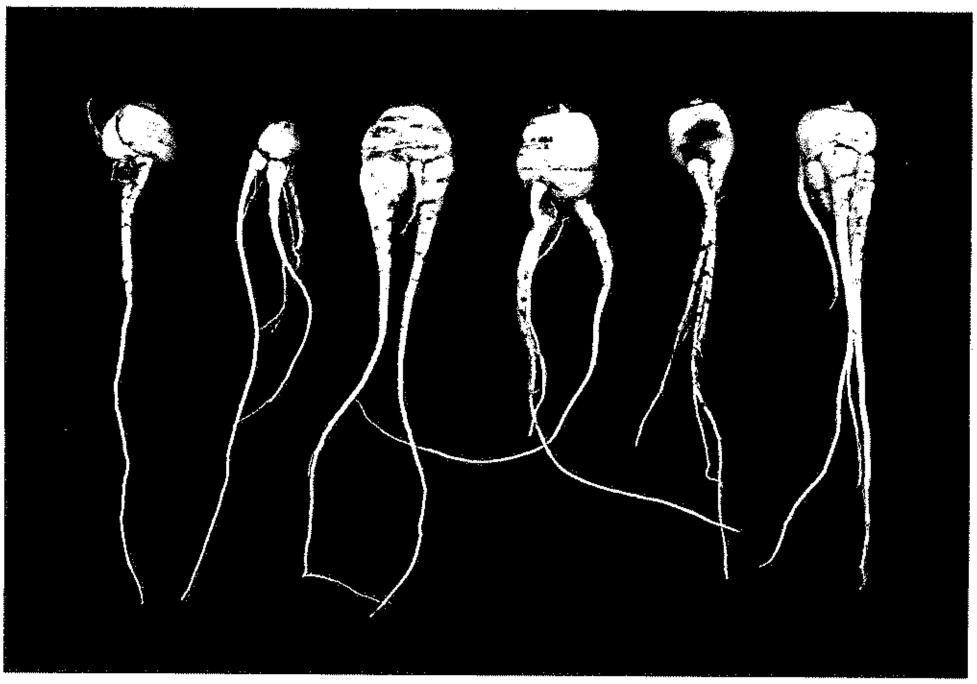
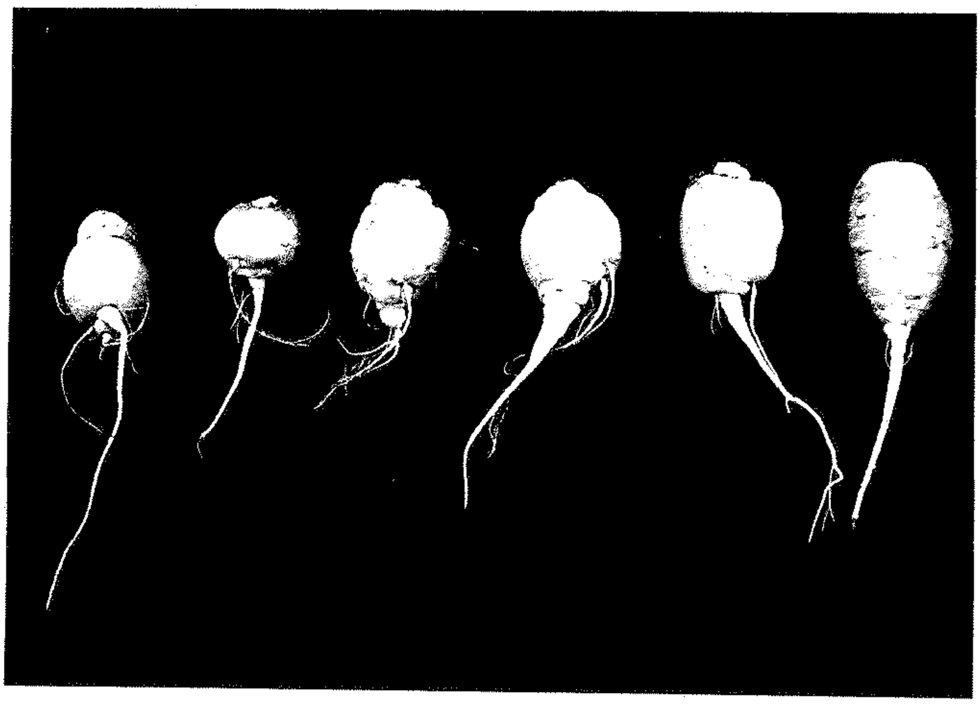
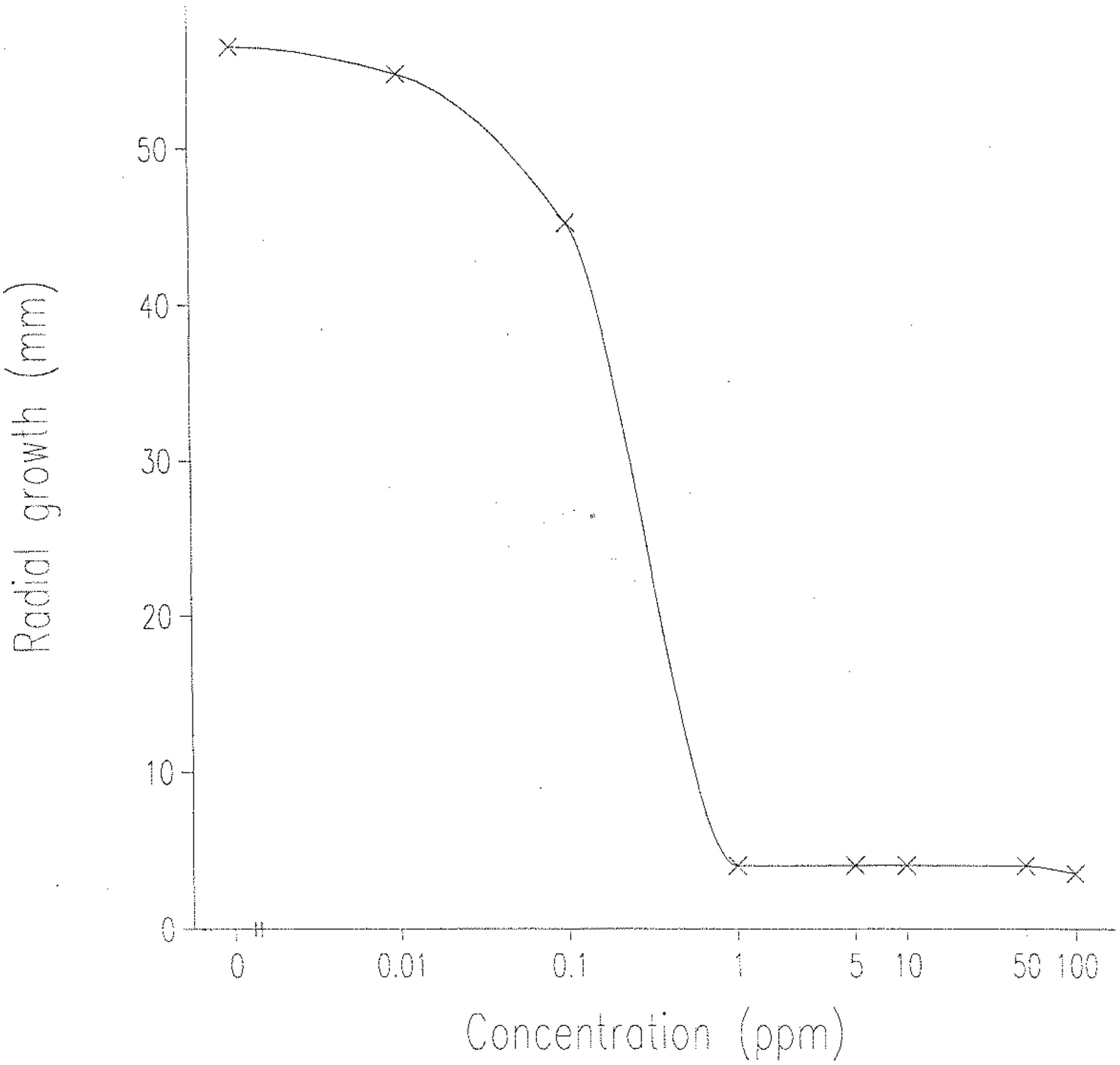


Figure 2. 'Cobby' growth of carrots resulting from infection by *Pythium sulcatum* when the carrots were grown in compost artificially infested with the fungus



Appendix 5

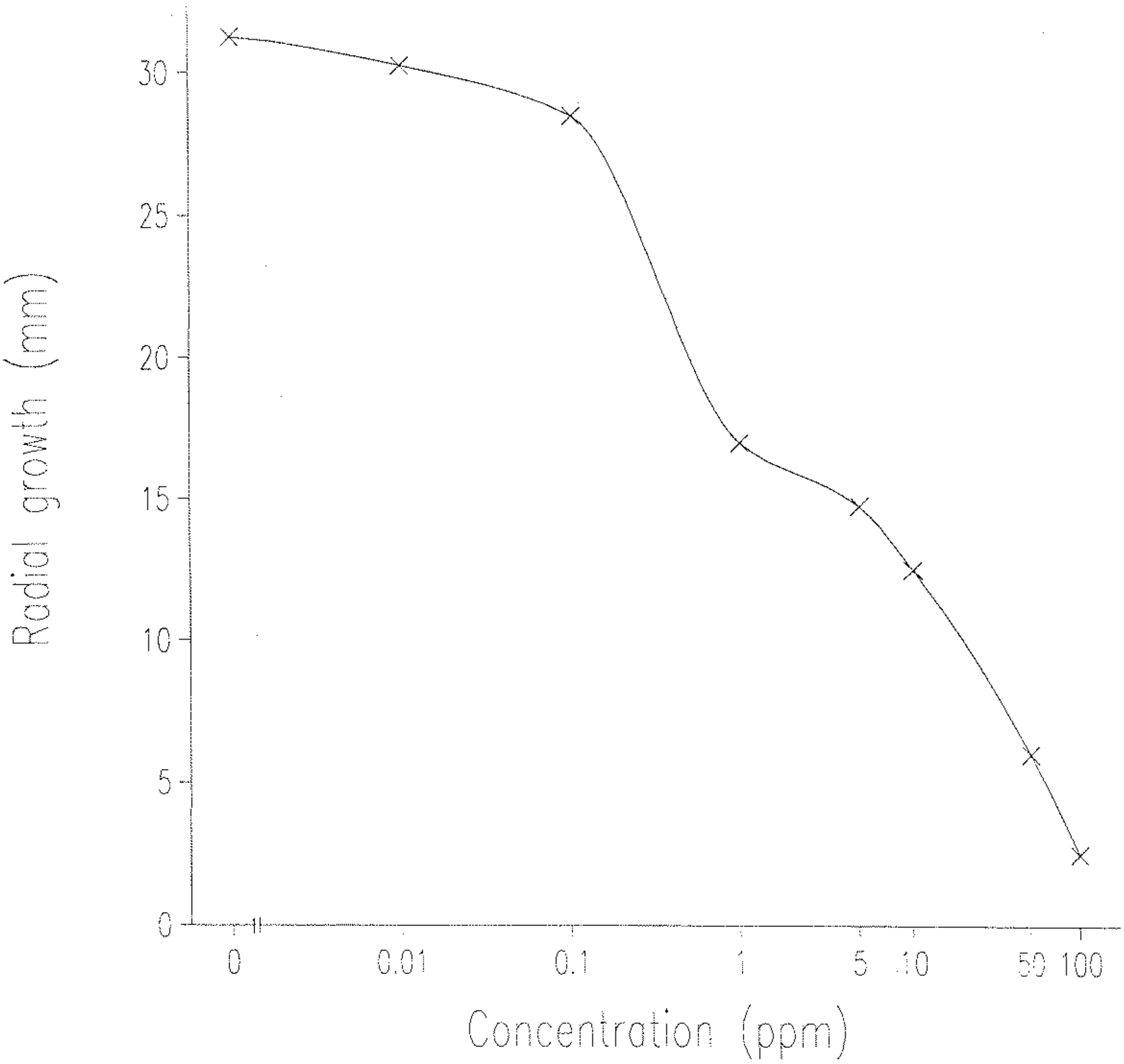
Figure 1. The response of *Pythium violae* to metalaxyl.



ppm=μg/ml

Appendix 5

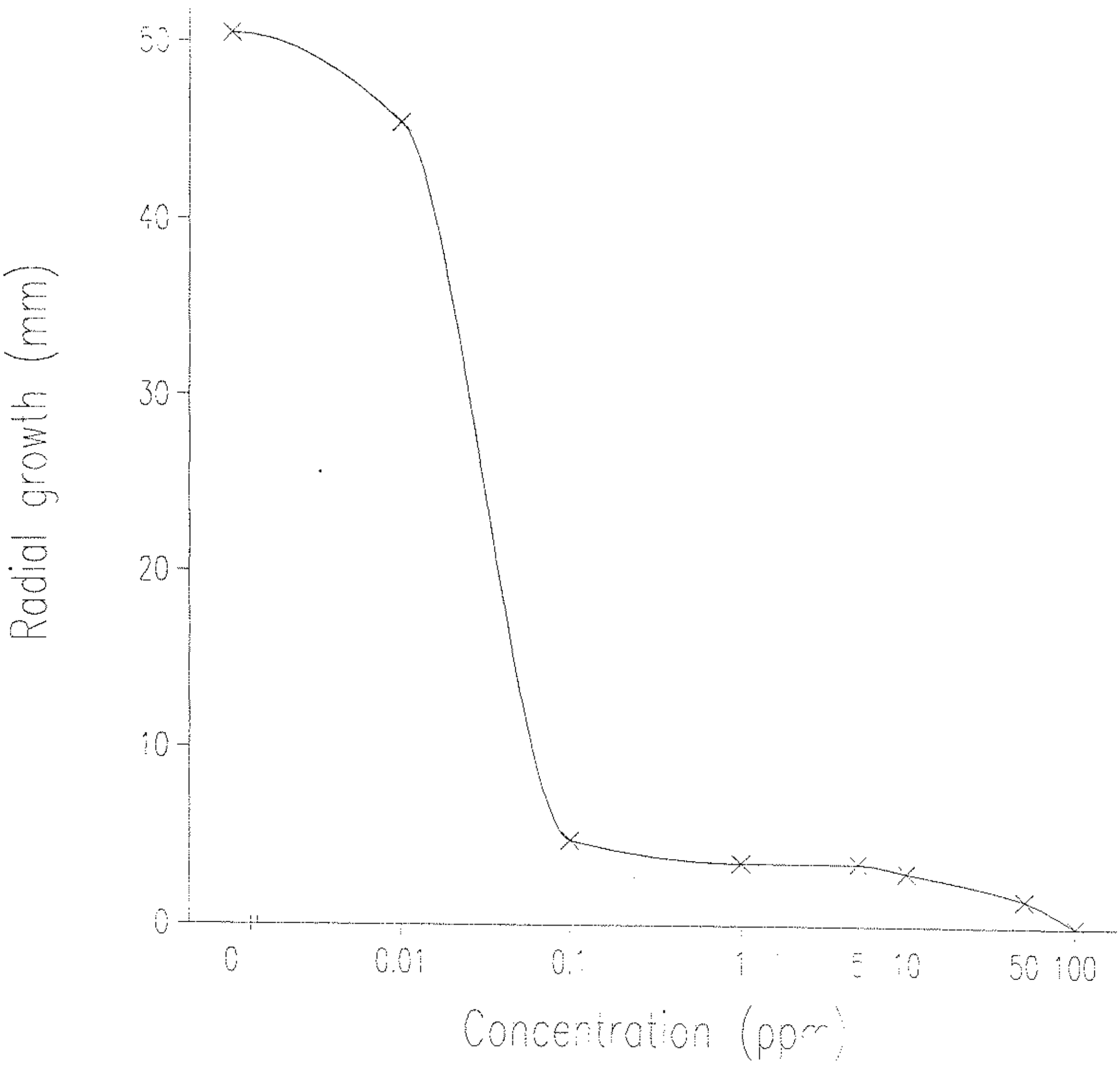
Figure 2. The response of *Pythium sulcatum* to metalaxyl.



ppm=μg/ml

Appendix 5

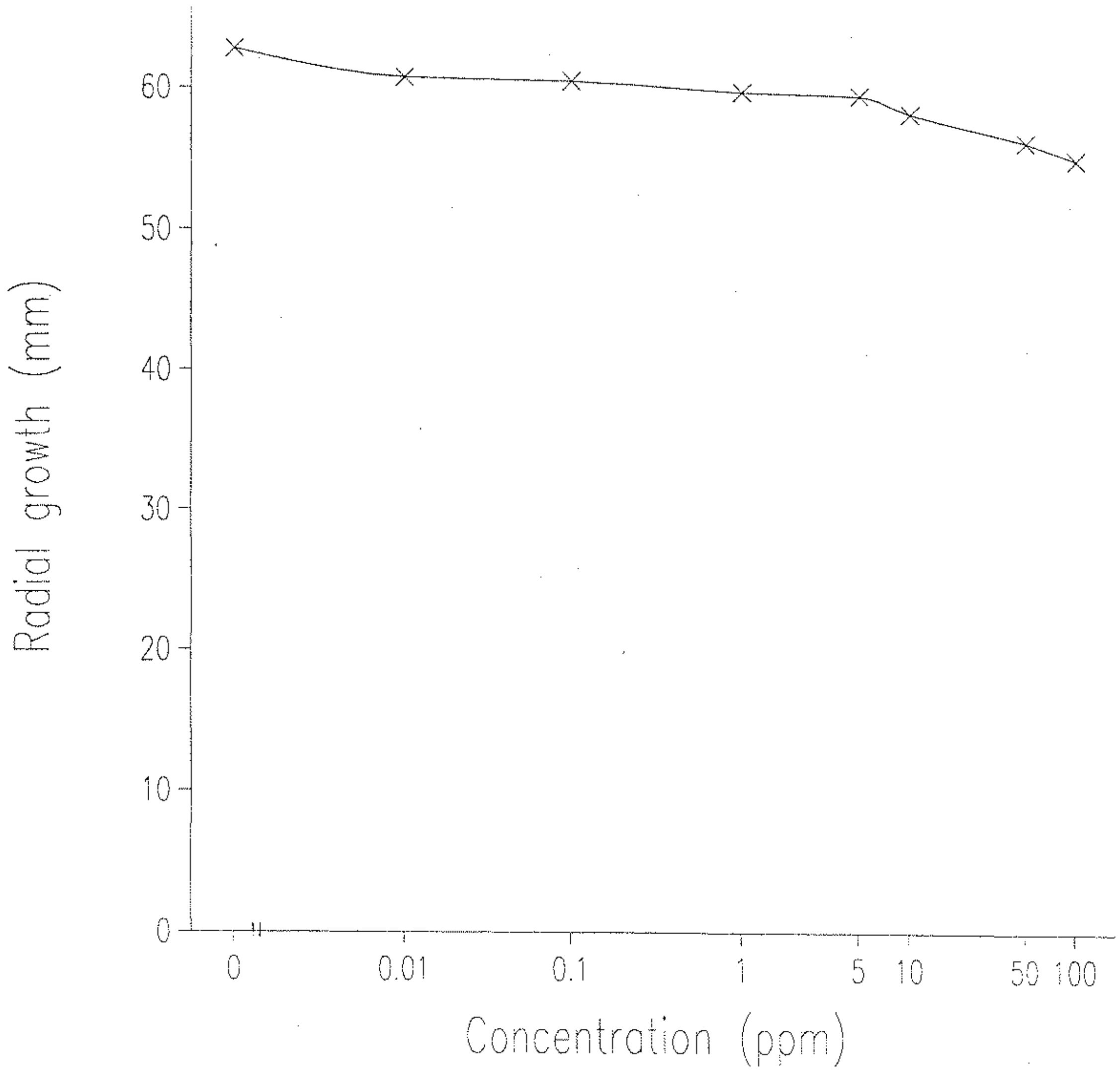
Figure 3. The response of *Pythium violae* to A9408B.



ppm=μg/ml

Appendix 5

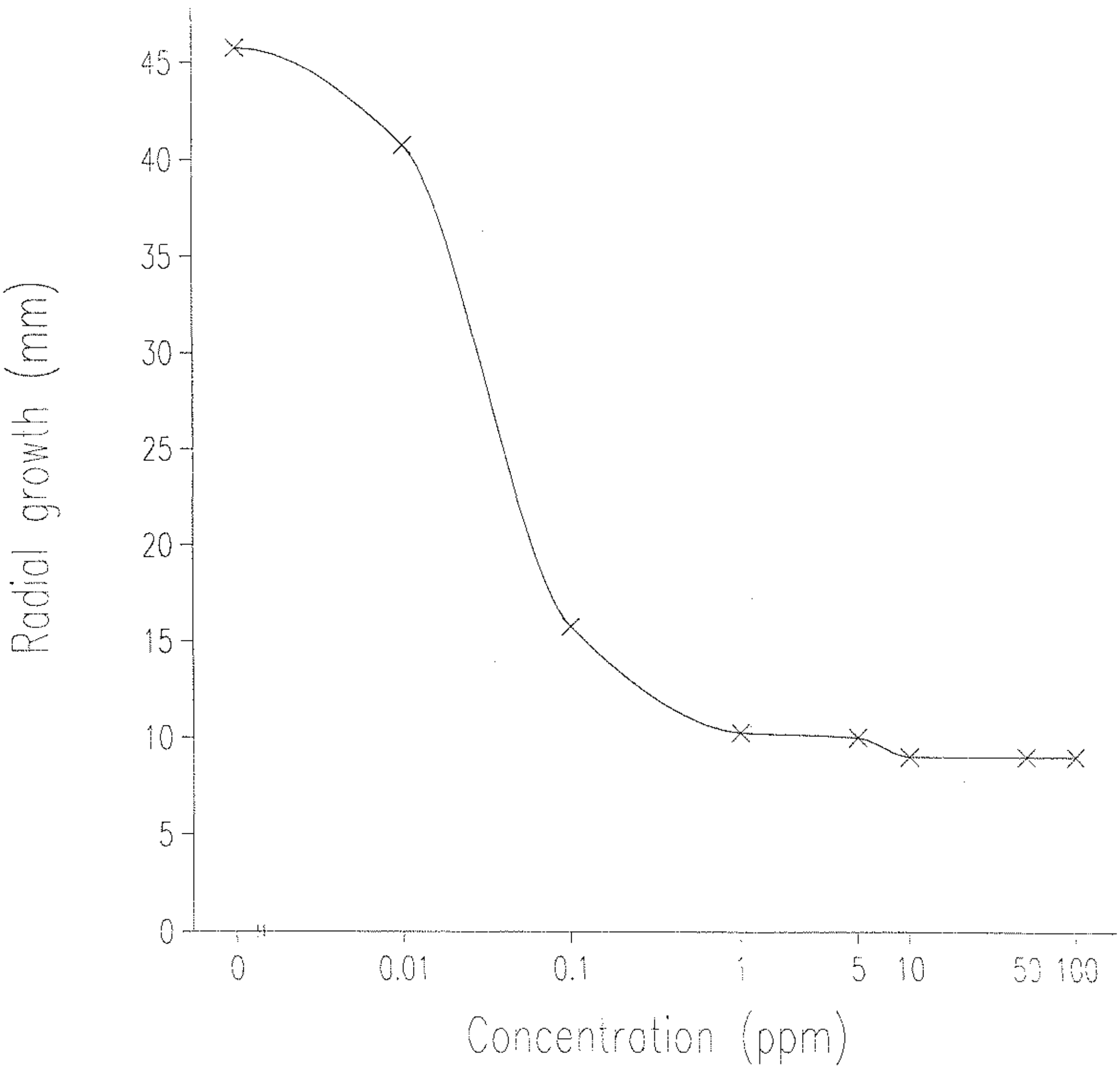
Figure 4. The response of *Pythium violae* to A9180A.



ppm=μg/ml

Appendix 5

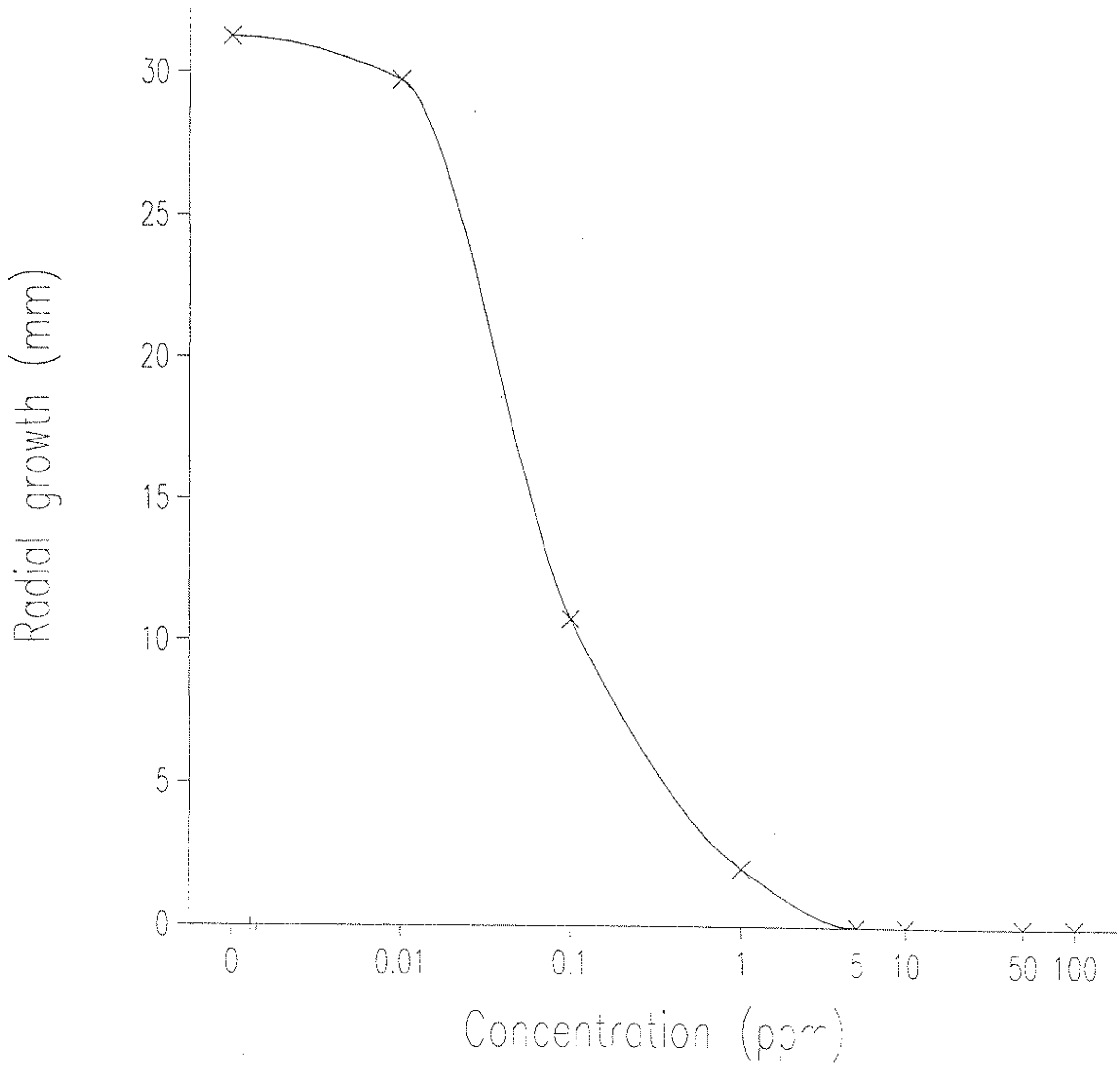
Figure 5. The response of *Pythium sulcatum* to hymexazole.



ppm=μg/ml

Appendix 5

Figure 6. The response of *Pythium sulcatum* to ICIA5504.



ppm=μg/ml

Appendix 6

Sowing, spraying and harvest dates in year 2.

Soil number	Sowing date	Spraying date	Harvest dates	Number of days to the harvest
1	1.8.1996	15.8.1996	11.12.1996	131
			8.1.1997	159
			4.2.1997	186
			5.3.1997	215
2	8.8.1996	21.8.1996	18.12.1996	131
			16.1.1997	160
			11.2.1997	186
			11.3.1997	214
3	22.8.1996	10.9.1996	9.1.1997	141
			5.2.1997	168

Appendix 4

Figure 1. Fanging effect from extreme attack of *Pythium violae* on carrots growing in compost artificially infested with the fungus

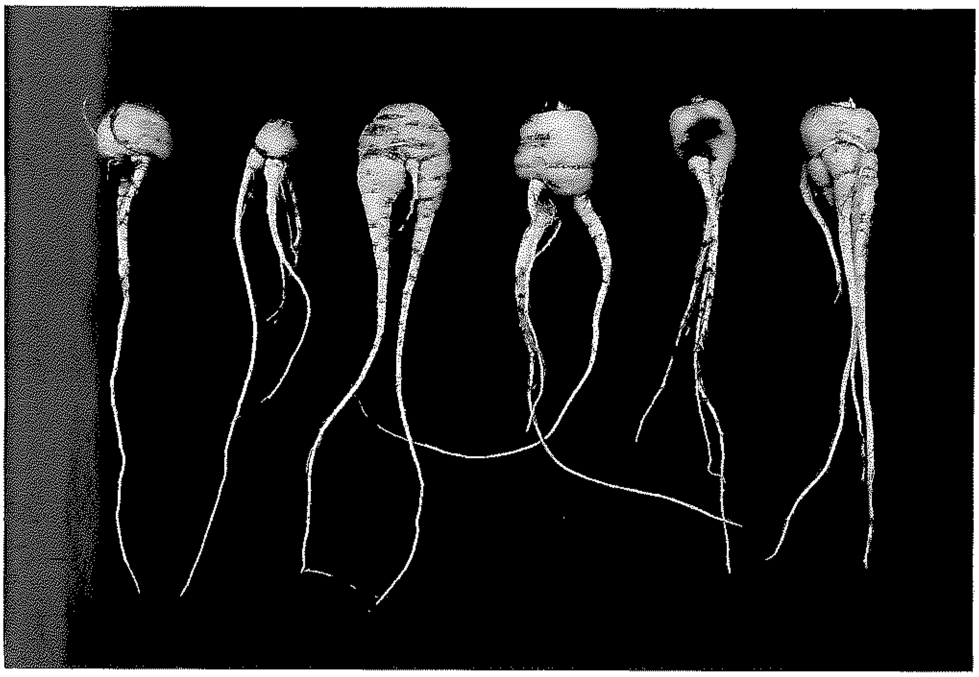


Figure 2. 'Cobby' growth of carrots resulting from infection by *Pythium sulcatum* when the carrots were grown in compost artificially infested with the fungus