

**New fungicides and strategies for
sustainable management of Sclerotinia
and Rhizoctonia diseases on
vegetable crops in A**

Dr Hoong Pung
Peracto Pty Ltd

Project Number: VG05090

VG05090

This report is published by Horticulture Australia Ltd to pass on information concerning horticultural research and development undertaken for the vegetables industry.

The research contained in this report was funded by Horticulture Australia Ltd with the financial support of Peracto Pty Ltd and the vegetables industry.

All expressions of opinion are not to be regarded as expressing the opinion of Horticulture Australia Ltd or any authority of the Australian Government.

The Company and the Australian Government accept no responsibility for any of the opinions or the accuracy of the information contained in this report and readers should rely upon their own enquiries in making decisions concerning their own interests.

ISBN 0 7341 2881 9

Published and distributed by:
Horticulture Australia Ltd
Level 7
179 Elizabeth Street
Sydney NSW 2000
Telephone: (02) 8295 2300
Fax: (02) 8295 2399

© Copyright 2012



Horticulture Australia



New fungicides and strategies for sustainable management of Sclerotinia and Rhizoctonia diseases on vegetable crops in Australia

HAL Project VG05090

**Final Report
15 May 2010**

By

Dr Hoong Pung

Peracto Pty Ltd

ABN: 97 109 472 559

Head Office: 16 Hillcrest Road
Devonport, Tas 7310 Australia

Telephone: +61 3 6423 2044

Facsimile: +61 3 6423 4876

Web: www.peracto.com

Project Number: VG05090

Report written by: Hoong Pung

Principal Investigator: Hoong Pung, Peracto Pty Ltd
16 Hillcrest Road, Devonport Tasmania 7310
Phone (03) 6423 2044
Fax (03) 6423 4876
Email: hpung@peracto.com

Report Date: 15 May 2010

Research personnel for investigative and efficacy trial studies

Hoong Pung, Susan Cross and Paul Florissen
Peracto Pty Ltd
16 Hillcrest Road
Devonport, Tasmania 7310

Research personnel for residue trial studies from Peracto Pty Ltd

Kate Allen, Bronwyn Haller, Jane Floyed and Belinda Ingram in Tasmania
Elizabeth Fields and Ryan Blackney in Victoria
Paul Florissen and Chris Monsour in Queensland
Richard Porter in South Australia
Mark Sumner in Western Australia

Laboratory analysis for chemical residue

Scott Winner
AgriSolutions Australia
75 Thompson Street
Deception Bay, Queensland 4508

Purpose of project

The main aim of this project was to conduct efficacy and residue trial studies to collate the necessary efficacy and residue data to expedite the registration of boscalid, a new fungicide active ingredient for long-term commercial use on vegetables in Australia. The other aim of the project was to investigate and develop disease management strategies for soilborne Rhizoctonia disease control in vegetables.

Acknowledgements

This project has been funded by HAL using the vegetable industry levy, voluntary contributions from Nufarm Australia Limited and matched funds from the Australian Government. The assistance of vegetable growers who provided the trial sites in Tasmania, Victoria, South Australia, New South Wales, Queensland, Western Australia and Victoria, is gratefully acknowledged.

Disclaimer

Any recommendations contained in this publication do not necessarily represent current Horticulture Australia Limited policy. No person should act on the basis of the contents of this publication, whether as to matters of fact or opinion or other content, without first obtaining specific, independent professional advice in respect of the matters set out in this publication.



Table of Contents

Media Summary	2
Technical Summary	3
PART I: INVESTIGATIONS ON RHIZOCTONIA DISEASES IN VEGETABLES AND DISEASE CONTROL METHODS	6
Introduction.....	6
1. Investigations on <i>Rhizoctonia solani</i> in cropping soils and vegetable crops	7
Summary	7
Introduction	7
Materials & methods.....	8
Results.....	10
Discussion	19
Conclusions	21
References	22
2. Screening of non-chemical soil treatments for suppressing Rhizoctonia in pot trials.....	23
Summary	23
Introduction.....	23
Materials & methods.....	23
Results.....	25
Discussion	27
3. A preliminary field study to evaluate gypsum and fungicide applications within a commercial green bean crop.....	28
Summary	28
Aims.....	28
Materials & methods.....	28
Results.....	29
Discussion	29
4. A field trial to evaluate pre-plant non-chemical soil treatments in Rhizoctonia inoculated soil.....	30
Summary	30
Aims.....	30
Materials & methods.....	30
Results & discussion	30
5. Screening of fungicides for Rhizoctonia control in pot trials	31
Summary	31
Introduction.....	31
Materials & methods.....	31
Results.....	33
Discussion	35
6. Field trials to evaluate various products applied as in-furrow spray soil applications.....	36
Summary	36
Aims.....	36
Materials & methods.....	36
Discussion	40
7. Effects of plant varieties and seed treatments	41
Summary	41
Introduction.....	41
Materials and methods	41
Results and discussion.....	42
General Discussion.....	45
Recommendations	47
Technology Transfer	48
Appendix	49

MEDIA SUMMARY

Sclerotinia diseases are major threats to the sustainable production of many vegetable crops in Australia. They affect a wide range of vegetable crops. The availability of effective fungicides is critical for the management of Sclerotinia diseases. Green beans and lettuces are the two main vegetable crops that are highly susceptible to Sclerotinia diseases, where losses can range from 20% to 100%. In 2004, when procymidone was withdrawn from use on bean and lettuce crops in Australia, boscalid was identified as a suitable replacement fungicide. Boscalid is a new active ingredient that had not been registered for use in any vegetable crops in Australia. Currently, it is used under a temporary permit on the condition that the product will be registered for long term use in Australia. This project was aimed at facilitating trial studies to collate the necessary efficacy and residue data to expedite the registration of this product for long-term commercial use on vegetables in Australia. Efficacy trials in this project showed that boscalid was as effective as procymidone for Sclerotinia control under most conditions. However, similar to procymidone and other fungicides, its level of disease control can be influenced by crop variety, plant vigour and weather conditions. Boscalid was also shown to be highly effective on Botrytis. Trials in this project also established that Amistar (azoxystrobin, Syngenta Crop Protection Pty Ltd) has little or no effect in controlling Sclerotinia. Under relatively dry conditions, Du-Wett, a new spray adjuvant, was shown to enhance disease control by Filan. Twenty-three residue trials were conducted to provide the necessary data to support Filan (boscalid, Nufarm Australia Limited) registration in Australia for long term use in vegetables that are susceptible to Sclerotinia diseases: leafy vegetable groups, legume vegetables, brassica vegetables and root vegetables. The residue trials were conducted under Good Laboratory Practice (GLP) in Tasmania, Queensland, Victoria, South Australia and Western Australia. Applications to the Australian Pesticides and Veterinary Medicines Authority (APVMA) are in progress and its registration for use in leafy vegetable groups, legume vegetables, brassica vegetables and root vegetables is expected in 2011.

Although *Rhizoctonia solani* is believed to be a limiting factor in crop productivity by many vegetable growers, there has been limited knowledge on the pathogen and its impact on vegetable crops. Not all Rhizoctonia can cause disease problems and it is important to find out which sub-groups are common in vegetable soils and whether they are pathogenic to vegetables. In this project, *R. solani* AG2.1 was the most common sub-group in vegetable soils, where it was found in 83% of the soil samples. AG2.1 was also shown to be the highly pathogenic to vegetables, causing severe damping off on peas, beans, cauliflowers and lettuces. In vegetables, there is also a lack of management options to control Rhizoctonia diseases. Trials were therefore conducted to evaluate novel non-chemical treatment methods as well as fungicides for their efficacy in controlling Rhizoctonia diseases. Seed treatments with azoxystrobin, fludioxonil or tolclofos-methyl were found to be more effective than Captan or Thiram in preventing early seedling damping off due to Rhizoctonia. Azoxystrobin and tolclofos-methyl applied as in-furrow soil applications at sowing were found to be the highly effective in preventing Rhizoctonia infections in infected soil. Azoxystrobin, applied as seed or in-furrow soil treatments caused a delay in seedling emergence, whereas tolclofos-methyl had no phytotoxic effects. Other non-chemical soil treatments including gypsum, molasses and biocontrol agents have little or no effect in soils inoculated with high levels of Rhizoctonia AG2.1.

TECHNICAL SUMMARY

PART I: Investigations on Rhizoctonia diseases in vegetables and disease control

Although Rhizoctonia is believed to be a limiting factor in crop productivity by many vegetable growers, there has been limited knowledge on its distribution and impact on vegetable crops. Previous research on Rhizoctonia diseases had been hampered by the complexity of the pathogen and the difficulties in studying the impact of the disease below ground. *R. solani* is not a single species, but a collection of non-interbreeding populations. Not all Rhizoctonia can cause disease problems and it is important to find out which sub-groups are common in vegetable soils and understand their impact in vegetable crop production. In this project, soil samples from intensive vegetable cropped fields were investigated to gain a better understanding of Rhizoctonia and their pathogenicity on vegetables. DNA soil tests showed that *R. solani* AG2.1 was the most common sub-group in vegetable soils, where it was found in 83% of the soil samples. AG3 and AG4 were found in 35% and 25% of the soil samples, respectively. AG2.2 was rarely detected. AG2.1 was also showed to be the highly pathogenic to vegetables, causing severe damping off on peas, beans, cauliflowers and lettuces. *R. solani* AG2.2 and AG4 were also pathogenic. However, AG3, a major pathogen on potato, was not pathogenic to these vegetables.

In a bioassay study on soils from 24 paddocks to bait root pathogens using green beans, Rhizoctonia was found in association with bean root rot in 63% of the soil samples. Under cold and wet conditions, seedling establishment from the untreated green bean seeds was very poor with less than 46% plants surviving in 42% of the soil samples. In paddocks, which had been used intensively for vegetable production including green beans, other bean root pathogens, namely *Aphanomyces euteiches*, *Fusarium* spp., *Pythium* spp. and *Thielaviopsis basicola*, were also present. This demonstrates that the causes of root diseases can be complex, and that often more than one root pathogen was involved. Seed quality and field conditions are also factors that can reduce crop establishment. It is therefore, vital to identify all causal pathogens, as well as non-pathological factors, before disease control measures are considered.

Because of a lack of understanding on Rhizoctonia diseases in vegetables, there is a lack of management options to prevent Rhizoctonia damping off as well as root and stem infections. Therefore, many trials were conducted in this project to evaluate novel non-chemical treatment methods as well as fungicides for their efficacy in controlling Rhizoctonia diseases. The trials were conducted in soils inoculated with *R. solani* AG2.1 in pots and in the field using peas and green beans. *R. solani* AG2.1 were shown to cause severe pre- and post-emergence damping off in the inoculated soils. On green beans, it also restricts root development, hence reducing plant growth. Under ideal humid and warm conditions, it also causes above-ground infections on lower stems and bean pods.

Non-fungicide treatments such as gypsum, molasses, saw dust, sugar, bacteria and fungal biocontrol agents were screened for Rhizoctonia control in inoculated pot soil. Sawdust mixed into soil at 3% w/v had the highest rate of seedling survival, with 74% survival at 46 days after sowing (DAS). Sawdust applied at 1% w/v and molasses at 1% and 1.5% w/v also increased seedling survival to a range of 30% to 34% compared to 6% in the untreated control. The rates of molasses and sawdust applied in the trial studies were very high and hence are not practical for commercial use. However, the promising results in some studies indicate the importance of the role of organic matter in suppressing Rhizoctonia diseases. Methods that can increase organic matter in soil such as green manure, re-cycling of organic waste products and various organic amendments may be useful in suppressing low to moderate levels of soilborne pathogens.

Gypsum granules mixed into pot soil at 2% to 5% w/v showed some activity for Rhizoctonia control, but it was not as effective as molasses. Gypsum applied onto the soil surface and then drenched into the soil had no effect. Gypsum is relatively insoluble and therefore must be mixed thoroughly into soil so that it can come into contact with the fungal pathogen in order to have any effect. Unlike fungicide chemicals, gypsum does not completely inhibit growth. Instead, it only suppresses the pathogen by delaying its mycelial spread. These findings showed the limitations of using gypsum for Rhizoctonia control as it must be applied at very high rates and be well distributed in soil. It is possible that gypsum soil application may be more effective in suppressing low levels of the pathogen that is more likely to occur naturally in the field. Crops may also benefit from gypsum soil application, as it is also a fertiliser and soil improver. The biocontrol agents based on *Bacillus lydicus* and *Trichoderma* spp. had little or no effect on Rhizoctonia, under high disease levels in soils.

In initial screening trials, the fungicides, azoxystrobin (Amistar, Syngenta Crop Protection Pty Ltd), phosphorous acid (Agri Fos, Agrichem Industries Pty Ltd), boscalid (Filan, Nufarm Australia Limited), fludioxonil + cyprodinil (Switch, Syngenta Crop Protection Pty Ltd), propiconazole + cyproconazole (Tilt, Syngenta Crop Protection Pty Ltd) and tolclofos-methyl (Rizolex, Sumitomo Chemical Australia Pty Ltd) showed potential for controlling *Rhizoctonia* and increasing seedling survival and plant growth in a pot trial. Not all fungicides, however, are suitable for soil applications and only Amistar and Rizolex were selected for further studies. In two *Rhizoctonia* inoculated field trials, Amistar and Rizolex applied as in-furrow soil applications at sowing, were also found to be the highly effective in preventing *Rhizoctonia* infections and consistently increased green bean seedling establishment and growth. Rizolex applied at 2 L/ha and Amistar applied at 4 L/ha or 10 mL/100 m row, as in-furrow spray applications, were effective in controlling the pathogen. Rizolex was less effective at the lower rate of 1 L/ha. Amistar soil treatment was observed to cause a delay in seedling emergence, whereas Rizolex had no such phytotoxic effect. Therefore, Rizolex was the safer fungicide to use. This product is currently registered for use in potatoes as seed piece treatment and as in furrow soil application for *Rhizoctonia* control.

Bean seed treatments containing azoxystrobin, fludioxonil or tolclofos-methyl were found to be much more effective than Captan or Thiram in preventing early seedling damping off due to *Rhizoctonia*. Azoxystrobin seed treatment may also delay seedling growth.

PART II: Development of boscalid for Sclerotinia disease control in vegetables

Sclerotinia diseases by *Sclerotinia sclerotiorum* and *S. minor* are major threats to the sustainable production of many vegetable crops in Australia. They affect a wide range of vegetable crops, such as green beans, brassicas, carrots, lettuces, peas, potatoes, swedes and turnips. Green beans and lettuces are the two main vegetable crops that are highly susceptible to Sclerotinia diseases, where losses can range from 20% to 100%. Until 2004, many growers relied solely on procymidone (sold as Sumisclex and Fortress) for Sclerotinia control as it gave very consistent and effective control. In late 2004, procymidone was withdrawn from use on green bean and lettuce crops in Australia, due to concerns on its safety to human health and the environment. Early trial studies conducted in 2002 and 2003, showed that boscalid (Filan) was as effective as procymidone against *S. minor* and *S. sclerotiorum* on green bean and lettuce crops.

Boscalid is a new class of fungicide, benanilide, that had not been registered for use in any vegetable crops in Australia. Currently, it is used on green beans and lettuces under a temporary permit, on the condition that the product will soon be registered for long term use in Australia. The aim of the second part of this project was to carry out trial studies to collate the necessary data in order to expedite the application to register this product for long-term commercial use on vegetables in Australia.

In this project, eight efficacy trials were conducted in Tasmania and Queensland on green beans and lettuces to establish that Filan is effective in controlling Sclerotinia diseases. In all except for one trial at Gympie, boscalid was shown to be as effective as procymidone for Sclerotinia control. The level of disease control by the fungicide, however, can be influenced by crop variety, plant vigour and weather conditions. In Gympie, Queensland, when highly susceptible bean cultivars were sown under extremely high disease pressure and favourable weather conditions in June to July, procymidone was more effective than boscalid.

Boscalid was also shown to be highly effective on Botrytis. Trials in this project also showed that Amistar (azoxystrobin) has little or no effect in controlling Sclerotinia.

A field trial was conducted to evaluate the potential of a new organosilicone and organic blend of spray adjuvant, Du-Wett™. It was compared against Activator™, which was the most commonly used non-ionic surfactant used with Filan™ spray applications for Sclerotinia control on green beans in Tasmania. Du-Wett was shown to enhance disease control by Filan, under relatively dry conditions. Activator did not improve the disease control by Filan, but showed a trend of slightly more disease, when it was applied with Filan. Following this trial study, green bean growers in Tasmania had stopped using Activator as a spray adjuvant with the fungicide applications.

Twenty-three residue studies were conducted to establish the Maximum Residue Limits (MRLs) for various types of vegetables: leafy vegetable groups, legume vegetables, brassica vegetables and root vegetables. Field residue trials were conducted under Good Laboratory Practice (GLP) in Tasmania, Queensland, Victoria, South Australia and Western Australia. All chemical analysis of plant samples from

the residue trials were carried out in Queensland, under GLP. Detailed reports on all the efficacy and residue trial studies and chemical analysis have been completed and sent to Nufarm Australia Limited. Applications to the APVMA are in progress and the fungicide is expected to be registered for use in 2011 on leafy vegetable groups, legume vegetables, brassica vegetables and root vegetables.

PART I: INVESTIGATIONS ON RHIZOCTONIA DISEASES IN VEGETABLES AND DISEASE CONTROL METHODS



Introduction

The aim of Part I of this project was to conduct investigations to gain a better understanding of Rhizoctonia disease in vegetables and to conduct feasibility studies on potential fungicides, biofungicides as well as other novel methods for Rhizoctonia control.

Growers identify Rhizoctonia as an increasingly major threat to the sustainable production of many vegetable crops in Australia. *Rhizoctonia solani* is the most widely recognised species of Rhizoctonia, and was identified initially on potato plants in 1858 by Julius Kühn (Ceresini 1999). *Rhizoctonia solani* can reproduce sexually but generally exists as mycelium and/or sclerotia. *Thanatephorus cucumeris* is the teleomorph or sexual stage of the pathogen. Knowledge on the pathogen and its impact on vegetable crops has been limited. Previous research on Rhizoctonia diseases had been hampered by the complexity of the pathogen and the difficulties in studying the impact of the disease below ground. Studies have shown that *Rhizoctonia solani* is a group of fungi that looked similar in taxonomy, but actually consists of many genetically distinct fungal groups (Anderson 1982, Carling & Leiner 1986, Ogoshi 1987, Ceresini 1999). Approximately 12 of these AG groups have been described, of which AG2.1, AG2.2, AG3, AG4 and AG8 are commonly associated with vegetable and cereal crop diseases. The pathogenicity of some AG groups can be specific to plant host type, while others have a relatively wide range of hosts (Anderson 1982). Apart from research on potatoes, there have been little or no studies on the type of *R. solani* AG groups, inoculum density or pathogenicity on the major vegetable crops in Australia. Hyphal anastomosis (a fusion of hyphae together to establish their relatedness) has been used to identify and group similar types after their isolation from infected host plant tissues. The determination of inoculum density of the pathogen in soils is based on baiting with susceptible host plants, seeds, stem sections or other organic materials. More recently, molecular methods developed to identify the AG groups have facilitated research on them in plants and soils.

With the lack of understanding on the levels and types of Rhizoctonia sub-groups, there is a lack of management options to prevent Rhizoctonia damping off as well as root and stem infections on maturing plants. Some fungicides such as azoxystrobin, fludioxonil and tolclofos-methyl have been found to be effective against Rhizoctonia on major crops such as potato and cereal crops, but there is little or no interest to examine their potential use on vegetables. Calcium sulphate, based on natural gypsum, had been shown to have potential for use to help suppress Sclerotinia wilt disease in pyrethrum. Field trials over the past five years have shown that calcium sulphate applied with procymidone can enhance Sclerotinia control, enhance infected plant recovery and increase yields, when compared to fungicide alone. Similar field trials on broad acre crops like soya bean and canola carried out in the USA also showed the synergistic effects of calcium sulphate when applied with fungicides. Since 2003, the pyrethrum industry in Tasmania has adopted the use of calcium sulphate plus procymidone mixture for Sclerotinia wilt disease control. In 2004, many green bean growers also started applying a mixture of calcium sulphate plus boscalid for Sclerotinia control with satisfactory results. Recently, *in vitro* studies conducted in Tasmania showed that calcium sulphate could help delay the pathogen's mycelial growth as well as sclerotia production of both Sclerotinia and Rhizoctonia.

1. Investigations on *Rhizoctonia solani* in cropping soils and vegetable crops

Summary

Soil tests were conducted in 2006 and 2007, on soils from 40 paddocks that had been used intensively for vegetable crop production for the presence and levels of important *Rhizoctonia solani* sub-groups. AG2.1 was the most common *R. solani* sub-group in the soil samples from 40 paddocks, being detected in 83% of paddocks. The other *R. solani* sub-groups, AG3, AG4 and AG2.2 were detected in 35%, 25% and 8% of the paddocks, respectively. AG8, which can seriously reduce yield in cereal crops, was not detected in any of the soil samples. The effects of each sub-group on various vegetable crops are currently being examined in further studies in order to get a better understanding of their impact on various vegetable crops.

In bioassay tests of soils from 24 paddocks to bait root pathogens with green beans, almost all the soils had root pathogens. *Rhizoctonia* was found in association with bean root rot in 63% of the soil samples. In 42% of the soil samples, only the *Rhizoctonia* pathogen could be observed on the root rots. In paddocks which had been used more intensively for bean production, root rot by other pathogens, namely *Thielaviopsis basicola* and *Aphanomyces euteiches*, two major root pathogens on beans were also present. Under cold and wet conditions in pots, seedling establishment from the untreated green bean seeds was very poor. Seedling emergence and survival was less than 46% in 42% of the soil samples. This study demonstrated that the causes of root diseases can be complex, and it is vital to identify all causal pathogens, as well as non-pathological factors, before disease control measures are considered. Further studies are currently being conducted under the new green bean project VG08043 to develop methods for detecting *Thielaviopsis basicola* and *Aphanomyces euteiches* in soil samples.

Introduction

Increasingly, *R. solani* has been recognised as consisting of a collection of fungal isolates that look similar in taxonomy but are different genetically (Anderson 1982). A concept of using anastomosis groups (AG) to describe each unique *R. solani* type based on hyphal anastomosis (a fusion of hyphae together to establish their relatedness) has now gained wide acceptance among plant pathologists (Anderson 1982, Ogoshi 1987). More recently, molecular methods based on DNA analysis, developed to assist in identifying the separate AG sub-groups of *R. solani*, have facilitated research in identification, pathogenicity, host specificity and economic impact of the different sub-groups.

Even though *R. solani* has often been associated with root rot and yield decline in vegetable crops, there have been little or no studies to establish or confirm their presence and impact on crop productivity. Therefore, preliminary studies were conducted in this section to test and identify the type and levels of *R. solani* present in paddocks that had been intensively used for vegetable crop production. Although up to 12 AG groups have been described, many are non-pathogens or are crop specific. The SARDI DNA tests used in this study were aimed at determining the presence of AG2.1, AG2.2, AG3, AG4 and AG8, the sub-groups that are identified as being associated with important potato and cereal crop diseases.

In addition to soil tests for *Rhizoctonia* sub-groups, additional quantitative and qualitative data were collated with bioassay tests to bait root pathogens and assess their impact on seedling growth of a vegetable crop, as well as to conduct field examinations to observe crop growth and determine causes of poor crop growth. Green beans were used as a benchmark vegetable crop in this study, because of their high susceptibility to a wide range of root pathogens. Pathogenicity tests were also conducted in soil inoculated with the selected AG groups.

Materials & methods

Soil sampling

Soil samples were collected from 40 paddocks in Tasmania, Victoria and Queensland. Each sample consisted of an aggregate of 20 soil core samples taken to a depth of 15 cm in a 'W' formation across each paddock, which were then bulked together before use for soil tests or bioassays. Soil tests based on DNA analysis for *R. solani* sub-groups and their levels were conducted at the SARDI laboratory for DNA analysis. The SARDI DNA tests were developed to determine the presence and levels of AG2.1, AG2.2, AG3, AG4 and AG8, which represent the most studied sub-groups that are associated with important crop diseases. Where possible, DNA tests for other pathogens or pests, such as *Spongospora subterranea* (powdery scab), *Streptomyces scabies* (common scab), *Verticillium dahliae* (*Verticillium* wilt) and *Pratylenchus* nematode species, were also conducted for growers' interest.

The locations of the paddocks were marked using a global positioning system (GPS) for future reference. Most of the paddocks were selected because they have been used for intensive vegetable and potato production, and yield decline has been experienced on these soils. Many of the paddocks were sown with green beans in the 2006/07 season. Roots of green bean plants are generally highly susceptible to stem, hypocotyl and root rot by various soilborne pathogens, including *Rhizoctonia*, and hence are ideal for studies on root pathogens and their impact on crop establishment, growth and yield.

Bioassay tests

Bioassay tests were conducted in a pot trial using green beans, with 24 soil samples collected initially in October 2006 (Sample No. 1-24) to bait *R. solani* and other root pathogens. Twenty green bean seeds were sown in each pot, and there was only one pot per soil sample. At 19, 27 and 43 days after sowing (19DAS, 27DAS and 43DAS), seedling emergence and survival were recorded as a percentage of the total number of seeds initially sown. At 43DAS, surviving plants were also assessed for fresh shoot weights and roots were rated for root rot severity. Thin sections of roots were examined for root pathogens. Roots of surviving plants were assessed for root rot severity as described below.

Root rot severity ratings:

- 0 = no hypocotyl discoloration & no root rot
- 1 = some superficial hypocotyl rot, light root pruning, with good root branching
- 2 = superficial hypocotyl rot and moderate root pruning
- 3 = severe hypocotyl rot and moderate root pruning
- 4 = severe hypocotyl rot and severe root pruning
- 5 = severe stunted or dying plant with very small roots

Root rot severity index = $\frac{[(1 \times \text{no. plants in rating 1}) + \dots + (5 \times \text{no. plants in rating 5})]}{\text{no. surviving plants}} \times 100$

Field observations

Paddocks where the soil samples were collected (above) were also monitored after sowing with green beans, carrots, onions and potatoes. Roots of plants were examined for root rot and *R. solani*, as well as for the presence of other root pathogens. Qualitative descriptions on the establishment and growth of the commercial crops were also recorded.

Pathogenicity study

Garden pea, green bean, cauliflower and lettuce were evaluated in sandy loam soil that was inoculated with five different sub-groups of *R. solani* in 3 L pots. Two isolates of AG2.1 were used in the study – one was isolated from an infected bean stem and another from a potato stem. All pots, except the uninoculated control were inoculated with each *R. solani* sub-group by mixing Rhizoctonia colonised millet seeds into the top 2 L of soil at a rate of 30 g/pot (1.5% w/v). After inoculation with Rhizoctonia, the soil was wet to field capacity and each pot was sown with 20 untreated vegetable seeds. The study was set up outdoors during spring time in 2007. The trial design for each crop was randomised complete block with four replicates. Seedlings were counted and the percentages of seedlings that survived were then tabulated from the number of seed sown.

Results

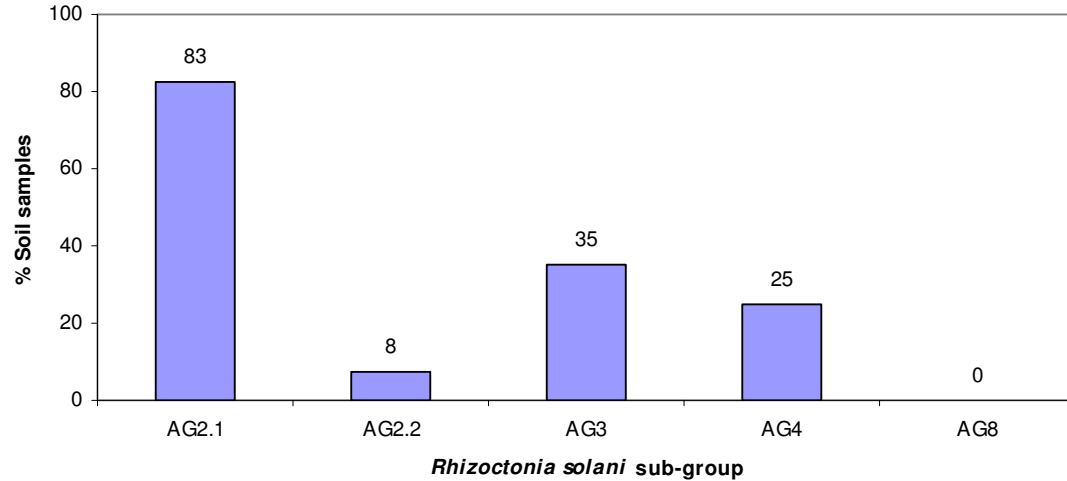
Table 1.1 - Soil tests for *R. solani* AG groups and common soilborne pathogens in 40 intensively cropped paddocks

Sample No.	Sample Code	Grower	Cropping practice	Soil type	Location	pg DNA / g soil										
						AG2.1	AG2.2	AG3	AG4	AG8	<i>Spongospora subterranea</i>	<i>Streptomyces scabies</i>	<i>V. dahliae</i>	<i>P. neglectus</i>	<i>P. thornei</i>	<i>P. penetrans</i>
1	AA63283	NB	vegetable, pasture, potato	Red Ferrosol	Sassafras, TAS	258	0	9	27	0	798	0	na	na	na	na
2	AA63284	NB	vegetable, pasture, potato	Red Ferrosol	Sassafras, TAS	2227	0	8	0	0	5860	0	na	na	na	na
3	AA63285	AM	vegetable, pasture, potato	Red Ferrosol	Sassafras, TAS	174	0	0	281	0	9670	0	na	na	na	na
4	AA63286	AM	vegetable, pasture, potato	Red Ferrosol	Sassafras, TAS	144	0	103	0	0	26038	0	na	na	na	na
5	AA63287	DP	vegetable, pasture, potato	Red Ferrosol	Sassafras, TAS	612	0	0	60	0	42702	0	na			
6	AA63288	GR	vegetable, pasture, potato	Red Ferrosol	Sassafras, TAS	49	0	0	0	0	36692	0	na	na	na	na
7	AA63289	MR	vegetable, pasture, potato	Red Ferrosol	Thirlstane, TAS	102	0	718	14276	0	80	0	na	na	na	na
8	AA63290	JP	vegetable, pasture, potato	Red Ferrosol	East Sassafras, TAS	1	0	0	45	0	6209	0	na	na	na	na
9	AA63291	JP	vegetable, pasture, potato	Red Ferrosol	East Sassafras TAS	7	0	5	24	0	15748	0	na	na	na	na
10	AA63292	JP	vegetable, pasture, potato	Red Ferrosol	East Sassafras TAS	0	0	165	0	0	21005	0	na	na	na	na
11	AA63282	LB	vegetable, pasture, potato	Red Ferrosol	Forth, TAS	14	0	2	33	0	3189	0	na	na	na	na
12	AA63276	DB	vegetable, pasture, potato	Red Ferrosol	Upper Burnie, TAS	525	0	66	0	0	31195	14	13	0	0	1
13	AA63277	BC	vegetable, pasture, potato	Red Ferrosol	Flowerdale, TAS	33	0	0	0	0	84	83	0	0	0	0
14	AA63278	WE	vegetable, pasture, potato	Brown sandy clay loam	Flowerdale, TAS	88	5	0	0	0	27259	0	13	0	0	0
15	AA63279	BH	vegetable, pasture, potato	Red Ferrosol	Sisters Creek, TAS	16	0	6	0	0	14482	0	0	0	0	0
16	AA63280	BH	vegetable, pasture, potato	Red Ferrosol	Sisters Hills, TAS	7364	0	0	0	0	17283	0	0	0	0	0
17	AA63281	CD	vegetable, pasture, potato	Red Ferrosol	Sisters Hills, TAS	183	0	0	0	0	11009	288	1	0	0	55
18	AA63293	CD	vegetable, pasture, potato	Red Ferrosol	Sisters Hills, TAS	1	0	0	0	0	75619	32	0	0	0	0
19	AA63294	JC	vegetable, pasture, potato	Red Ferrosol	Sisters Hills, TAS	54	0	8	0	0	70389	0	0	0	0	0

Table 1.2 - Soil tests for *R. solani* AG groups and common soilborne pathogens in 40 intensively cropped paddocks (Cont.)

Sample No.	Sample Code	Grower	Cropping practice	Soil type	Location	pg DNA / g soil										
						AG2.1	AG2.2	AG3	AG4	AG8	<i>Sporogospira subterranea</i>	<i>Streptomyces scabies</i>	<i>V. dahliae</i>	<i>P. neglectus</i>	<i>P. thornei</i>	<i>P. penetrans</i>
20	AA63295	JC	vegetable, pasture, potato	Red Ferrosol	Montumana, TAS	404	0	36	0	0	53593	0	1	0	0	0
21	AA63465	JC	vegetable, pasture, potato	Red Ferrosol	Sisters Hills, TAS	952	0	20	30	0	4059	0	0	1	0	0
22	AA63466	BH	vegetable, pasture, potato	Red Ferrosol	Boat Harbour, TAS	679	0	0	0	0	28055	0	24	0	0	0
23	AA63467	BH	new ground, one potato crop	Grey sandy loam	Wynyard, TAS	24	427	0	0	0	19	0	0	0	0	0
24	AA63468	JB	vegetable, pasture, potato	Red Ferrosol	Sassafras, TAS	2	0	0	0	n/a	1626	0	na	na	na	na
25	AA63469	RB	pasture, vegetable, cereals, potato between long break	Red Ferrosol	Forthside, TAS	0	0	0	181	n/a	3	0	na	na	na	na
26	AA63470	NW	pasture, vegetable, potato between long breaks	Red Ferrosol	Table Cape, TAS	0	0	0	0	n/a	90	0	na	na	na	na
27	AA63471	OT	vegetable, pasture, potato	Red Ferrosol	Wesley Vale, TAS	0	0	0	0	n/a	26370	62	na	na	na	na
28	AA63472	GR	vegetable	Sandy loam	Heatherton, Vic	0	0	0	0	n/a	0	67	na	na	na	na
29	AA63473	HF-C	vegetable	Sandy clay loam	Cambridge, TAS	2	0	0	0	n/a	0	42	na	na	na	na
30	AA63474	HF-C	vegetable	Sandy clay loam	Cambridge, TAS	26	0	0	653	n/a	0	63	na	na	na	na
31	AA63475	HF-R	vegetable	Clay loam	Richmond, TAS	100	0	0	0	n/a	11	0	na	na	na	na
32	AA63476	HF-R	vegetable	Clay loam	Richmond, TAS	8	0	0	0	n/a	0	0	na	na	na	na
33	AA63477	QL	vegetable	Clay loam	Glenore Grove, Qld	0	0	0	0	n/a	0	0	na	na	na	na
34	AA63478	DS	vegetable	Black clay	Gatton, Qld	0	0	0	0	n/a	0	0	na	na	na	na
35	AA63479	AR	vegetable, pasture, potato	Red Ferrosol	Natone, TAS	124	0	37	0	0	90716	0	na	na	na	na
36	AA63480	AR	vegetable, pasture, potato	Red Ferrosol	Natone, TAS	7	1	0	0	0	102794	0	na	na	na	na
37	AA63481	AR	vegetable, pasture, potato	Red Ferrosol	Natone, TAS	30	0	0	0	0	54659	115	na	na	na	na
38	AA63482	AR	vegetable, pasture, potato	Red Ferrosol	Natone, TAS	2	0	0	0	0	78386	0	na	na	na	na
39	AA63483	AR	vegetable, pasture, potato	Red Ferrosol	Natone, TAS	458	0	0	0	0	30612	0	na	na	na	na
40	AA63484	AR	vegetable, pasture, potato	Red Ferrosol	Natone, TAS	987	0	4	0	0	41776	37	na	na	na	na

n/a = not available because not tested for it

Figure 1.1 - The frequency of *Rhizoctonia solani* anastomosis groups in 40 soil samples**Table 1.3 - *R. solani* sub-groups in soil tests, and seedling emergence and survival in bioassay tests with the same soils from 24 paddocks**

Sample No.	Sample Code	Soil type	<i>Rhizoctonia solani</i> (pg DNA / g soil)					% Seedling emergence (19DAS)	% survival (27DAS)	% survival (43DAS)
			AG2.1	AG2.2	AG3	AG4	AG8			
1	AA63283	Red Ferrosol	258	0	9	27	0	85	85	75
2	AA63284	Red Ferrosol	2227	0	8	0	0	50	50	45
3	AA63285	Red Ferrosol	174	0	0	281	0	65	65	65
4	AA63286	Red Ferrosol	144	0	103	0	0	65	65	65
5	AA63287	Red Ferrosol	612	0	0	60	0	60	70	55
6	AA63288	Red Ferrosol	49	0	0	0	0	70	70	70
7	AA63289	Red Ferrosol	102	0	718	14276	0	40	40	40
8	AA63290	Red Ferrosol	1	0	0	45	0	30	30	30
9	AA63291	Red Ferrosol	7	0	5	24	0	45	55	55
10	AA63292	Red Ferrosol	0	0	165	0	0	50	55	55
11	AA63282	Red Ferrosol	14	0	2	33	0	50	50	50
12	AA63276	Red Ferrosol	525	0	66	0	0	30	35	35
13	AA63277	Red Ferrosol	33	0	0	0	0	60	60	60
14	AA63278	Brown sandy clay loam	88	5	0	0	0	30	30	30
15	AA63279	Red Ferrosol	16	0	6	0	0	40	50	50
16	AA63280	Red Ferrosol	7364	0	0	0	0	95	95	95
17	AA63281	Red Ferrosol	183	0	0	0	0	65	65	60
18	AA63293	Red Ferrosol	1	0	0	0	0	50	50	50
19	AA63294	Red Ferrosol	54	0	8	0	0	75	80	80
20	AA63295	Red Ferrosol	404	0	36	0	0	30	30	30
21	AA63465	Red Ferrosol	952	0	20	30	0	80	80	80
22	AA63466	Red Ferrosol	679	0	0	0	0	40	40	40
23	AA63467	Grey sandy loam	24	427	0	0	0	10	10	10
24	AA63468	Red Ferrosol	2	0	0	0	-	25	25	25

DAS = Days after Sowing

Figure 1.2 - Seedling emergence in bioassay tests on 24 soil sample at 19 days after sowing

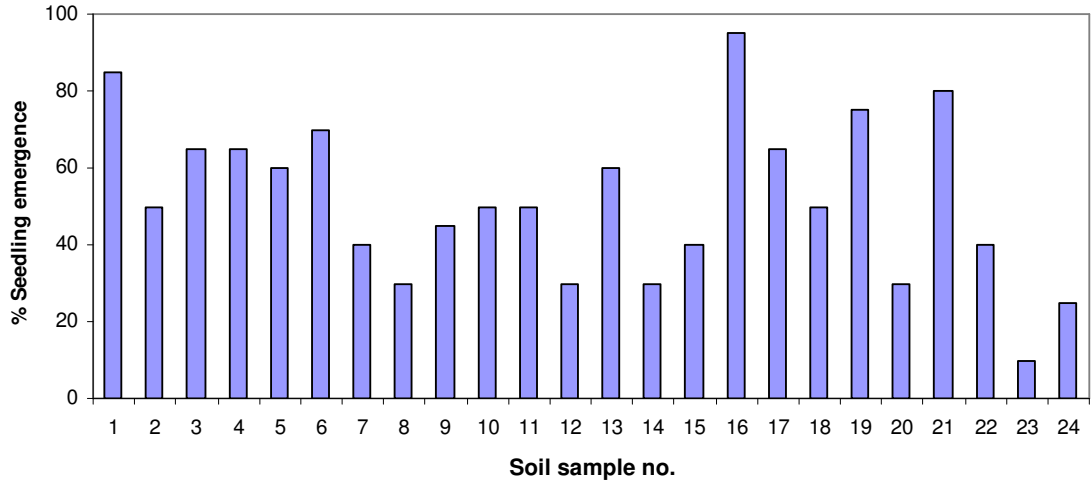


Figure 1.3 - Seedling emergence range at 19 days after sowing

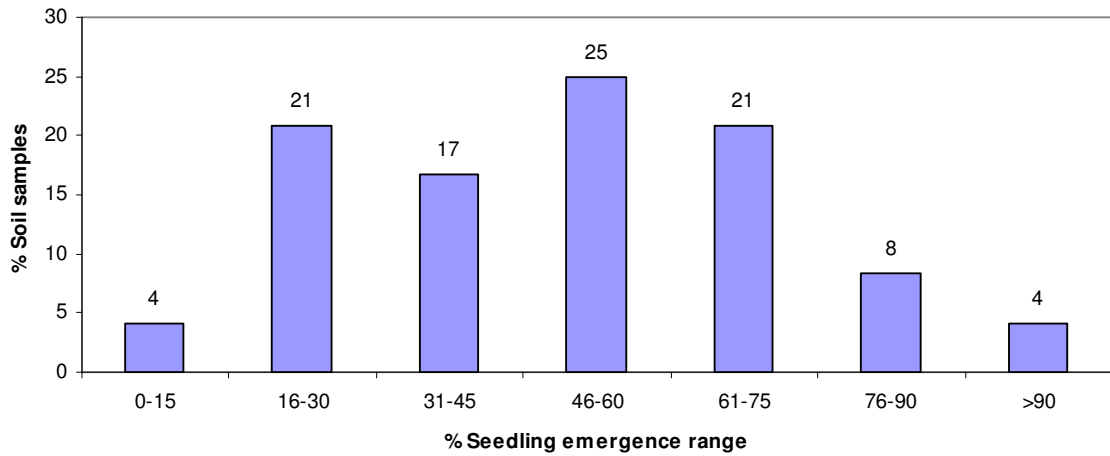


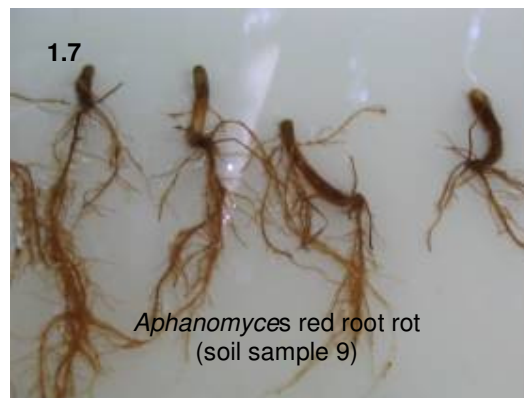
Table 1.4 – Green bean seedling total and average fresh shoot weight, root rot severity and description, and major pathogens associated with the root rots

Paddock & Soil Sample No.	Sample Code	Fresh shoot weight (g/all plants)	Average fresh shoot weight/plant	Disease severity rating (1-5)	Root rot	Major pathogens associated with root rot
1	AA63283	52.52	3.50	3.9	black rot	<i>Thielaviopsis</i>
2	AA63284	31.23	3.47	3.6	brown rot	<i>Rhizoctonia</i> , <i>Pythium</i>
3	AA63285	53.64	4.13	3.8	black rot	<i>Thielaviopsis</i>
4	AA63286	56.39	4.34	3.5	brown rot	<i>Rhizoctonia</i> , <i>Pythium</i>
5	AA63287	63.62	5.78	3.8	black rot	<i>Thielaviopsis</i> , <i>Rhizoctonia</i>
6	AA63288	49.80	3.56	3.9	brown rot	<i>Rhizoctonia</i>
7	AA63289	48.70	6.09	3.9	black rot	<i>Thielaviopsis</i>
8	AA63290	38.93	6.49	4.0	black rot	<i>Thielaviopsis</i> (main disease), <i>Aphanomyces</i>
9	AA63291	40.53	3.68	4.1	red brown rot	<i>Aphanomyces</i>
10	AA63292	21.09	1.92	3.8	black & red brown rot	<i>Thielaviopsis</i> (main disease), <i>Aphanomyces</i> , <i>Rhizoctonia</i>
11	AA63282	10.56	2.11	2.2	brown rot	<i>Rhizoctonia</i>
12	AA63276	33.53	4.79	2.0	light brown rot	<i>Aphanomyces</i>
13	AA63277	16.96	2.83	1.8	light brown rot	unknown
14	AA63278	19.72	6.57	2.0	light brown rot	<i>Rhizoctonia</i>
15	AA63279	15.86	3.17	3.6	light brown rot	<i>Rhizoctonia</i>
16	AA63280	87.84	4.62	3.4	light brown rot	<i>Rhizoctonia</i>
17	AA63281	47.10	3.93	3.3	light brown rot	<i>Rhizoctonia</i>
18	AA63293	19.66	3.93	2.2	light brown rot	<i>Rhizoctonia</i>
19	AA63294	51.72	3.23	3.2	light brown rot	<i>Rhizoctonia</i>
20	AA63295	11.22	3.74	1.3	light brown rot	<i>Rhizoctonia</i>
21	AA63465	16.74	2.09	4.0	light brown rot	<i>Thielaviopsis</i> , <i>Rhizoctonia</i>
22	AA63466	11.34	2.83	4.0	light brown rot	<i>Rhizoctonia</i>
23	AA63467	4.76	2.38	1.0	no obvious rot	no obvious pathogens, only poor emergence
24	AA63468	29.11	5.82	3.8	black rot	<i>Thielaviopsis</i> , <i>Pythium</i>

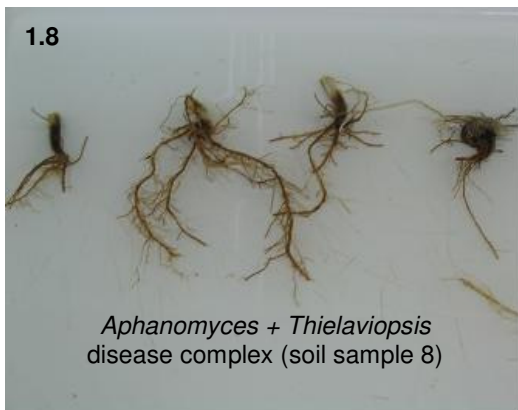
Figures of bean roots from bioassay tests



Light brown to brown rot due to *Rhizoctonia* infections on beans (Figures 1.4-1.5)



Black rot due to *Thielaviopsis* (Figures 1.6) and red brown rot due to *Aphanomyces* (Figures 1.7) on beans



Severe bean root rots due to disease complexes by *Thielaviopsis* + *Aphanomyces* (Figures 1.8) and *Thielaviopsis* + *Rhizoctonia* (Figures 1.9)

Field sampling and observations of crops in the paddocks

Table 1.4 - Field observations in 2007

Paddock No.	Location	Current crop	Field observations
1	Sassafras	green bean	Sparse to bare patches in the bean crop. Smaller plants were affected by black root rot due to <i>Thielaviopsis</i> . Roots break off easily when pulled, due to the root rot.
2	Sassafras	green bean	Crop terminated early due to very high weed pressure and shortage of water.
3	Sassafras	n/a	No crop sown, under pasture.
4	Sassafras	n/a	No crop sown, under pasture.
5	Sassafras	green bean	Very high levels of undecomposed grass residue due to dry soil conditions. Uneven plant sizes. Relatively small plants at harvest due to a lack of water. Root rot caused by <i>Thielaviopsis</i> and <i>Rhizoctonia</i> .
6	Sassafras	green bean	Poor crop establishment with large, sparse to bare patches in the bean crop compounded by poor soil conditions. Relatively small plants at harvest suffering from early crop senescence. Severe rot on lower tap roots.
7	Thirstane	green bean	Black rot more severe, with stunted plants in the area close to the fence line, where less water was applied in irrigations. Black and brown root rots caused by <i>Thielaviopsis</i> and <i>Rhizoctonia</i> .
8	East Sassafras	n/a	No crop sown, under pasture.
9	East Sassafras	green bean	Reduced plant density and poor growth. Reddish brown rot due to <i>Aphanomyces</i> noted on the seedlings, but at close to harvest, secondary <i>Penicillium</i> rot was noted on many rotten roots.
10	East Sassafras	green bean	High levels of undecomposed cereal crop residue. Uneven plants with sparse patches due to root rot complex by <i>Aphanomyces</i> and <i>Thielaviopsis</i> . Basal stem rot caused by a white sterile fungus that originated from rotting cereal crop residue.
11	Forth	n/a	No vegetable crop sown.
12	Upper Burnie	n/a	No crop sown, fallow.
13	Flowerdale	green bean	High levels of undecomposed grass residue. Relatively even crop establishment with some sporadic small and stunted plants. Stunted plants had abnormal swollen hypocotyl end with no tap root.
14	Flowerdale	green bean	High levels of undecomposed grass residue. Relatively even crop establishment. Some stunted plants had abnormal swollen hypocotyl end with no tap root. <i>Rhizoctonia</i> observed on some roots with brown rot.
15	Sisters Creek	potato	Even plant sizes in potato crop. Powdery scab was common on the potato tubers.
16	Sisters Hills	green bean	Uneven plant sizes, small stunted plants had swollen hypocotyl end with no tap root and few lateral roots. Some light brown root discoloration of healthy plants due to <i>Rhizoctonia</i> .
17	Sisters Hills	green bean	Very uneven plant sizes with many small plants. Small, stunted plants had swollen hypocotyl ends with no tap root and few lateral roots. Some light brown root discoloration of the medium size plants due to <i>Rhizoctonia</i> .
18	Sisters Hills	onion	Uneven plant size and hence uneven bulbs. Plants had relatively shallow root systems with a depth of approximately 15 cm. This may be due to drought conditions and shallow irrigation. <i>Rhizoctonia</i> was observed on the surface of some onion roots but did not cause lesion or root discoloration.
19	Sisters Hills	green bean	Good crop establishment with relatively even plant sizes; a few small plants with stunted and swollen hypocotyl ends with no tap roots. Some light brown root discoloration of medium size plants due to <i>Rhizoctonia</i> .
20	Montumana	green bean	Uneven plant sizes, many small plants with swollen hypocotyl end and no tap root. A few small plants have brown hypocotyl rot due to <i>Rhizoctonia</i> .
21	Sisters Hills	n/a	No crop sown, under pasture.
22	Boat Harbour	n/a	Crop not sown, fallow after carrots.
23	Wynyard	green bean	Uneven crop establishment, patches of poor emergence, most roots had red brown hypocotyl and root discoloration. No obvious root pathogens could be found in association with the above discoloration. Poor sandy loam soil with very low organic matter.
24	Sassafras	pasture	Under pasture. The previous bean crop sown in 2006 had severe root rot causing the crop to wilt and senesce early due to a root disease complex by <i>Thielaviopsis</i> and <i>Pythium</i> .

Figures of green bean crops and samples from the paddocks in field observations



Uneven crop establishment and severe root rot of stunted seedlings (Figures 1.10-1.11)

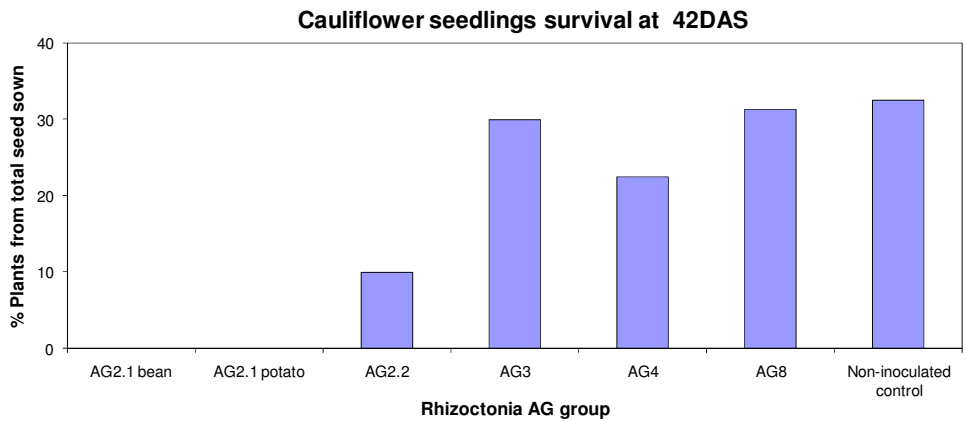
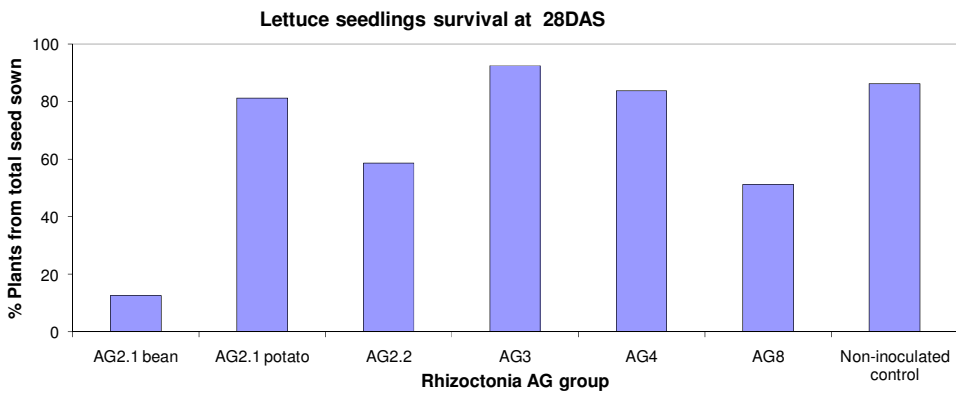
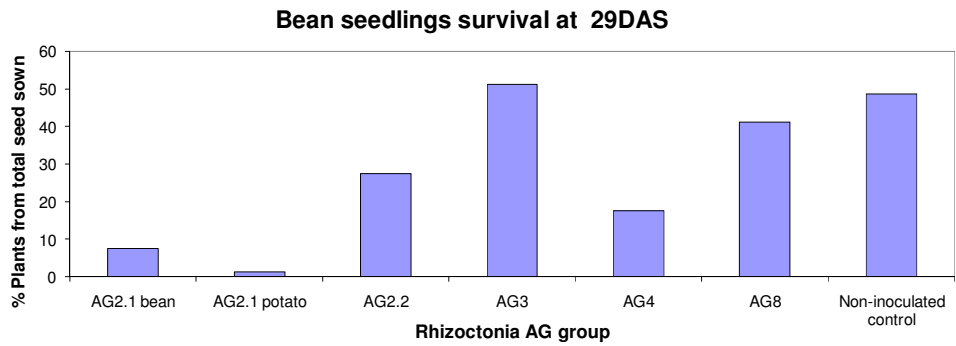
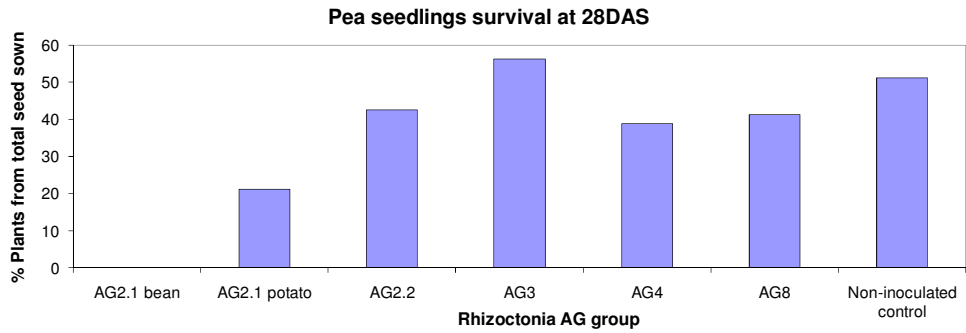


Early crop senescence due to root rot and constrictions of lower tap roots (Figures 1.12-1.13)



Typical stunted plants due to abnormal root growth observed in paddock nos. 16-22 (Figures 1.14-1.15)

Figure 1.16 Pathogenicity of *R. solani* AG isolates on green peas, green beans, lettuces and cauliflower



* DAS = days after sowing

Discussion

Soil tests for *R. solani* sub-groups

- AG2.1 was the most common *R. solani* sub-group in the soil samples from 40 paddocks, being detected in 83% of paddocks. In inoculated soil, studies have shown that *R. solani* AG2.1 infections of peas, green beans and pumpkin reduced root biomass and caused constrictions of lower stems and roots, thus affecting the overall plant growth and productivity (Pung & Cross 2007).
- The other *R. solani* sub-groups, AG3, AG4 and AG2.2 were detected in 35%, 25% and 8% of the paddocks, respectively. AG8 was not detected in any of the soil samples. The effects of these sub-groups on various vegetable crops are currently being examined in further studies, in order to gain a better understanding of their impact on vegetable crop production.
- *R. solani* AG3 is the principal cause of Rhizoctonia black scurf on potato tubers, and appears to be specific to potatoes (Carling & Leiner 1986, Campion et al 2003). Many of the paddocks surveyed were also typically used for potato production. Black scurf on potato tubers due to black sclerotia formed on the surface of tubers by *R. solani* is the most obvious symptom of Rhizoctonia disease on potato crops. Most of the other AG groups are believed to have a broad host range and the levels of damage cause by them may be dependent on field and crop conditions. In France, AG2.1 isolates did not cause black scurf on potato tubers, but at very high levels, it can cause deformities and corky lesions on tubers (Campion et al 2003).

Bioassay tests

- In the bioassay tests with green beans, soils in the pots were kept relatively wet and cool during the trial, with two irrigations per day. The bean seedling emergence and survival at 19 days after sowing (19DAS) was highly variable, ranging from 10% to 95% (Table 1.3, Figure 1.2). Figure 1.4 shows the percentage of soil samples at various levels of seedling survival. The seedling emergence and survival were more than 60% in only approximately one third of the soil samples, and less than 46% in approximately 42% of the soil samples. These figures indicate that seedling establishment from the untreated green bean seeds was very poor.
- There was no linear correlation between the populations of *R. solani* AG2.1 and the seedling survival at 43DAS ($R^2 = 0.1952$) (Table 1.3). There were also no obvious correlations between the other AG groups and seedling survival. Apart from *R. solani*, there were also other pathogens such as *Thielaviopsis*, *Aphanomyces* and *Pythium* that can impact on seed germination and seedling survival. The causes of poor seedling establishment and growth can be complex and each paddock, location or farm may have its own unique sets of contributing factors.
- The examination of roots from the bioassay tests at 43DAS indicated that *R. solani* was common in most of the soil samples. It was found in association with bean root rot in 63% of the soil samples. In 42% of the soil samples, only the *R. solani* pathogen was observed on the root rots. Root rot caused by Rhizoctonia was generally light brown to brown in colour (Figures 1.4-1.5). There were also other root pathogens, particularly *Thielaviopsis basicola* and *Aphanomyces euteiches*, which caused severe black root rot and red brown root rot (Figures 1.6-1.7). Roots with more than one pathogen typically had worse root rot compared to those affected only by a single pathogen (Figures 1.8-1.9).
- It is noteworthy that the two devastating bean root pathogens, *Thielaviopsis* and *Aphanomyces*, were frequently found in the Sassafra and Thirlstane areas in paddock nos. 1-10 and 24, which have been more intensively used for green bean production, compared to those located west of Wynyard in paddock nos. 12-23. Further studies are currently being conducted in a new project VG08043 on these pathogens in green beans.

- The worst seedling emergence and survival, with 10% seedlings, was recorded in sample no. 23, taken from a sandy loam soil from Wynyard. Although this soil had the highest level of *R. solani* AG2.2, no *Rhizoctonia* hyphal growth or other root pathogens could be found on hypocotyls and roots of the surviving seedlings. This is surprising, as *R. solani* AG2.2 has been described as a serious pathogen to green beans, causing lesions on hypocotyls and roots (Hagedorn & Hanson 2005). It is possible that this pathogen only caused poor seedling emergence in this soil type. Other unknown non-pathological causes related to the type of soil in the paddock are also believed to be contributing factors to the poor seedling emergence, as uneven crop establishment is commonly found in other paddocks with the same soil type. The second worst seedling emergence and survival, at 25%, was recorded in sample no. 24, a soil from Sassafras. There was little or no *R. solani* detected in the soil test for this sample, and no *R. solani* growth could be found in association to root rots. Instead, *T. basicola* and *Pythium*, observed in root rot tissues, were the major pathogens.
- The outcomes of the bioassay tests indicate that root rot could be caused by a range of root pathogens, which may also interact with one another to cause a root disease complex, which can result in a more severe root rot than that cause by a single pathogen. Soil tests developed to detect soilborne pathogens may have to be crop specific and be able to cover the range of important and damaging pathogens for that crop, in order to be useful for commercial use.
- The impact of the root pathogens and root rot severity on seedling growth could not be properly measured in the bioassay study, because plant growth was affected by plant competition in the limited space available in each pot (Table 1.4).

Field observations

- Most of the paddocks were selected because they had been used for intensive vegetable and potato production, and yield decline had been experienced on these soils. Most of the green bean crops in the paddocks appeared to have better seedling establishment compared to the bioassay tests. The differences in emergence between the bioassay tests and the field crops may be due to the relatively warm and dry field soil conditions in Tasmania in 2006/07. Furthermore, all of the bean seeds used in the bioassay study were untreated, whereas seeds in commercial crops had been treated with commercial fungicide seed treatments (Apron and Thiram, Maxim, Dynasty or Captan). Crop establishment in the paddocks would have been substantially enhanced by the fungicide seed treatments.
- Fungal pathogens found in association with root rots and discolouration of bean plants in the paddocks were consistent with those observed in bean seedling roots in the bioassay tests (Table 1.4). Black and red brown root rot due to *Thielaviopsis* and *Aphanomyces* were common in the Sassafras area, while light brown root rot caused by *Rhizoctonia* was common in the Sisters Creek area.
- In addition to root rot, sparse plant densities and stunted seedlings with swollen and tapered roots were consistently observed in paddock nos. 16-22 in areas west of Wynyard (Figures 1.14-1.15). No pathogens or crop management practices and field factors could be identified in association with the symptoms. The symptoms were later traced to abnormalities associated with the seed batch used. The same root symptoms were observed on seedlings grown from the same seed batch sown in pasteurised soil in pots.

Pathogenicity of *R. solani* AG isolates

- AG2.1 isolates were shown to be the most pathogenic to peas, green beans and cauliflowers, causing severe damping off on seeds and seedlings. Although not as pathogenic as AG2.1, AG2.2 and AG4 were shown to cause some damping off on pea, green bean and cauliflower. AG3 and AG8 had little or no effects on them.
- With lettuce, only the AG2.1 bean isolate was highly pathogenic, but not the AG2.1 potato isolate. This indicates that there is variability in the pathogenicity of isolates from the same sub-group. AG2.2 and AG8 also reduced seedling lettuce seedling survival.
- AG3 had no effects on all the vegetables.

Conclusions

- In soil tests, the most common type of *R. solani* sub-group was AG2.1, which was detected in 83% of soils collected from 40 paddocks that had been intensively used for vegetable and potato production.
- The bioassay tests indicated that root rot could be caused by a range of root pathogens, which may also interact with one another to cause a root disease complex, and which often resulted in a more severe root rot than that caused by a single pathogen. Soil tests developed to detect soilborne pathogens may have to be crop specific and be able to cover the range of important and damaging pathogens for that crop, in order to be useful for commercial use.
- In the bioassay tests, *Rhizoctonia* was found in association with bean root rot in 63% of the soil samples. In 42% of the soil samples, only the *Rhizoctonia* pathogen was observed on the root rots. However, bean root damage caused by other pathogens, such as *Thielaviopsis* and *Aphanomyces*, was more severe than that caused by *Rhizoctonia*.
- In the bioassay tests, under relatively cold and wet conditions, seedling establishment from the untreated green bean seeds was very poor. The seedling emergence and survival was less than 46% in approximately 42% of the soil samples.
- In the field observations, poor crop establishment and growth was associated to root rots, root pathogens, more intensive use of paddocks for green bean production and poor soil conditions, as well as poor seed quality.
- *R. solani* AG2.1 that was isolated from stem rot on green beans was shown to be the most pathogenic isolate on seedlings grown from untreated seeds of pea, bean, cauliflower and lettuce.

References

Anderson, N. A. , 1982. The genetics and pathology of *Rhizoctonia solani*. Ann. Rev. Phytopathol. 20: 329-47.

Campion, C., Chatot, C., Perraton, B. & Andrivon, D., 2003. Anastomosis groups, pathogenicity and sensitivity to fungicides of *Rhizoctonia solani* isolates collected on potato crops in France. European J. Pl. Pathol. 109 (9): 983-992.

Carling, D. E., & Leiner, R. H. 1986. Isolation and characterization of *Rhizoctonia solani* and binucleate *R. solani* – like fungi from aerial stems and subterranean organs of potato plants. Phytopathol. 76: 725-729.

Ceresini, P., 1999. *Rhizoctonia solani*. www.cals.ncsu.edu/course/pp728/Rhizoctonia/Rhizoctonia.html.

Hagedorn, D.J. & Hanson, L.E. , 2005. Rhizoctonia root rot. In “Compendium of bean diseases”, pp19-20, 2nd edition. The American Phytopathological Society, St. Paul, Minnesota, U.S.A.

Ogoshi, A. , 1987. Ecology and pathogenicity of anastomosis and intraspecific groups of *Rhizoctonia solani* Kuhn. Ann. Rev. Phytopathol. 25: 125-43.

Pung, H. & Cross, S. Evaluation of new seed dressings for improved disease and insect control in vegetable crops. Project VG04021. Final Report 2007.

2. Screening of non-chemical soil treatments for suppressing *Rhizoctonia* in pot trials

Summary

Three pot trials were conducted in Devonport, Tasmania, to evaluate gypsum, sawdust and molasses and biocontrol agents, applied as pre-plant soil treatments for the control of *R. solani* AG2.1 on green beans and garden peas in soil inoculated with *R. solani* AG2.1. The pre-plant soil treatments were mixed into soil and maintained at field capacity for one month prior to sowing. *R. solani* AG2.1 caused severe damping off and drastically reduced seedling emergence and survival of green beans and peas. Sawdust mixed into soil was effective in controlling *Rhizoctonia* damping off, especially at the high rate of 3% w/v. Molasses applied at 1% to 2% also showed potential for *Rhizoctonia* control. Gypsum mixed into soil showed weak control. Gypsum applied onto the soil surface and then drenched into the soil had no effect. The biocontrol agents based on *Bacillus lydicus* and *Trichoderma* spp. had little or no effect on *Rhizoctonia*.

Introduction

Gypsum and calcium sulphate granules have been shown to suppress the mycelial growth of *Rhizoctonia* from four week old cultures in *in vitro* tests. Organic amendments such as molasses, sawdust and green manure are believed to help suppress *Rhizoctonia* pathogens. Therefore, two pot trials were conducted to screen these materials for their ability to suppress *R. solani* AG2.1. *R. solani* AG2.1 is the most common *Rhizoctonia* found in paddocks that have been intensively cropped with vegetables.

Materials & methods

Three studies were conducted in sandy loam soil that was inoculated with *R. solani* AG2.1 in 3 L pots (19.5 cm x 18.5 cm). Soil in all pots were inoculated with *R. solani* AG2.1 bean isolate by mixing *Rhizoctonia* colonised millet seeds into the top 2 L of soil at a rate of 20 g/pot (or 1.0% w/v) in pot trials 1 and 3, and at a rate of 15 g/pot (or 0.75% w/v) in pot trial 2. All the soil treatments were applied at the same time as *Rhizoctonia* inoculum as described in the treatment details below. Following *Rhizoctonia* inoculations and soil treatments, soil in the pots were wet to field capacity and kept under relatively cool conditions (10-15°C) for approximately 4 weeks prior to sowing. Twenty seeds were then sown in each pot: trials 1 and 3 were sown with untreated green bean seeds, and trial 2 was sown with untreated garden pea seeds. Garden peas were used in trial 2 because the trial was conducted in winter, when green beans cannot be grown due to the cold conditions. The trial design was randomised complete block with four replicates. Pots were irrigated with an overhead sprinkler every day.

Seedling emergence and seedling survival were assessed by recording the number of seedlings in each pot and they were tabulated as a percentage of the 20 sown bean, or garden pea seeds. In the final assessment, all shoots of surviving plants were cut and weighed as a measure of plant size. The roots of the surviving plants were also washed and rated for root rot severity rating of 0 to 5. Root rot severity rating: 0 = no root rot; 1 = no hypocotyl rot, slight root discoloration; 2 = < 10 % hypocotyl rot, some root discoloration; 3 = 11-30% hypocotyl rot, root discoloration; 4 = 31-60% hypocotyl rot, root discoloration and 5 = >60% hypocotyl rot, root discoloration.

Treatment details**Table 2.1. Treatment details in Trial 1 on green beans**

Pre-plant treatment	Product rate (w/v)	Application method
Untreated control	-	With pathogen and water only applied
Gypsum (Micro-Gyp) drench 1%	1%	20 g gypsum broadcast onto soil surface, and then drenched into top soil with water
Gypsum (Hi-Ag) mixed 1%	1%	Gypsum mixed thoroughly into top 2 L soil at 20, 40 and 60 g for 1%, 2% and 3%, respectively
Gypsum (Hi-Ag) mixed 2%	2%	
Gypsum (Hi-Ag) mixed 3%	3%	
Molasses mixed 1%	1%	Thick viscous molasses diluted with warm water and then applied at the appropriate rate and mixed thoroughly into the top 2 L soil
Molasses mixed 1.5%	1.5%	
Sawdust mixed 1%	1%	20 and 60 g sawdust mixed thoroughly into top 2 L soil
Sawdust mixed 3%	3%	

Gypsum used was calcium sulphate Hi-Ag™, produced by Processed Gypsum Products, which consists of fine gypsum granules that have been passed through a 2 mm sieve (91% purity, 21.2% Ca, 16.8% S). Sawdust was from Eucalyptus hardwood obtained from a local plant nursery.

Table 2.2. Treatment details in Trial 2 on garden peas

Pre-plant treatment	Product rate (w/v)	Application method
Non-inoculated control	n/a	No pathogen, water only applied
Untreated control	n/a	With pathogen and water only applied
Agm Trichoderma	0.01%	Prepare suspensions at the listed product rates in 1 L water and then apply and mix 100 ml of the suspension into the top 2 L soil
Micro Plus	0.01%	
SoilGard	0.25%	
Molasses 10%	1.0%	Diluted in 100 ml water and then mix into the top 2 L soil
Molasses 20%	2.0%	
Gypsum (Hi-Ag) 2.5%	2.5%	Mix 50 g for 2.5% or 100 g for 5.0% into the top 2 L soil
Gypsum (Hi-Ag) 5%	5.0%	
Gypsum (Micro-Gyp) 2.5%	2.5%	

The gypsum used was Hi-Ag™ and Micro-Gyp™, both were calcium sulphate produced by Processed Gypsum Products. Hi-Ag was fine gypsum granules that have been passed through a 2 mm sieve and Micro-Gyp™ was a wettable powder of gypsum. SoilGard™ was based on *Trichoderma* spp. produced Certis USA

Table 2.3. Trial details for Trial 3 on green beans

Pre-plant treatment	Product rate (w/v)	Application method
Non-inoculated control		No pathogen, water only applied
Untreated control		With pathogen and water only applied
Contans	0.50%	Prepare suspensions at the listed product rates in 1 L water and then apply and mix 100 ml of the suspension into the top 2 L soil
Micro-Plus	0.50%	
0.5% Molasses	0.50%	
1.0% Molasses	1.00%	

Contans was based on *Coniothyrium minitans*, a fungal biocontrol agent, produced by Prophya.

Results

Table 2.4. Effects of pre-plant soil treatments in *Rhizoctonia* inoculated soil in Trial 1 on beans

Pre-plant soil treatment	Seedling emergence/survival % of seed sown			Root rot severity index (0-5) 46DAS	Total fresh shoot weight of surviving plants/pot 46DAS
	12DAS	20DAS	46DAS		
Untreated control	5 e	8 d	6 c	1.5	31.0 bcd
Gypsum drench 1%	3 e	5 d	3 c	2.5	21.0 d
Gypsum granules 1%	5 e	9 d	6 c	2.1	28.7 cd
Gypsum granules 2%	14 cde	18 cd	16 bc	1.6	53.7 bcd
Gypsum granules 3%	10 de	14 cd	14 bc	2.6	52.3 bcd
Molasses 1%	28 bc	30 bc	31 b	1.8	66.0 ab
Molasses 1.5%	29 b	34 b	30 b	2.6	56.8 abc
Sawdust 1%	31 bcd	35 bc	34 b	2.0	64.5 ab
Sawdust 3%	68a	76a	74a	2.3	82.5 a

Means followed by same letter do not significantly differ ($P = 0.05$)
DAS = Days after sowing

Figure 2.1. Pre-plant treatment effects on seedling survival in Trial 1

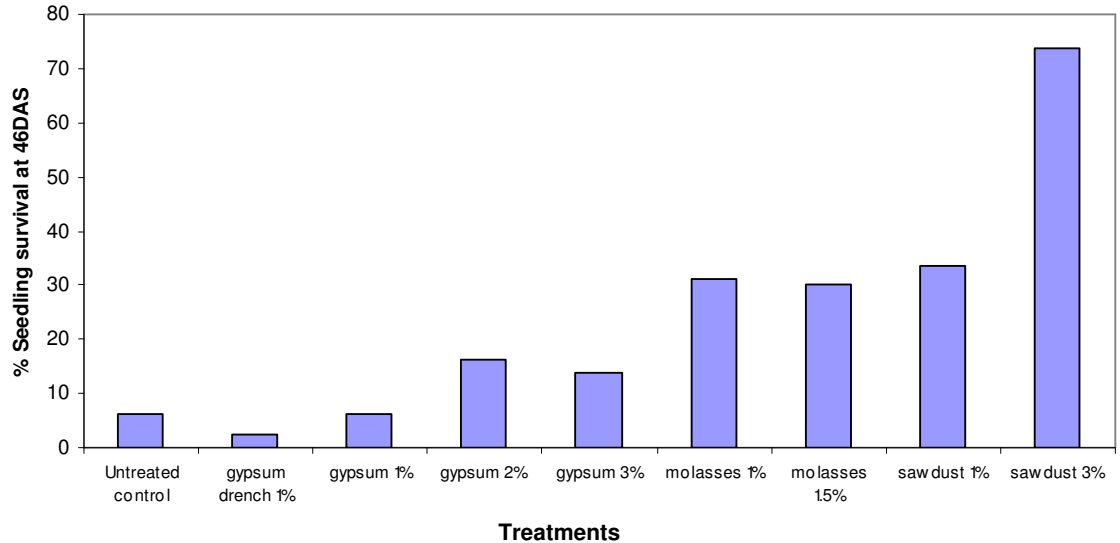


Table 2.5. Effects of pre-plant soil treatments in *Rhizoctonia* inoculated soil in Trial 2 on peas

Treatment	Emergence/survival (% plants from total seed sown)		Total fresh shoot weight of surviving plants/pot
	14DAS	27DAS	28DAS
Untreated control	10 cd	10 cd	0.96 cde
Micro Plus	4 d	1 e	0.41 de
SoilGard	3 d	1 e	0.07 e
Gypsum (Hi-Ag) 5.0%	5 d	5 de	0.84 cde
Gypsum (Hi-Ag) 2.5%	14 cd	14 c	1.94 cd
Gypsum (Micro-Gyp) 2.5%	13 cd	16 c	1.46 cde
Agm Trichoderma	20 c	21 c	2.65 c
Molasses 1%	76 b	75 ab	20.48 ab
Molasses 2%	89 a	90 a	24.01 a
Non-inoculated control	69 b	65 b	15.22 b

Means followed by same letter do not significantly differ (P = 0.05)
 DAS = Days after sowing

Figure 2.2. Pre-plant treatment effects on seedling survival in Trial 2

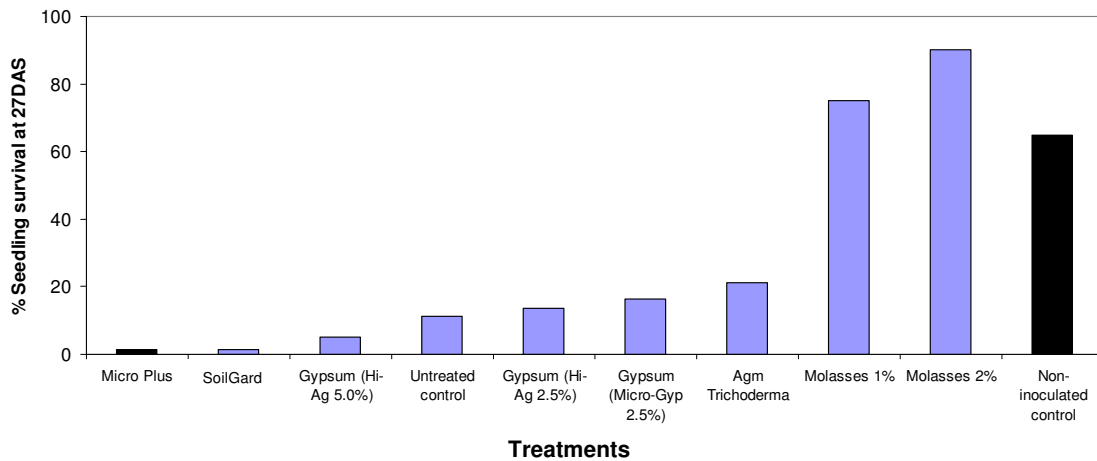
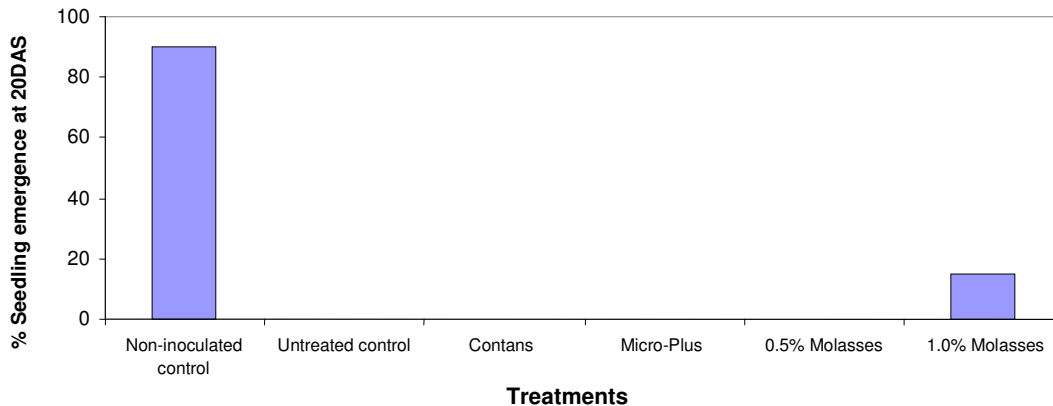


Figure 2.3. Pre-plant treatment effects on seedling survival in Trial 3



Discussion

- *R. solani* AG2.1 was highly pathogenic to both peas and green beans. Its major effect was in reducing seedling emergence and survival. Only 10% or less of the seed sown emerged and survived as seedlings in the inoculated and untreated controls (Figures 2.1 - 2.3).
- In Trial 1, pre-plant applications of sawdust and molasses into *Rhizoctonia* inoculated soils suppressed the pathogen and significantly increased seedling emergence and survival compared to the untreated control (Table 2.1, Figure 2.1). Sawdust applied at 3% w/v had the highest rate of seedling survival, with 74% survival at 46DAS. Sawdust applied at 1% w/v and molasses at 1% and 1.5% w/v also increased seedling survival at the range of 30% to 34%. In comparing the two materials, molasses is considered to have greater potential for commercial use as it is readily available, relatively low cost and is not bulky. Hence, further studies were conducted with it in Trial 2.
- In Trial 1, gypsum granules and the gypsum drench applications did not significantly improve seedling emergence compared to the untreated inoculated control (Table 2.1, Figure 2.1). However, there was a trend of increases in the percentage of seedling emergence and survival with the gypsum granule applications at the higher rates of 2% and 3%. In laboratory studies, gypsum was shown to suppress *R. solani* AG2.1 by delaying its mycelial growth. Unlike fungicide chemicals, it does not completely inhibit growth. *Rhizoctonia* level and growth into soil may have been too high for gypsum to have any impact.
- In Trial 2, molasses again showed potential in suppressing the pathogen and substantially increased pea seedling survival. Gypsum and Agm *Trichoderma* also showed some potential, but they were not as effective as molasses.
- The biocontrol agents, Micro Plus based on *Bacillus lydicus*, SoilGard based on *Trichoderma* spp. had no effect on *Rhizoctonia*.
- In Trial 3, the pathogen was highly pathogenic because of very active fungal growth from the colonised millet seed inoculum, and little or no seedlings emerged and survived in the inoculated soil, regardless of the pre-plant soil treatments. Only 15% plants survived in the soil treated with 1.0% molasses.

3. A preliminary field study to evaluate gypsum and fungicide applications within a commercial green bean crop

Summary

A field trial was conducted in a paddock that was known to have high levels of *Rhizoctonia* in Thirlstane, Tasmania in 2006/07, where fungicide and gypsum soil treatments were applied to the soil surface and raked and irrigated in, to determine their effects on root diseases on green beans within a commercial crop. The paddock was selected because a relatively high level of *Rhizoctonia* AG4 was detected in soil tests. After sowing, surface soil treatments were applied. Gypsum granules were broadcast onto small plots at 200 kg/ha and 1000 kg/ha, and then raked in to approximately 50 mm deep. The fungicides, Amistar 250 SC at 2 L/ha, Filan 500 WG at 1 kg/ha, Rizolex 500 SC at 1 L/ha and Thiram 800 WP at 1 kg/ha were first applied onto gypsum granules as a carrier, which were then broadcast and raked into the top soil. The entire crop in the paddock was kept relatively dry due to the dry weather conditions as well as by the grower in order to reduce the impact of root disease in the crop. As a consequence, the root rot disease was not severe and appeared to have no detrimental effects on crop establishment and growth. No significant differences could be found in the root rot severity rating and black root rot incidence between the soil treatments.

Aims

Gypsum granules and fungicides coated onto gypsum granules were applied onto the soil surface in order to evaluate their potential for damping-off and root rot control within a commercial green bean crop.

Materials & methods

The trial was set up in a paddock that was known to have high levels of *Rhizoctonia* (see soil sample 7 in Table 1.1). The *R. solani* sub-groups, AG2.1 and AG4 were detected in a DNA soil test. AG3 also detected at the site was a host specific pathogen, pathogenic to potato crops only. After green bean seeds had been sown and the soil surface compacted with a heavy roller, the soil treatments were applied. The trial design was randomised complete block with 2 m by 5 m plot size and four replicates. The fungicides were first applied to gypsum granules as carriers. The treated gypsum was then broadcast at a rate of 200 kg/ha and incorporated to a depth of approximately 50 mm with a rake. The trial area was irrigated soon after the treatment applications.

Table 3.1 – Fungicide and gypsum treatment details

No.	Treatment	Product rate (kg, or L /ha)	Active ingredient (g ai/ha)	Application method
1	Untreated control (raked)	N/a	N/a	Soil raked
2	Amistar 250 SC	2 L/ha	500 g	Soil raked soon after broadcasting of fungicide treated gypsum
3	Filan 500 WG	1 kg/ha	500 g	
4	Rizolex 500 SC	1 L/ha	500 g	
5	Thiram 800 WP	1 kg/ha	800 g	
6	Gypsum (low rate)	200 kg/ha	-	Soil raked soon after broadcasting of gypsum granules
7	Gypsum (high rate)	1000 kg/ha	-	

Assessments for seedling emergence and survival were conducted at 28 days after sowing (28DAS) by recording the number of seedlings in 2 plant rows x 3 m in each 10 m² plot. The numbers of small and stunted seedlings were also recorded. At 74DAS, 20 consecutive plants in the middle row of each plot were collected and assessed for fresh shoot weight and root rot severity. Root rot severity was rated as described in Section 2. The incidence of black root rot due to *Thielaviopsis* was also assessed.

Results

Table 3.2 - Treatment effects on plant density, root rot rating and percentage roots with black rot

No.	Treatment	28DAS	74DAS	
		Plant density/plot	Root rot severity rating (0-5)	% Roots with black rot by <i>Thielaviopsis</i>
1	Untreated control	112	3.4	65
2	Amistar 250 SC	122	3.1	45
3	Filan 500 WG	105	3.4	31
4	Rizolex 500 SC	104	3.3	54
5	Thiram 800 WP	113	3.2	46
6	Gypsum (low rate)	119	3.5	48
7	Gypsum (high rate)	110	3.4	66
p-value		0.173	0.426	0.113

DAS = Days after sowing

Discussion

The paddock was selected for a *Rhizoctonia* study because DNA soil tests indicated this paddock had a relatively high level of AG4 and a low level of AG2.1, and both were found to be pathogenic to green beans (Figure 1.16). Crop establishment and growth within the trial area was excellent. Although all plant roots had brown or black root discoloration, the severity was considered to be moderate and it appeared to have little or no obvious impact on plant growth.

The main type of root disease noted in this paddock was black root rot caused by the pathogen *Thielaviopsis basicola*. There was also a brown root discoloration, which may be due to a complex of root pathogens including *Rhizoctonia* and *Fusarium*. This study demonstrated that in intensively cropped paddocks, more than one soilborne pathogen is often involved in root diseases.

The gypsum and fungicide soil treatments had no significant effect on plant densities, root rot severity and black root rot incidence. The root rot was not severe and appeared to have no detrimental effect on crop establishment and growth. Weather conditions were relatively dry and warm, and the bean crop was kept relatively dry during the growing season by the grower in order to minimise the impact of root rot disease in the entire commercial crop in the paddock. This appeared to have limited any damage by the root diseases. Black root rot and *Rhizoctonia* infections tend to be more severe under wet and cold conditions. No significant differences could be found in the root rot severity rating and black root rot incidence.

4. A field trial to evaluate pre-plant non-chemical soil treatments in *Rhizoctonia* inoculated soil

Summary

A field trial was conducted at Forthside Vegetable Research Station, Tasmania, in 2007/08, where molasses and two biocontrol products based on *Bacillus lydicus* and *Trichoderma virens* were applied as spray applications to the seed furrow at sowing for *Rhizoctonia* control in inoculated soil. Apart from a small difference in plant densities between the inoculated and non-inoculated soil, there were no obvious differences in the root rot severity rating and plant growth.

Aims

Molasses and two biocontrol products, Micro Plus based on *B. lydicus* and SoilGard based on *T. virens* were applied as spray applications to the seed furrow at sowing for *Rhizoctonia* control in inoculated soil. These were compared against two untreated controls – an inoculated control and a non-inoculated control, with *Rhizoctonia*.

Materials & methods

Two furrows were prepared 200 mm deep in each soil bed (1.6 m x 20 m long). *R. solani* AG2.1 inoculum was spread along the furrow at the rate of 10 g of colonised millet seed per metre row. No inoculum was applied in the non-inoculated control plots. Plot size was 1.6 metre by 4 metre. The trial design was a randomised complete block with four replicates. Spray treatment applications were applied with a water volume of 280 L/ha to the seed furrow after the fungal inoculum application. The furrows were then covered, irrigated once a week and then green bean seeds were sown four weeks later at a depth of 150 mm deep along the furrows. The trial area was irrigated with overhead sprinklers twice a week.

Results & discussion

Table 4.1 - Treatment effects on plant density, root rot rating and fresh shoot weight

Treatment	Rate	Plant density (2 x 3 m row)		Root rot index	Fresh shoot weight
		12DAS	60DAS	(0-5) 60DAS	(kg/m bed) 60DAS
Inoculated control	N/a	91 bc	87	2.4	0.73
Molasses	28 L	89 bc	89	2.3	0.66
Micro Plus™	4 kg	95 b	99	2.2	0.73
SoilGard™	4 kg	98 b	92	2.2	0.69
Non-inoculated control	N/a	111 a	110	2.3	0.88
p-value		0.001	0.137	0.244	0.214

Means followed by same letter do not significantly differ (P=0.05, LSD)

DAS = Days after sowing

The highest plant density was recorded in the non-inoculated control, where no *Rhizoctonia* inoculum was applied into the seed furrows. The trial was carried out during summer time, when soil was relatively hot and dry following the inoculum and pre-plant treatment applications. Bean seeds were sown one month after the inoculum and soil treatments. Low levels of root disease indicated that the pathogen probably did not survive well under the hot and dry conditions. Apart from the difference in plant densities between the inoculated and non-inoculated soil, there were no obvious differences in the root rot severity rating and plant growth.

5. Screening of fungicides for *Rhizoctonia* control in pot trials

Summary

Two pot trials were conducted in 2006 and 2007 to screen fungicides for their efficacies in controlling *R. solani* AG2.1. One trial was sown with green beans and the other with garden peas. Thirteen fungicide active ingredients were screened for their efficacies against *Rhizoctonia*. The fungicides were applied either as a soil drench after sowing or mixed into soil as treated granules. There were two sets of untreated controls, one inoculated with the pathogen and the other was non-inoculated. *R. solani* AG2.1 was highly pathogenic to peas and green beans, causing severe damping off in the untreated and inoculated control. *R. solani* AG2.1 is the most common *Rhizoctonia* found in paddocks that have been intensively cropped with vegetables. The main effects of *R. solani* AG2.1 was the reduction in seedling emergence and survival under wet and cold conditions. The fungicides, Amistar, Agri-Fos, Filan, Tilt, Switch and Rizolex showed potential for controlling *Rhizoctonia* and increasing seedling survival and total fresh shoot weights. Seed treatments with Maxim XL, alone or in combination with the biocontrol agent Micro Plus, were not effective under the high disease pressure.

Introduction

Two pot trials were conducted to screen and identify fungicides that have activities in controlling *R. solani*. Fungicides were applied either as seed treatment, soil drench application or mixed into soil as treated clay or polymer granules. Treated fertilisers, bentonite clay particles, polymer granules had been used successfully as slow release chemical granules in order to extend root protection from white root rot control for up to 100 days after sowing on onions.

Materials & methods

Various fungicides were screened for their potential to control *Rhizoctonia* as seed treatments or as pre-plant or post plant soil applications in two pot trial studies in 2006 and 2007. The pot trials were conducted in sandy loam soil in 3 L pots. All pots, except the non-inoculated control, were inoculated with *R. solani* AG2.1 by mixing *Rhizoctonia* colonised millet seeds into the top 2 L of soil in 3 L pots at a rate of 2.5% w/v and 0.75% w/v in Trial 1 and Trial 2, respectively. After inoculation with *Rhizoctonia*, the soil was wet to field capacity and each pot was sown with 20 untreated seeds at 20 mm deep. Trial 1 was sown with green beans and Trial 2 was sown with garden peas. The trials were set up consecutively with two crop varieties according to the weather conditions: the green beans were sown under relatively warm conditions in November 2006 to January 2007 and the garden peas were sown under relatively cold conditions in August to October 2007. Green beans are highly susceptible to cold and frost damage in April to October in Tasmania.

In Trial 1, all chemical treatments were applied as drench applications with 100 ml water per pot soon after seed sowing. In Trial 2, treatments were applied either as seed treatments, soil drench applications or mixed into soil as treated granules. The treated granules consisted of bentonite clay granules or encapsulated as slow release polymer granules, which were mixed to the desired quantity into the top 2 L soil. The pot trial was kept indoors for the first 2 week period under relatively cool conditions at 10-15°C, before being re-located to an outdoor compound, in order to slow down seed germination and seedling emergence, and optimise impact of the *Rhizoctonia* pathogen. Seedling emergence and survival, root rot severity and the fresh shoot weights of plants were recorded.

Table 5.1 - Treatment details for Trial 1 on green beans

Product	Active ingredient	Soil treatment rate		Application schedule
		Product/pot	Active ingredient (g/pot)	
Inoculated control	N/a	nil	nil	Drench soil in each pot with 200 mL water only.
Amistar SC	azoxystrobin	0.2 mL	0.05	Prepared diluted suspensions at the appropriate concentrations and bulked to a water volume of 200 mL, which was drenched onto soil in each pot after sowing.
Cabrio	pyraclostrobin	0.1 mL	0.025	
Switch	fludioxonil + cyprodinil	0.1 g	0.063	
Rizolex	tolclofos-methyl	0.1 mL	0.05	
LEM 17	LEM 17	0.4 mL	0.08	
Filan	boscalid	0.1 g	0.05 g	
Thiram	thiram	0.1 g	0.08	
Tilt	propiconazole	0.1 mL	0.025	
Agri-Fos 600	phosphorous acid	0.3 mL	0.18	
Non-inoculated control	N/a	nil	nil	

Table 5.2 - Treatment details for Trial 2 on garden peas

Treatment	Active ingredient	Soil treatment rate		Application method
		Product/pot	Active ingredient (g/pot)	
Inoculated control	N/a	nil	nil	None
Maxim XL treated seed	fludioxonil + metalaxyl	nil	nil	Seed treatment @ 50 ml/100 kg seed
Maxim XL + Micro Plus treated seed	fludioxonil + metalaxyl + <i>Bacillus lydicus</i>	nil	nil	Seed treatment @ 50 ml/100 kg + 220 g/100 kg
Amistar	azoxystrobin	0.2 mL	0.05 g	Post plant soil treatment: prepared and drenched 100 mL of the diluted suspension onto soil surface in each pot immediately after untreated seed had been sown.
Agri-Fos 600	phosphorous acid	0.3 mL	0.18 g	
Filan	boscalid	0.1 g	0.05 g	
Rizolex	tolclofos-methyl	0.1 mL	0.05 g	
Terraclor	terraclor	0.075 g	0.056 g	
Tebuconazole bentonite	tebuconazole	10 g	0.215 g	Pre-plant soil treatment: treated granules mixed into top 2 L soil, one day before untreated seed were sown.
Bayfidan bentonite	triademinol	10 g	0.188 g	
Tebuconazole in polymer	tebuconazole	10 g	0.215 g	
Non-inoculated control	nil	nil	nil	None

Results

Table 5.3 - Treatment effects on *Rhizoctonia* AG2.1 on beans in Trial 1

Pre-plant soil treatment *	Seedling emergence/survival (% plants from total seed sown)			Root rot severity rating (0-5)	Total fresh shoot weight of surviving plants (g)
	12DAS	20DAS	55DAS		
Inoculated control	13 d	20 c	16 f	1.5 c	68.0 e
Cabrio	21 bcd	25 c	20 ef	1.5 c	100.5 bcde
Thiram	21 bcd	33 bc	23 def	1.8 bc	97.5 cde
Amistar SC	20 cd	39 abc	29 cdef	2.3 abc	82.5 de
LEM 17	29 abc	40 abc	36 bcdef	2.1 abc	143.0 ab
Agri-Fos 600	33 abc	44 abc	40 abcde	2.4 ab	128.0 abc
Filan	38 ab	44 abc	41 abcde	2.5 ab	137.5 abc
Tilt	40 ab	53 ab	43 abcd	2.7 a	148.5 a
Switch	39 abc	50 ab	48 abc	2.0 abc	124.0 abcd
Rizolex	41 ab	56 ab	54 ab	2.7 a	147.0 a
Non-inoculated control	55 a	60 a	60 a	2.6 ab	141.0 abc

* Treatments were sorted in an ascending order according to the seedling survival at 55DAS
 Means followed by same letter do not significantly differ (P=0.05, LSD)
 DAS = Days after sowing

Figure 5.1 - Bean seedling survival at 55DAS

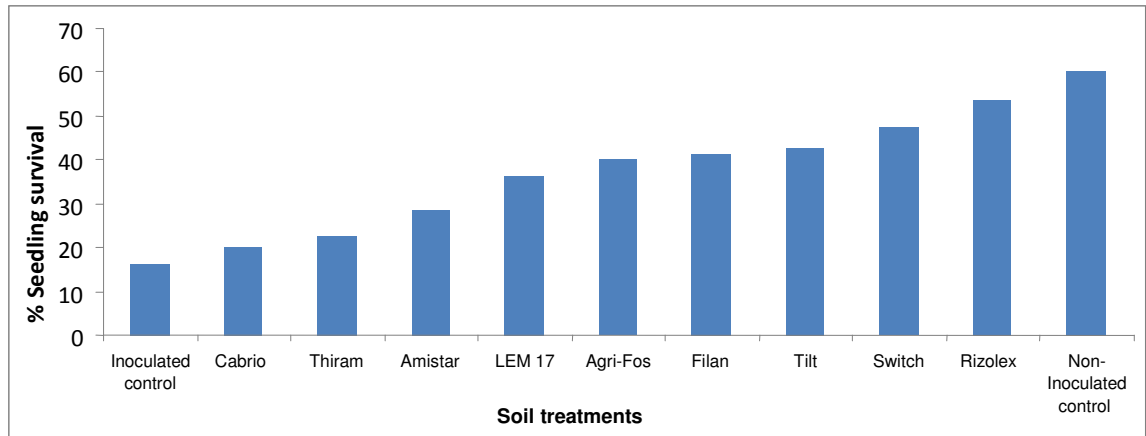
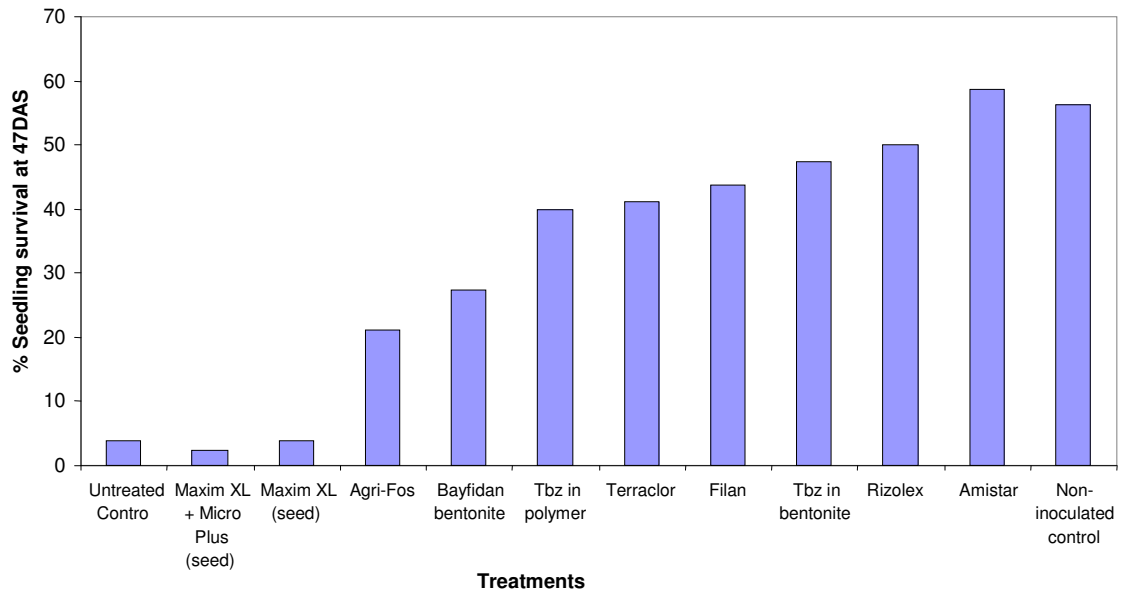


Table 5.4 – Treatment effects on *Rhizoctonia* AG2.1 on peas in Trial 2

Treatment	Emergence/survival % plants from of seed sown			Total fresh shoot weight of surviving plants/pot
	14DAS	28DAS	47DAS	47DAS
Inoculated control	4 f	4 e	4 f	1.88 f
Maxim XL treated seed	4 f	4 e	4 f	1.35 f
Maxim XL + Micro Plus treated seed	6 ef	6 de	3 f	1.04 f
Agri-Fos 600	21 de	21 cd	21 e	14.13 de
Bayfidan in bentonite	33 cd	38 bc	28 de	5.63 ef
Tebuconazole (tbz) in polymer	44 abc	44 ab	40 cd	23.74 abc
Terraclor	40 bc	43 ab	41 bcd	19.15 cd
Filan	43 abc	44 ab	44 abc	20.57 bcd
Tebuconazole (tbz) in bentonite	46 abc	49 ab	48 abc	14.01 de
Rizolex	50 ab	48 ab	50 abc	22.12 bcd
Amistar	46 abc	59 a	59 a	31.77 a
Non-inoculated control	59 a	59 a	56 ab	29.16 ab

Means followed by same letter do not significantly differ ($P=0.05$, LSD)

DAS = Days after sowing

Figure 5.2 - Pea seedling survival at 47DAS

Discussion

- *R. solani* AG2.1 was highly pathogenic to peas and green beans, causing severe damping off in the untreated and inoculated control (Tables 5.3 and 5.4). The main effect of *R. solani* AG2.1 was in reducing seedling emergence and survival. The differences in the total fresh shoot weights were mainly due to the total number of surviving seedlings. There was only low levels of root rot in the surviving plants.
- In Trial 1 on green beans, Agri-Fos, Filan, Tilt, Switch and Rizolex were shown to be effective as soil drench applications in controlling *Rhizoctonia* and increasing seedling survival and total fresh shoot weights (Table 5.3). The performance of Amistar and LEM 17 were more variable.
- In Trial 2 on peas, Amistar, Rizolex, Filan and Terraclor gave effective disease control, when applied as soil drench applications.
- Seed treatments with Maxim XL, alone or in combination with the biocontrol agent Micro Plus, were not effective under the high disease pressure.
- Trial 2 also showed that tebuconazole and triademinol applied in slow release granules were phytotoxic, with stunted plants. Bayfidan was more phytotoxic than tebuconazole. Tebuconazole coated granules had been used successfully to extend the control of onion white rot in infected soil. However, this study showed that the use of such slow release granules is not feasible for peas or green beans because of phytotoxicity and concerns on chemical residues. Unlike most vegetable crops that are grown for 2 to 3 months, onions are typically grown for up to 4 to 6 months under cool conditions in Tasmania.

6. Field trials to evaluate various products applied as in-furrow spray soil applications

Summary

Two field trials were conducted to screen fungicides and non-fungicides for disease control at Forthside Vegetable Research Station, Tasmania in 2007 and 2008 in *Rhizoctonia* inoculated soil. *R. solani* AG2.1 was highly pathogenic to green beans in both trials, causing severe damping off. Apart from reducing seedling emergence and survival, it also caused above ground infections on stem bases and pods. Rizolex applied at 2 L/ha and Amistar applied at 4 L/ha or 10 mL/100 m row, as in-furrow spray applications, were effective in controlling the pathogen. Rizolex was less effective at the lower rate of 1 L/ha. Amistar soil treatment caused a delay in seedling emergence, whereas Rizolex had no phytotoxic effects. The other two fungicides, Quintozene and Rovral were also not effective. All the non-fungicides including molasses were not effective.

Aims

Various fungicide chemical and non-chemical products were applied as in-furrow spray applications to evaluate their potential for *Rhizoctonia* control in inoculated soil. These were compared against two untreated controls – one inoculated and one not inoculated with *Rhizoctonia*.

Materials & methods

Two seed furrows were prepared 200 mm deep in each soil bed (1.6 m x 36 m long). *R. solani* AG2.1 inoculum was spread along the furrow at the rate of 10 g of colonised millet seed per metre row. No inoculum was applied in the non-inoculated control plots. Plot size was 1.6 metre by 4 metre with two plant rows. The trial design was randomised complete block with four replicates. In Trials 1 and 2, in-furrow treatment applications were applied as in-furrow spray applications with a water volume of 280 L/ha to the seed furrow after the fungal inoculum application and sown with green beans at 50 mm spacing. Trial 1 was sown with green beans cv. Celtic that had been treated with Captan and Thiram, and Trial 2 was sown with green beans cv. Flavor Sweet that had been treated with Thiram. The trial was irrigated by overhead sprinklers twice a week. Seedling emergence and survival, root rot severity, hypocotyl rots, pod infections and yields were recorded.



Figure 6.1: In-furrow spray application after inoculum and seed sowing

Table 6.1 - Treatment details for Trial 1 on green beans

Treatment	Active ingredient	Product rate	Application method
Untreated inoculated control	Nil	Nil	Nil
Amistar	azoxystrobin	4 L/ha	In-furrow band application with 280 L/ha water
Quintozene	quintozene	10 kg/ha	
Rizolex	tolclofos-methyl	2 L/ha	
Molasses	molasses	28.4 L/ha	
Sugar	sugar	28.4 kg/ha	
Micro Plus	<i>Bacillus lydicus</i>	10 kg/ha	
Micro Gyp	calcium sulphate	4 kg/ha	
SoilGard*	<i>Trichoderma</i> spp.	4 kg/ha	

Table 6.2 - Treatment details for Trial 2 on green beans

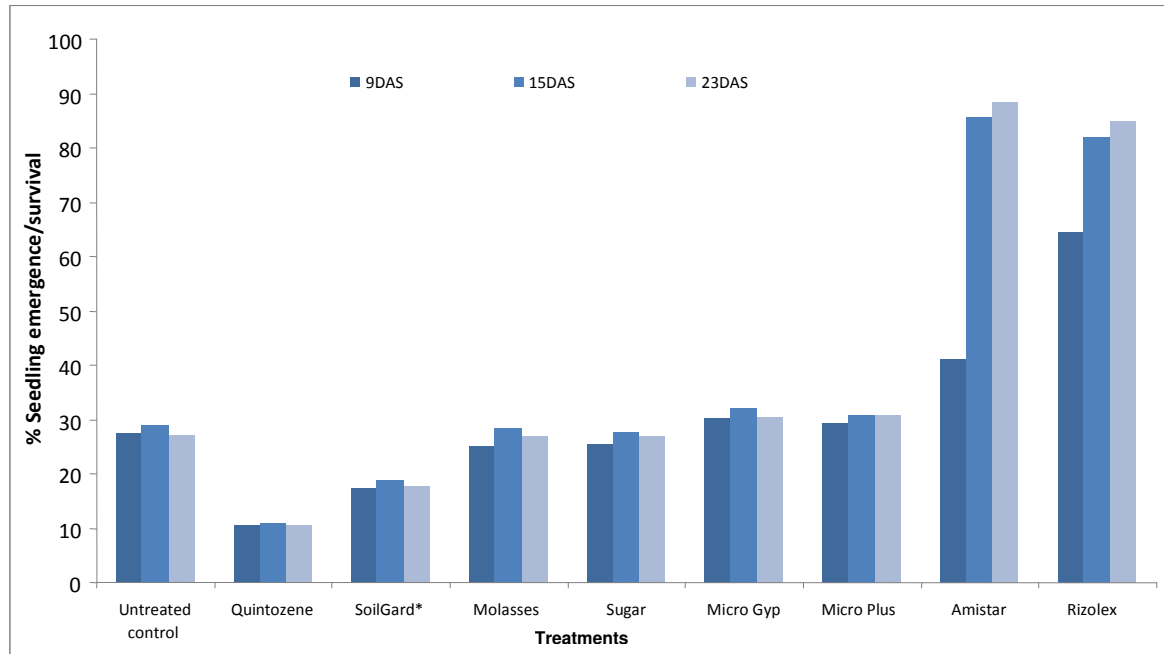
Treatment	Active ingredient	Product rate	Application method
Inoculated control	Nil	Nil	Nil
Des-O-Germ in furrow	quaternary ammonium disinfectant	100 ml/100 L	In-furrow spray application with 280 L/ha water
Rovral Aquaflo in furrow	iprodione	1.0 L/ha	
Rizolex in furrow	tolclofos-methyl	1.0 L/ha	
Rizolex in furrow + molasses	tolclofos-methyl + molasses	1.0 L/ha + 1% (5.7 L/ha)	
Rizolex in furrow	tolclofos-methyl	2.0 L/ha	
Amistar in furrow	azoxystrobin	10 ml/100 m row	Nil
Non-inoculated control	Nil	0.00	

Table 6.3 - Effects of in-furrow soil treatment applications on green beans in Trial 1

Product	Emergence/survival (% plants from total seed sown)			% Pod infected by <i>Rhizoctonia</i>	Pod yield per plot (kg/4m row)
	9 DAS	15 DAS	23DAS		
Untreated control	28 bc	29 b	27 b	9.8 b	3.8 cde
Amistar	41 b	86 a	88 a	0.3 c	6.7 ab
Quintozene	11 c	11 c	11 c	23.5 a	1.5 e
Rizolex	64 a	82 a	85 a	2.0 c	9.2 a
Molasses	25 bc	28 b	27 b	10.8 b	3.1 cde
Sugar	25 bc	28 b	27 b	15.5 ab	3.4 cde
Micro Plus	29 bc	31 b	31 b	13.3 ab	5.0 bc
Micro Gyp	30 bc	32 b	30 b	9.5 b	4.2 bcd
SoilGard*	17 c	19 bc	18 bc	15.3 ab	2.0 de

Thiram + Captan commercially treated bean seed used in the whole trial
 Means followed by same letter do not significantly differ (P=.05, LSD)
 DAS = Days after sowing

Figure 6.2. Treatment effects on the emergence and survival of green beans in Trial 1 *

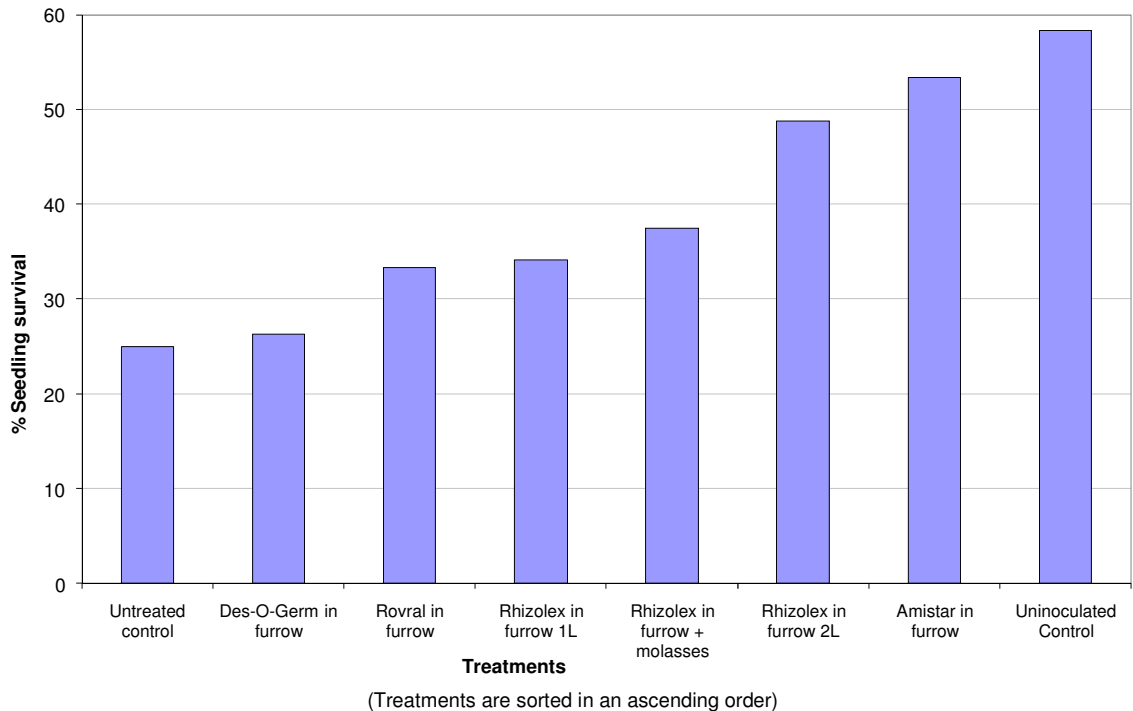


* Note that except for untreated control, all treatments were sorted in an ascending order

Table 6.4 - Effects of in-furrow soil treatment applications on green beans in Trial 2

Product	Rate	Emergence/survival (% plants from total seed sown/plot)		Root rot index	%Plants with hypocotyl rot
		27DAS	48DAS	48DAS	48DAS
Untreated control	0.00	30	25	3.0	65
Des-O-Germ in furrow	100 ml/100 L	28	26	2.4	77
Rovral Aquaflo in furrow	1.0 L/ha	38	33	2.2	41
Rizolex in furrow	1.0 L/ha	34	34	1.9	14
Rizolex in furrow + Molasses	1.0 L/ha + 5.7 L/ha	38	38	1.9	16
Rizolex in furrow	2.0 L/ha	54	49	1.9	18
Amistar in furrow	10 ml/100 m row	63	53	1.5	9
Uninoculated control	0.00	60	58	1.2	5
p-value		0.159	0.182		

Thiram commercially treated bean seed used in the whole trial
DAS = Days after sowing

Figure 6.3. Treatment effects on the survival of green beans in Trial 2 at 48DAS

Discussion

- Under relatively cool and wet conditions, *R. solani* AG2.1 was highly pathogenic to green beans in the inoculated field trials conducted over two seasons in 2008 and 2009. It caused severe damping off in the untreated control (Tables 6.3 - 6.4, Figures 6.2 - 6.3). The major effect of *R. solani* AG2.1 was in reducing seedling emergence and survival. In 2008, in Trial 1, the pathogen also produced basidiospores, the sexual spores, causing above ground infections on stem bases and causing rot on pods that were in contact with the soil. In 2009, in Trial 2, *Rhizoctonia* cankers were noted on hypocotyls.
- In Trial 1, Amistar at 4 L/ha and Rizolex at 2 L/ha, applied as in-furrow spray applications, were highly effective in controlling *Rhizoctonia*, significantly increasing seedling survival and yield of beans. Other in-furrow treatments with Quitozene, SoilGard, molasses, sugar, gypsum and Micro Plus were not effective.
- In Trial 2, even though there were no significant differences in all the data, there was a trend of improved seedling survival with Amistar and Rizolex, applied as in-furrow spray applications. Rizolex appeared to be less effective, when applied at the lower rate of 1 L/ha.
- In both trials, Amistar soil treatment was observed to cause a delay in seedling emergence, whereas Rizolex showed no such phytotoxic effects.



Figure 6.4: *Rhizoctonia* root cankers (1) and green bean pod rots (2 and 3)

7. Effects of plant varieties and seed treatments

Summary

Three trials were conducted to examine the susceptibility of green bean varieties and fungicide seed treatments in *Rhizoctonia* inoculated soil. *R. solani* AG2.1 was consistently shown to be highly pathogenic to green beans, causing severe damping off. Under humid and warm conditions, the pathogen was also shown to produce the perfect fungal stage, causing above ground infections on stem and pod. All green bean varieties were susceptible to *R. solani* AG2.1. Although there appeared to be differences in the susceptibility of bean varieties to *Rhizoctonia*, these differences seemed to be related to the different fungicide seed treatments that were applied by the seed companies. Thiram and Captan, which are commonly used as seed treatments were not effective in preventing *Rhizoctonia* damping off under high disease pressure. Fungicide seed treatments with azoxystrobin and fludioxonil were established to be more effective for *Rhizoctonia* control, when applied in addition to Thiram.

Introduction

A field trial and a pot trial were conducted to determine if there were differences between the commercial green bean and carrot varieties to *R. solani* AG2.1. A further study was conducted with a pot trial to establish the effects of seed treatments on *Rhizoctonia* damping off.

Materials and methods

The field trial was conducted in *Rhizoctonia* inoculated soil at the Forthside Vegetable Research Station, Tasmania. *Rhizoctonia* was applied in the field trial as described in Trial 6, with four replicates. The two pot trials were set up in inoculated soil using the method described in Trial 5 using 1.0% w/v inoculum and three replicates. *R. solani* AG2.1 grown in colonised millet seeds was used as the inoculum. All the seeds had been treated with fungicides as described in the treatment details. All trial designs were randomised complete block. Green bean varieties that were most commonly sown by growers in Tasmania were used. All seed was imported and had already been treated with fungicides at their country of origin. In the third trial, green beans cv. Flavor Sweet that had been commercially treated with Thiram were used as the control standard, and additional fungicide, Amistar, Dynasty or Rizolex, was added as a polymer seed coating. Seedling emergence and survival, root rot severity, pod infections and yields were recorded.

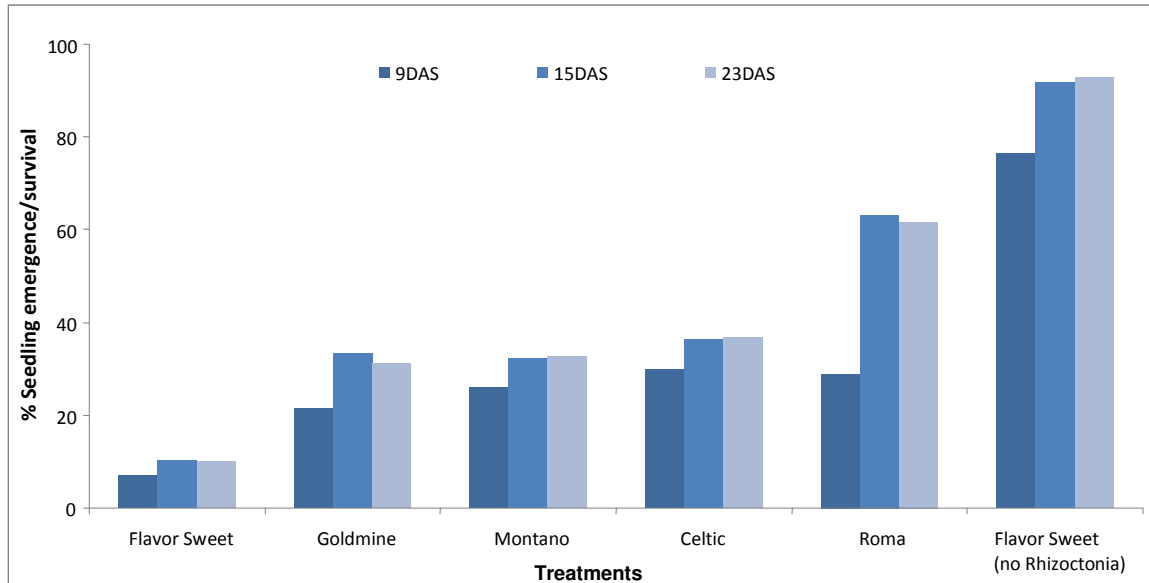
Results and discussion

Table 7.1. The effects of green bean varieties and their respective commercial seed treatments to *Rhizoctonia* in a field inoculated trial at Forthside in Trial 1

Bean variety	Seed treatment	Emergence/survival (% plants from the total seed sown)		
		9DAS	15DAS	23DAS
Flavor Sweet	thiram	7 e	11 d	10 e
Celtic	azoxystrobin + fludioxinol	30 b	37 c	37 c
Montano	maxim + metalaxyl-M	27 bc	33 c	33 c
Roma	captan + thiram	29 b	63 b	62 b
Goldmine	captan	22 bcd	33 c	31 cd
Flavor Sweet (no <i>Rhizoctonia</i> control)	thiram	77 a	92 a	93 a

Means followed by same letter do not significantly differ ($P=0.05$, LSD)
DAS = Days after sowing

Figure 7.1. The effects of *Rhizoctonia* on bean varieties in Trial 1

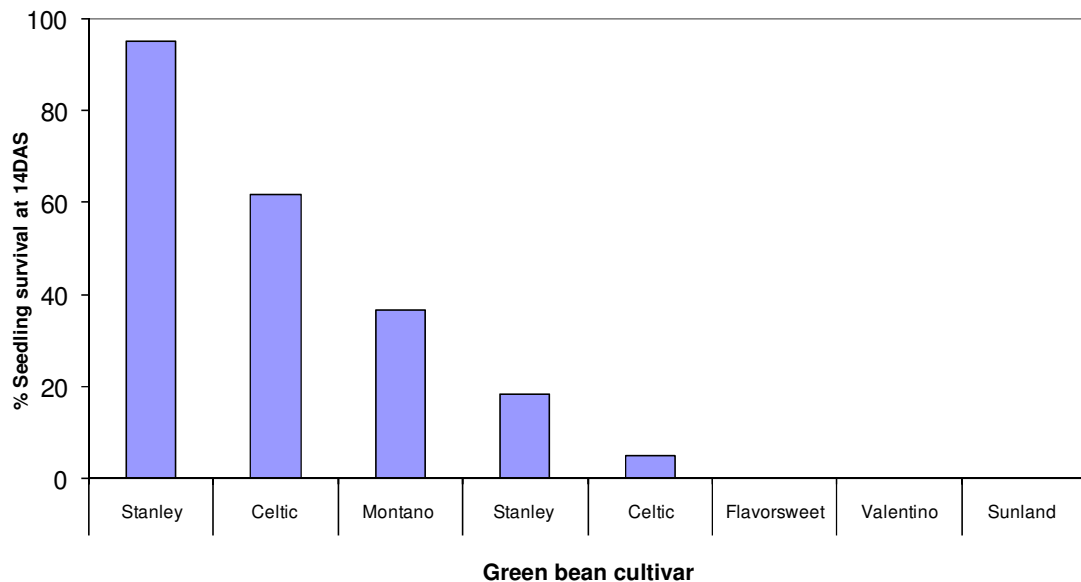


(Treatments sorted in an ascending order according to the seedling emergence levels)

Green bean cv. Flavor Sweet was highly susceptible to *Rhizoctonia* under cool and wet conditions and high disease pressure (Table 7.1). Roma, which had the largest size seed, was the least susceptible because the seedlings were larger and more vigorous in their growth compared to the other varieties with smaller seeds. The other varieties, Goldmine, Montano and Celtic, appeared to be intermediate in their susceptibilities. Note that differences in the fungicide seed treatments between the seed variety may also have some influence on their susceptibility to damping off by *Rhizoctonia*.

Table 7.2. The effects of bean varieties and their respective commercial seed treatments to *Rhizoctonia* in an inoculated pot trial in Trial 2

Variety	Seed treatment	% Seedling survival at 14 days after sowing	
		Non-inoculated control soil	Soil inoculated with <i>Rhizoctonia</i>
Stanley	Azoxystrobin/Fludioxonil/Metalaxyl	93	87
Celtic	Fludioxonil	82	38
Montano	Fludioxonil/Metalaxyl	95	18
Stanley	Fludioxonil/Metalaxyl	95	10
Celtic	Captan/Thiram	90	5
Flavorsweet	Thiram	87	0
Valentino	Captan	98	0
Sunland	Untreated	57	0

Figure 7.2. The effects of bean varieties and seed treatments in *Rhizoctonia* inoculated soil in Trial 2

There appeared to be a correlation between the seed treatments and their susceptibility to damping off by *Rhizoctonia* (Table 7.2). In the *Rhizoctonia* inoculated soil, the differences between the two different seed treatments used for Stanley and Celtic, indicated that seed treated with azoxystrobin + fludioxonil appeared to be most effective in preventing *Rhizoctonia* damping off, followed by fludioxonil. Metalaxyl had no effect on *Rhizoctonia* and hence was excluded in Figure 7.2. Captan and Thiram are broad spectrum fungicides and they appeared to be less effective.

As a consequence to the seed treatments, Flavor Sweet, Valentino and Celtic that were treated with Captan and/or Thiram were highly susceptible to *Rhizoctonia*. Sunland was a local untreated seed variety. Stanley that was treated with both azoxystrobin and fludioxonil was the least susceptible. In comparing varieties that had been treated with fludioxonil, Celtic appeared to be less susceptible than Stanley and Montano.

Table 7.3. Details of seed treatments and on green beans cv. Flavor Sweet in Trial 3

Treatment	Basic commercial seed treatment	Additional seed treatment	Rhizoctonia inoculum in soil
Non-inoculated control	Thiram	None	No Rhizoctonia applied
Inoculated control	Thiram	None	Soil inoculated with Rhizoctonia
+ Amistar	Thiram	Amistar 0.125 g ai/kg seed	
+ Dynasty	Thiram	Dynasty CST 0.13 g ai/kg seed	
+ Rizolex	Thiram	Rizolex 3 g ai/kg seed	
+ Rizolex	Thiram	Rizolex 6 g ai/kg seed	

ai = active ingredient; Dynasty was a seed dressing, which contains three active ingredients (75 g ai/L azoxystrobin, 12.5 g ai/L fludioxonil and 37.5 g ai/L metalaxyl-M); Amistar was based on 250 g ai/L azoxystrobin and Rizolex was based on 500 g ai/L tolclofos-methyl

Table 7.4. The effects of green bean and carrot varieties to Rhizoctonia in an inoculated pot trial in Trial 3

Treatment	Additional seed treatment	Emergence/survival (% plants from total seed sown/pot)	
		20DAS	43DAS
Non-inoculated control	None	90	90
Inoculated control	None	0	0
+ Amistar	Amistar 0.125 g ai	72	68
+ Dynasty	Dynasty 0.13 g ai	53	57
+ Rizolex	Rizolex 3 g ai	72	68
+ Rizolex	Rizolex 6 g ai	55	53

DAS = Days after sowing

In the Thiram only treated seed, 100% seedling mortality was recorded in all three pots (Table 7.4). The addition of Amistar, Dynasty or Rizolex substantially improved seedling survival. This indicated that Thiram has little or no control on Rhizoctonia under wet conditions and high disease pressure. This study verifies the observations in Trial 2 that azoxystrobin, fludioxonil and Rizolex were more effective than Thiram in controlling Rhizoctonia. Azoxystrobin and fludioxonil were relatively new fungicides that had been developed for use in seed dressings by Syngenta in recent years. Thiram, an old broad spectrum fungicide, is still commonly used in treating vegetable seeds. Rizolex had been developed for use as seed piece treatment and soil in furrow treatment for Rhizoctonia control only in potato crops in Australia.

GENERAL DISCUSSION

There is a lack of understanding of *Rhizoctonia* distribution, genetic diversity, pathogenicity and host range in vegetable crops in Australia. Although all the *R. solani* fungal isolates looked the same morphologically, they are actually a collection of genetically diverse groups whose hyphae either repel one another or fused together and exchanges genetic materials (anastomosis). The pathogen had been separated into 12 different anastomosis groups (AGs). Not all AGs can cause disease problems and it is important to find out which AGs are common in vegetable soils and understand their impact in vegetable crop production. In this project, 40 soil samples from intensive vegetable production regions, mainly in Tasmania and a few from Victoria and Queensland, were tested for the presence and levels of important AGs that can cause disease problems, namely AG2.1, AG2.2, AG3, AG4 and AG8. The soil tests showed that the most common AG in the vegetable soils is AG2.1, which was found in 83% of the soil samples. AG3 and AG4 were found in 35% and 25% of the soil samples, respectively.

AG2.1 isolates were shown to be the most pathogenic to peas, green beans and cauliflowers, causing severe damping off on seeds and seedlings. Although not as pathogenic as AG2.1, AG2.2 and AG4 were shown to cause some damping off on pea, green bean and cauliflower. AG3 and AG8 had little or no effects on them. With lettuce, only the AG2.1 bean isolate was highly pathogenic, but not the AG2.1 potato isolate. This indicates that there is variability in the pathogenicity of isolates from even the same AG group. AG2.2 and AG8 also reduced lettuce seedling survival. AG3, a major pathogen on potato, had been found to have no effect on green beans, green peas, lettuces, cauliflowers and carrots in this project. AG3 is believed to be present in all paddocks in Tasmania, where potatoes are part of the crop rotation. However, it is often not detected in soil because the pathogen is believed to survive poorly in soil and is often found on infected tubers or roots of volunteer potatoes.

A bioassay study was also conducted on 24 of the soil samples collected from paddocks sown with green beans in 2006, in order to examine the effect of *Rhizoctonia* on damping-off and root rot severity. Green beans are highly susceptible to *Rhizoctonia* damping off and hypocotyl rots, and hence are ideal for use in bioassays and in studies to screen disease control methods. *Rhizoctonia* was found in association with bean root rot in 63% of the soil samples. In 42% of the soil samples, only the *Rhizoctonia* pathogen could be observed on the root rots. In the pathogenicity study, *Rhizoctonia* AG2.1 caused severe pre- and post-emergence damping off. *Rhizoctonia* also causes hypocotyl rot or basal stem rots. It may restrict root growth of surviving plants and hence could cause early crop senescence as the plant foliage increases in size. Under warm, wet and humid conditions, the pathogen also produced basidiospores, the sexual spores, which can cause above ground infections on stem bases and cause rot on pods that were in contact with the soil or close to the ground.

In paddocks which had been used more intensively for bean production, other root pathogens, namely *Aphanomyces euteiches* and *Thielaviopsis basicola*, the two major root pathogens of green beans were often present as well. This demonstrates that the cause of root diseases can be complex and often more than one soil pathogen is involved. Seed quality and field conditions are also factors that can reduce crop establishment. Therefore, it is vital to identify all causal pathogens, as well as non-pathological factors, so that appropriate action can be taken.

Non-fungicides such as gypsum, molasses, saw dust, sugar, bacteria and fungal biocontrol agents were also screened for *Rhizoctonia* control in inoculated pot soil. Sawdust and molasses were shown to have activity in suppressing *Rhizoctonia*, when applied as pre-plant soil treatments, one month prior to planting. Between these two materials, molasses is considered to have greater potential for commercial use as it is readily available, low cost and is not bulky like saw dust. A subsequent field study, however, showed that under high disease pressure and disease favourable conditions, molasses was less effective in preventing severe damping off. Although the molasses and saw dust were applied at relatively high rates that are not practical for commercial use, the results demonstrated the importance of the role of organic matter in suppressing *Rhizoctonia* diseases. Methods that can increase organic matter in soil such as green manure, re-cycling of organic waste products and various organic amendments may be useful as part of an integrated strategy to suppress and reduce soilborne diseases such as *Rhizoctonia*.

Gypsum granules mixed into soil at 2% to 5% w/v showed some activity for *Rhizoctonia* control, but it is

not as effective as molasses, under high disease pressure. Gypsum applied onto the soil surface and then drenched into the soil had no effect. This finding limits the potential of gypsum for *Rhizoctonia* control, as it must be applied at very high rates. Gypsum is relatively insoluble and therefore must be mixed thoroughly into soil so that it can come into contact with the fungal pathogen in order to have any effect. Unlike fungicide chemicals, gypsum does not completely inhibit growth. Instead, it only suppresses the pathogen by delaying its mycelial spread. It is possible that under lower disease pressure that is more likely to occur in the field, the gypsum soil application may be more beneficial as it is also a fertiliser and soil improver. The biocontrol agents based on *Bacillus lydicus* and *Trichoderma* spp. had little or no effect on *Rhizoctonia*.

The fungicides, Amistar, Agri-Fos, Filan, Tilt, Switch and Rizolex showed potential for controlling *Rhizoctonia* and increasing seedling survival and plant growth in a pot trial. Most of these fungicides were developed for use as foliar applications for foliar diseases. Very few fungicides are suitable for use in soil applications. Amistar (azoxystrobin) and Rizolex (tolclofos-methyl) had been developed for use as seed and soil treatments to control *Rhizoctonia* diseases in potatoes. In two *Rhizoctonia* inoculated field trials, these fungicides applied as in-furrow soil applications at sowing, were also found to be highly effective in preventing *Rhizoctonia* infections and increasing green bean crop establishment and plant growth. Rizolex applied at 2 L/ha and Amistar applied at 4 L/ha or 10 mL/100 m row, as in-furrow spray applications, were effective in controlling the pathogen. Rizolex was less effective at the lower rate of 1 L/ha. Amistar soil treatment caused a slight delay in seedling emergence under relatively cold conditions, whereas Rizolex had no such phytotoxic effects. Therefore, Rizolex is the safer fungicide to use.

Bean seed treatments containing azoxystrobin, fludioxonil or tolclofos-methyl were found to be much more effective than Captan or Thiram in preventing early seedling damping off due to *Rhizoctonia*. Azoxystrobin seed treatment was observed to slightly delay seedling growth. In considering fungicides for seed treatments, it should also be noted that often more than one root pathogen is involved. Hence, a broad spectrum fungicide or the use of a combination of selective fungicides tends to be more useful than a single selective fungicide in protecting the seed, seedlings and early root development.

RECOMMENDATIONS

- The development of commercial testing services to identify *R. solani* to their different AG groups and their inoculum levels in soil will assist growers in identifying and managing Rhizoctonia diseases. Research is currently in progress at SARDI under a new potato research program to develop soil tests for various soilborne pathogens in potato crops, which includes all the major *R. solani* AG groups.
- Further research is being conducted in the new project VG08043 on the two major soilborne diseases of green beans, *Aphanomyces euteiches* and *Thielaviopsis basicola*. This includes the role of *R. solani* in a root disease complex with these pathogens. Molecular soil test that can detect *Aphanomyces* and *Thielaviopsis* will also be developed and tested. Pre-plant soil tests that can detect *Aphanomyces* and *Thielaviopsis* as well as *R. solani* will be very useful to green bean growers in managing the soilborne diseases.
- The importance of the role of organic matter in suppressing Rhizoctonia diseases in this project indicates methods that can increase organic matter in soil such as green manure, re-cycling of organic waste products and various organic amendments, may be useful as part of an integrated strategy to suppress and reduce the soilborne diseases.
- Seed treatment is the most cost effective treatment for preventing Rhizoctonia damping off as well as in protecting seedlings from other soilborne diseases. Usually in root and damping off diseases, more than one soilborne pathogens is often involved. Therefore, in selecting seed treatment options, it is important to identify the major root pathogens for the vegetable crop type as well as in the area planted. With green beans, seed treatments containing azoxystrobin, fludioxonil or tolclofos-methyl were found to be much more effective than captan or thiram in preventing early seedling damping off due to Rhizoctonia.
- Amistar (azoxystrobin) at 4 L/ha or 10 mL/100 m row and Rizolex (tolclofos-methyl) applied at 2 L/ha, as in-furrow soil applications at sowing, were highly effective in preventing Rhizoctonia infections and increasing green bean crop establishment.

TECHNOLOGY TRANSFER

- Meetings with representatives from Nufarm Australia Ltd in 2006, 2007, 2008 and 2009. Detailed reports on all completed efficacy and residue trials and chemical analysis on Filan for Sclerotinia control sent to Nufarm Australia Ltd.
- Project findings were extended to vegetable agronomic consultants in a meeting at Laverton North, Victoria, on 20 February 2007.
- A workshop meeting was held on 8 August 2007 in Devonport to extend project findings
- Participation in the vegetable pathology workshop on 6-7 September 2007 in Melbourne.
- Project findings presented and discussed with other researchers and representatives from the agricultural chemical companies at the Rhizoctonia workshop on 17 October 2007 in Melbourne.
- Meeting with industry representatives, consultants and field officers from vegetable processors in Tasmania on the 26 October 2007 in order to extend useful research information from the Rhizoctonia workshop meeting in Melbourne.
- Project findings presented and discussed with other researchers and representatives from the agricultural chemical companies at the Sclerotinia workshop on 28 November 2007 in Devonport.
- Project findings extended to growers, consultants and field officers from vegetable processors in Tasmania at a bean disease workshop meeting on 14 March 2008 in Devonport (copies of the presentations are attached with this report).
- Provide a progress report for HAL Vegetable Annual Industry Report as requested in September 2008.
- Demonstrations of outcomes from pot trials on bean varieties, seed treatments and biocontrol options to representatives of the major green producers in Tasmania in January 2009.
- A Filan workshop meeting was held on 21-22 January 2010 in Latrobe, north west Tasmania, and Cambridge, south east Tasmania to provide growers with updates on Filan permit use and progress in the product registration for long term use.

APPENDIX

Product Details

Product	Active Ingredient (ai)	Concentration of active ingredient	Formulation
Agri-Fos 600 SC	phosphorous acid	600 g/L	Suspension concentrate
Amistar	azoxystrobin	250 g/L	Suspension concentrate
Cabrio	pyraclostrobin	250 g/L	Suspension concentrate
Captan	captan	800 g/kg	Wettable powder
Des-O-Germ	quaternary ammonium chloride	100 g/L	Liquid
Dynasty CST	azoxystrobin + fludioxonil + metalaxyl-M	75 g/L + 12.5 g/L + 37.5 g/L	Suspension concentrate
Filan	boscalid	500 g/kg	Water dispersible granule
Folicur	tebuconazole	430 g/L	Suspension concentrate
LEM 17	experimental	Not available	Suspension concentrate
Rizolex Liquid	tolclofos-methyl	500 g/L	Suspension concentrate
Switch	cyprodinil + fludioxonil	375 g/L + 250 g/L	Suspension concentrate
Terraclor	quintozene	750 g/kg	Wettable powder
Thiram	thiram	800 g/kg	Wettable powder
Tilt	propiconazole + cyproconazole	250 g/L + 80 g/L	Suspension concentrate
Agm Trichoderma	<i>Trichoderma</i> spp.	10 ⁶ colony forming units/g	Granules
Micro Plus	<i>Bacillus lydicus</i>	10 ⁷ colony forming units/g	Powder
SoilGard	<i>Trichoderma virens</i>	10 ⁶ colony forming units/g	Granules