Understanding the implications of pastures on the management of soil-borne diseases of seed potatoes

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Final Report
Horticulture Australia Project PT04001

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Purpose of project

In Australia, seed potato crops are often grown in rotation with a pasture phase. This project investigated the effects of the different pasture species on soil-borne pathogens and on disease and yield of potatoes. The aim was to establish principles for the management of rotations to minimise diseases of seed potatoes.

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1 Media Summary

Many seed potato crops are grown after a pasture phase of three or more years duration. Australian research has shown that pastures consisting of fodder legumes and grasses replenish soil nitrogen and restore soil structure after cropping. A field study in the Central Highlands of Victoria showed that a pasture phase can improve yields and reduce the build-up in populations of soil-borne pathogens. In the trial, yields were higher in potatoes grown after one and two-years of a pasture or fodder crop in a rotation than in continuously cropped potatoes. DNA tests provided evidence that populations of *Rhizoctonia solani* and *Spongospora subterranea*, which cause Rhizoctonia stem canker and powdery scab, were lower in rotations of perennial ryegrass/potato, white clover/potato and fodder Brassica/potato than in continuously cropped potatoes. However, the one and two years break from potatoes did not result in any significant reductions in the diseases Rhizoctonia stem canker, black scurf, black dot, silver scurf and powdery scab. This suggests that the rotations did not reduce pathogen populations below disease thresholds.

This study found that the 'break' from potatoes with pasture and fodder species had a greater effect on yield and pathogen populations than the individual components of the pasture, such as perennial ryegrass, white clover, or other crops such as fodder Brassica. The incidence and severity of diseases in potatoes did not differ between rotations of perennial ryegrass, white clover or fodder Brassica. This was supported by the DNA tests which showed no difference in populations of *R. solani* and *S. subterranea* between rotations of ryegrass, clover or fodder Brassica. White clover and fodder Brassica roots systems were found to support *R. solani*. However, the fungus was not found on ryegrass roots, and this suggests that selective removal of clover in pastures could potentially help manage Rhizoctonia stem canker.

The pasture phase clearly provides benefits for potato production, including a reduction in pathogen populations. Despite this, diseases remain a problem in the pasture-potato system. Other disease management options, such chemical treatment of seed potatoes, chemical treatment of soil, green manure crops, organic soil amendments, tillage practices and control of volunteer potatoes in the rotation, need be integrated in the pasture-potato rotation to further reduce or suppress pathogens and reduce disease.

2 Technical Summary

A field trial was established in the Central Highlands of Victoria (north-east of Ballarat) on a ferrosol soil (pH 5.2) to study the effects of different components of pasture (perennial ryegrass and white clover) and a fodder Brassica, in rotation with potatoes, on soil-borne pathogens, disease and yield of potatoes (2001-2005). Rotation treatments included: continuous potato cropping without a break; potatoes after one year of ryegrass, clover or Brassica (2002/03); potatoes after 2 years of ryegrass or clover (2003/04) and potatoes after a second cycle of ryegrass/potato or clover/potato (2004/05). As the one and two-year rotations were not in 'phase' (potatoes in the same season) a direct comparison between a 1-year and 2-year break from potatoes was not possible.

Rhizoctonia stem canker (*R. solani*), and the tuber diseases black scurf (*R. solani*), black dot (*C. coccodes*), silver scurf (*H. solani*) and powdery scab (*S. subterranea)* were common each season. Total and marketable yield in continuously cropped potatoes was significantly less than in potatoes cropped after one and two years of a pasture/fodder crop. However, disease incidence and severity generally did not differ significantly between continuous potatoes and potatoes in rotations. Deterioration of soil structure may have contributed to the reduced yields. DNA testing of soil after a second cycle of the one year rotation of ryegrass, clover or fodder Brassica with potatoes, found significantly lower quantities of *R. solani* AG3 (potato strain) and *S. subterranea* DNA in the rotation than in continuous potatoes, which shows that the break from potatoes reduced inoculum build-up.

Generally, there were no significant differences in disease incidence and severity and yields in potatoes after ryegrass, white clover or fodder Brassica. A 3-year study showed that fodder Brassica and white clover root systems supported epiphytic populations of *R solani* sclerotia and hyphae, whereas ryegrass roots did not. A beet seed bioassay indicated higher populations of *R. solani* in a clover/clover/potato treatment than in grass/grass, wheat/clover and continuous potato treatments. This suggests that clover and Brassica could potentially help maintain populations of *R. solani*. However, DNA tests found no significant differences in *R. solani* AG3 inoculum between rotations of ryegrass, clover and fodder Brassica, which suggests that clover and Brassica may not have a significant impact on populations of the pathogen. In contrast, higher populations of *R. solani* AG2-1 were found in the fodder Brassica rotation than in the clover, ryegrass or continuous potato treatments indicating the selection of Brassica specific strains of *R. solani* in that rotation.

The documented benefits of a pasture phase after cropping include the replenishment of soil nitrogen and restoration of soil structure. Our study showed that the breaks of one and two years with pasture and fodder species from potatoes improved yields and reduced the build-up inoculum of soil-borne pathogens. The break period had a greater impact than the individual pasture and fodder crops in the rotation, although there was some evidence that clovers and Brassica could support *R. solani*. Pathogen populations over the period of this trial were maintained above disease threshold levels in the pasture phase. Other disease management options, such chemical treatment of seed potatoes, chemical treatment of soil, green manure crops, organic soil amendments, tillage practices and control of volunteer potatoes in the rotation, need to be integrated in the pasture-potato rotation to further reduce or suppress pathogens and reduce disease.

3 The implications of pastures on the management of soil-borne diseases of potatoes – a review

3.1 Introduction

A significant proportion of the seed potato crop in Australia is grown after a pasture phase in mixed enterprise businesses involving livestock. In the past, seed potato production was restricted to regions that had well structured fertile soils and reliable spring and summer rainfall. However, growers now exercise greater flexibility in where they grow potatoes.

Regulations for certified seed potato production stipulate that early generation seed (G1-G3) can only be grown after a minimum 5-year break from potatoes and a minimum of a 3-year break for later generations (G4-G5). It is now common practice for seed growers to lease 'new' ground that has never been cropped to potatoes before. These areas are commonly long-term established pastures on the fringes of traditional production areas.

In a previous study we reviewed the general issue of the effects of rotation on potato diseases (de Boer 2003a). The single most important factor that influenced disease incidence and severity in potatoes was cropping frequency in rotations (Lamers *et al.* 1989; Vos and Van Loon 1989). Data from several field trials around the world showed that yield was reduced as the frequency of cropping potatoes in rotation increased. Diseases caused by nematodes and fungi were the major contributing factors to yield loss in these trials (Bollen *et al.* 1989; van Loon 1992; Vos and Van Loon 1989).

Generally, three years is considered to be a minimum break from potatoes in a rotation in order to maintain productivity. Optimum rotations will depend on local factors, including soil type, climate, crop species grown and disease pressure. Some research indicates that a 3-year rotation is sustainable in a well managed system (Peters *et al.* 2003).

3.1.1 The benefits of pasture

Common pasture rotations in Victoria comprise three or more years of a clover based pasture, followed by a potato crop, then one or two consecutive crops of oilseed Brassica, fodder Brassica, cereals, legumes or green manures.

To our knowledge, there are no published reports on the effects of pasture on potato production. However, there have been several studies of the impact of pastures and pasture legumes on the yield of wheat that serve to illustrate the potential benefits of the pasture phase to potato productivity.

Legume based pastures grown for 5-8 years accumulate nitrogen and restore soil structure after the cropping phase (Ellington 1986). Under Victorian conditions, pasture legume crops such as clovers and medics can increase soil nitrogen at rates of 20-80 kg/ha/year (Reeves *et al.* 1987). This stimulates the growth of grass in the pasture, as well as the following non-legume crops. The amount of nitrogen produced by one year of legume based pasture appears to be sufficient to provide a positive nitrogen balance for at least one wheat crop (Peoples *et al.* 1998; Zhang and Evans 2004).

Different legumes differ in their effect on the subsequent crop due to differing capacities for nitrogen fixation or differences in the amount of organic matter produced. The proportion of legume and grass in mixed pastures can affect the yield of subsequent crops. Moore and Grace (1998) predicted that each extra tonne of legume pasture would result in 1.12 tonne of extra grain yield/ha. Zhang and Evans (2004) found that the more dry matter produced by a clover pasture the greater the yield of the wheat crop.

Soil organic matter content is important not only for soil fertility but also for improving soil structure and water holding capacity. Root growth improves soil stability, and roots leak organic substances and slough off cells, which support fungi, earthworms and other soil organisms. All of these contribute to binding the soil particles together. As well as adding organic matter, the roots of plants form macro-pores in the soil, increasing aeration and water flow and decreasing density (Murphy and Harte 1992). There can be as much as 6 tonnes/ha of root material in the soil under a long term grass stand (Murphy and Harte 1992). Research in Canada has shown that potatoes add relatively little root residue (about 300 kg/ha) to the soil, compared with cereals (1500-2500 kg/ha) or grasses (3000-5000 kg/ha) (Carter 1996).

Reid and Goss (1981) reported that ryegrass and lucerne increase soil aggregate stability, while maize, tomato and wheat did the opposite. In cereal cropping, pasture breaks have been shown to increase the level of water stable soil aggregates compared to continuous cropping (King 1984). Beneficial effects have also been found in potato fields in Canada, where the soil structural stability at the soil surface (0-10cm) was improved by Italian ryegrass, red clover or barley grown in rotation with potatoes (Carter *et al.* 2003).

3.2 The influence of rotation on soil-borne diseases of potatoes

The following potato diseases are common in potato production in Australia

- Rhizoctonia stem canker and black scurf (*Rhizoctonia solani*)
- Powdery scab (*Spongospora subterranea*)
- Common scab (*Streptomyces scabies*)
- Silver scurf (*Helminthosporium solani*)
- Black dot (*Colletotrichum coccodes*)

Emerging problems include Verticillium wilt (*Verticillium dahliae*), the root knot (*Meloidogyne* spp.) and root lesion nematodes (*Pratylenchus* spp.).

3.2.1 Rhizoctonia Canker & Black Scurf (*Rhizoctonia solani*)

Rhizoctonia canker and black scurf, caused by the fungus *Rhizoctonia solani*, is widespread and common in Australia and affects emergence, plant growth, tuber yield and tuber quality (Baker 1970; Banville 1989; Banville *et al.* 1996; Hide *et al.* 1973).

Life-cycle

R. solani is a versatile soil-inhabiting fungus adapted to survival under a diverse range of conditions, affecting a wide range of crop species world-wide (Anderson 1982; Ogoshi 1987). The fungus can be found in native vegetation as well as in agricultural production

areas (Cother 1979; Ogoshi 1987). It survives in soil as sclerotia (red/black thick walled structures - 'black scurf' on potato skins) and as thick walled hyphae (fungal threads) in soil and on tubers or in plant debris (Papavizas *et al.* 1975). Under favourable conditions, the fungus grows actively in soil, colonising the roots of potatoes and other plants. The fungus can establish non-symptomatic endophytic relationships with other crops and weed plants (Carling *et al.* 1986) and can survive in organic matter in the absence of hosts. The sclerotia develop on the surface of tubers when potato plants begin to senesce. Chemicals leaching from maturing tubers signal the fungus to 'shut-down' for the season (Dijst 1990).

Anastomosis Groups

Rhizoctonia solani has a wide host range and is capable of infecting or colonising many different species of crops and weeds from many different plant families (Adams 1988; Anderson 1982; Ogoshi 1987). The fungus is divided into 13 sub specific groups called Anastomosis Groups (AGs) (Carling *et al.* 2002; 1991) based on somatic (vegetative) incompatibility responses between hyphae of genetically distinct isolates (anastomosis reactions) (Carling *et al.* 1988). Each of these groups has different degrees of specialisation to specific families of plants. Some have a high degree of host specificity. AG3, for example, is most commonly isolated from potatoes and AG8 from cereal roots (Banville *et al.* 1996).

Of the 13 anastomosis groups of *R. solani*, six have been associated with potatoes. These are AG1, AG2 (subgroups 2-1 and 2-2), AG3, AG4, AG5 and AG9 (Banville *et al.* 1996). Isolates of selected AGs most notably AGs 3, 4, 5 and 8 are capable of doing moderate to extensive damage to potato plants. Most of the sclerotia isolated from potato tubers are reported to belong to AG3 (Banville *et al.* 1996; de Boer 2003a; Petkowski *et al.* 2003).

Anastomosis groups of Rhizoctonia in potatoes in Australia

Six different AGs have been associated with potatoes in Australia (Balali *et al.* 1995; de Boer and Petkowski 2004; Petkowski *et al.* 2003; Petkowski and de Boer 2001). Balali *et al.* (1995) collected isolates of the fungus from stems, roots, tubers and soil in potato crops grown in Virginia and Lenswood in South Australia. Of 301 isolates tested, 90% were AG3 and 7% and 2% were AG4 and AG5, respectively. AG3 and 5 caused stem and root cankers and black scurf. AG4 caused stem cankers, severe root cankers and significantly reduced the number and volume of fine roots (feeder roots) but did not produce black scurf (Balali *et al.* 1995).

In Victoria isolates of AG2-1 and AG2-2 have been implicated as the cause of a Rhizoctonia wilt disease in potatoes (de Boer 2003a; de Boer and Petkowski 2004; Petkowski *et al.* 2003). However, this disease is very rare and only affects mature potato crops.

A study of potato crops in two districts of Victoria (de Boer 2003a) found not only AG3, 4 and 5, but also AG2 (subgroups AG2-1 and AG2-2). The AG3s were the most common, accounting for more than 60% of the isolates of the fungus, whilst the AG2s accounted for about 25% (Petkowski *et al.* 2003; Petkowski and de Boer 2001). The AG2s proved to be very pathogenic to potatoes as well as different *Brassica* species and red clover. This is the first report of AG2 causing significant damage to potatoes. It may be that the Australian strains differ to those found elsewhere. The AG2 groups are associated with Crucifers (*Brassica*) and Legumes, as well as many other crops (Hwang *et al.* 1996; Wong *et al.* 1985; Wong and Sivasithamparam 1985). The predominance of clover pastures and the use of

fodder Brassica crops in Central Highlands region where our studies were conducted may explain the presence of the *R. solani* AG2.

R. solani AG3, the 'potato strain' is able to grow and reproduce on the roots of *Brassica* species and clover forming an epiphytic relationship with its hosts in Australia (de Boer 2003a). Carling *et al.* (1986) reported a similar relationship between the pathogen and many other plant species. .

Anastomosis groups and crop rotation

The saprophytic survival capability of *R. solani* in debris of specific crops (Boosalis and Scharen 1959; Papavizas 1970; Papavizas *et al.* 1975; Specht and Leach 1987; Umaerus *et al.* 1989) and the pathogenicity of specific strains of *R. solani* selected by various preceding crops (Gudmestad *et al.* 1989; Johnston *et al.* 1994; Umaerus *et al.* 1989) will also influence the composition of soil populations pathogenic to potatoes (i.e. AG3) and consequently disease severity in succeeding crops.

Errampalli and Johnston in Canada (unpublished data) characterised *R. solani* AGs isolated from seven different rotation crops. They recorded 87% of the more aggressive AG3 and 13% of the less aggressive potato pathogen AG5 on potatoes grown after rotation crops. Within the rotation crops, barley harboured AG3, whereas winter wheat, orchard grass and ryegrass carried AG5. This suggests that barley may not be a good rotation crop for *R. solani* control.

The occurrence of AG3 has been shown to build up with increased cropping with potatoes and to gradually decline during the rotation period. Jaeger and Velvis (1989) found that after 3 or 4 years rotation, AG3 disappeared and was replaced by AG5. Gudmestat *et al*. (1989), found that AG4 and 5 were the most dominant groups in potatoes sampled from fields that did not have a history of potato cropping, whereas AG3 and 5 were dominant on potatoes taken from fields with a history of potatoes. The incidence and severity of Rhizoctonia damage was highest in fields with a history of potatoes.

The relationship between rotation, the frequency of potato cropping and *Rhizoctonia solani*

Cropping Frequency

There have been many studies on the effects of rotations on *R. solani* around the world. The results are likely to vary with differences in rotations systems, soil type, climates, cultivars and cultural practices. The most important factor determining the incidence and severity of disease is cropping frequency (Scholte 1987; Scholte 1989; Scholte 1992). The more frequently potatoes are grown in a rotation, the greater the incidence and severity of damage in potato crops (Carter and Sanderson 2001; Gilligan *et al.* 1996; Scholte 1992). Research in Canada report significantly less stem canker and black scurf in a 3-year rotation than a 2-year rotation (Carter and Sanderson 2001; Peters *et al.* 2004) and this was attributed to the break from potatoes , rather than an effect of a specific rotation sequence. The reduction in disease with reduced cropping frequency can be explained by a reduction in inoculum. Gilligan *et al.* (1996), found that the amount of inoculum in the soil quickly dropped off to low levels with longer rotations, after 1 year in a 6 year rotation and 2 years in a 4 year rotation. However, inoculum was quickly replenished after growing a potato crop.

Crop species and rotation sequences

The effects of specific crops or crop sequences that precede potatoes disease are not clear-cut and often contradictory. Recommendations to growers often mention grasses and cereals a good rotation crops for control of *R. solani* (Banville *et al.* 1996) although this is not clear from research data.

A number of studies have examined the prevalence of *R. solani* in rotation crops. However, often there has not been a correlation between this data and disease in the subsequent potato crop (Celetti *et al.* 1989a; b; Celetti *et al.* 1990). For example, Celetti *et al.* (1989b) reported a higher incidence of *R. solani* in clover stands than in Italian ryegrass and other crops. However, corresponding differences in Rhizoctonia stem canker levels in subsequent potato crops grown after the different rotations were not evident (Celetti *et al.* 1989a; b; Celetti *et al.* 1990). Johnston *et al* (1994) reported a higher incidence of *R. solani* sampled from clover plants than ryegrass or wheat but the incidence and severity of *R. solani* was no different in potatoes rotated with ryegrass and clover.

Larkin and Honeycutt (2006) report that potato crops rotated with clover or soybean, had higher levels of Rhizoctonia damage compared with potatoes rotated with barely and oil-seed Brassica crops. Scholte (1992) reported that *Rhizoctonia* disease levels were unaffected by rotation with sugar beet, maize, wheat or barley. The varied effects of rotation on *R. solani* indicate that rotation systems that are suited to local conditions and practices need to be developed.

Tillage and Rotation

Researchers in Canada have been conducting research on rotations which include conservation tillage (Carter and Sanderson 2001; Peters *et al.* 2004; Peters *et al.* 2003; Sturz and Carter 1995). They showed that a three-year rotation with barley and red clover reduced disease caused by *R. solani* over a two year rotation with barley in a minimum tillage system. There were indications that the soils minimum tillage in the three-year rotation were suppressive to R. solani Others have reported that reduced tillage systems significantly lowered soil populations of *R. solani* (Gudmestad *et al.* 1978; Leach 1993). Peters *et al.* (2004) also indicate that soil suppressiveness can also be linked to activities such as green manuring and the addition of organic amendments (Sturz *et al.* 1997). This work indicates that the effects of rotation can be enhanced by practices that improve soil health.

3.2.2 Powdery Scab (*Spongospora subterranea*)

Spongospora is an obligate parasite. This means it needs a live host to multiply and complete its life-cycle. The fungus survives in soil as clumps of spores called cystosori (spore balls) which are highly resistant to desiccation (Harrison *et al.* 1997). Research in Victoria found that spores were still viable after four years in soil (R. F de Boer, unpublished data). Circumstantial evidence suggests that they can last a decade or more.

Powdery scab disease of potatoes was reviewed recently by Harrison et al. (1997). There are two stages in the life cycle of this fungus. Individual spores in cystosori germinate under cool, wet conditions, probably stimulated by chemicals leaching from potato roots, and produce swimming spores (zoospores), which infect the fine hairs on the surface of potato

roots. The fungus multiplies in the root hairs producing another crop of zoospores and the cycle can be repeated in fresh root hairs. This process allows a rapid multiplication of the fungus before tubers are initiated. However, the zoospores are unlikely to survive in soil for long (days or weeks) and may not have an important role in the survival of this fungus in the long-term. In the second stage of the life cycle, a different process of multiplication occurs in infected roots and tubers resulting in the development of survival spores (cystosori) in root galls (seen as white galls or nodules on roots) and powdery scab pustules.

Research has shown that *Spongospora* is able to infect the roots of a very wide range of plant species in glasshouse tests (Harrison *et al.* 1997). The host list includes tomatoes, Solanaceous weeds, grasses, cereals, various clovers and fodder Brassica (de Boer 2001). However, these primary zoospores are unlikely to survive in soil.

Previous research was not able to demonstrate any effects of rotations or 4 years on scab incidence and severity (de Boer 2001; de Boer 2003a; de Boer and Theodore 1997). This is probably due to the longevity of the very resistant powdery scab cystosori. Wale (1987) considered that a 5-year rotation was too short for Scotland.

Powdery scab is sensitive to soil pH, temperature and water content (Harrison *et al.* 1997). There is anecdotal evidence of a higher risk of powdery scab in potatoes cropped after improved pastures (clover/grass pastures) that have been limed. Adding lime to soil can increase the risk of powdery scab. Rotations that improve soil structure could potentially reduce powdery scab. The incorporation of organic amendments into soil has been shown to reduce powdery scab in sandy soils (Crump and de Boer 2003) and such treatments could be integrated into a rotation.

3.2.3 Silver Scurf (*Helminthosporium solani)*

Silver scurf is one of the most common diseases of potato tubers around and is particularly common seed potatoes in Australia (de Boer and Wicks 1994). The fungus only affects the potato skin, reducing tuber quality but not yield. De Boer (1997) reviewed the biology and life-cycle of the pathogen. Until recently, it was believed that *H. solani* did not survive more that a few months in soil and was mainly perpetuated through the planting of infected seed potatoes. However, research in Victoria shows that the fungus is soil-borne and widespread in traditional potato cropping districts (de Boer 1997; 2003b). When disease-free mini-tubers (produced in a glasshouse) were planted after 8 years of pasture, 100% of their progeny were affected with silver scurf, indicating that inoculum of *H. solani* was soil-borne.

For a disease as common as silver scurf, surprisingly little is known about the ecology of the fungus and how it survives in soil. *H. solani* is not known to have any other hosts and does not grow on the roots of potato plants. A study in the USA found that the fungus can multiply on the roots of dead grasses (Merida and Loria 1994) which suggests that it may have some activity as a saprophyte in soil. Volunteer potatoes probably play an important role in the survival of this pathogen in the period between potato crops.

Previous research in Australia did not find any effects of rotations on silver scurf (de Boer 2003a). The planting of diseased seed will nullify the effects of rotation (de Boer 2003b). Peters et al. (2003) found that silver scurf was less severe after 3 years rotation compared with 2 years in minimally tilled fields and hypothesised that effect was related to the distribution of inoculum in soil.

H. solani is an enigma. It is clear that the fungus survives in Australian soil for several years or more. However, much more needs to be learnt about how the fungus survives without its potato host and whether it utilises other species, such as grasses, in the rotation.

3.2.4 Black Dot (*Colletotrichum coccodes*)

Colletotrichum coccodes infects both potatoes and tomatoes. The disease black dot is common and widespread in Australia as a tuber blemish disease of seed and commercial potatoes (de Boer and Wicks 1994) and has proved to be very common in field trials conducted in Victoria and South Australia (de Boer 1997; 2003a; b). Black dot gets its name from the abundant small, dot-like black sclerotia that are found on senescent and dead potato roots, stolons and stems below and above the ground, as wells as on tubers (Dillard 1992). The fungus progressively colonises the roots, stems, stolons and tubers of potato plants as the crop develops. Severely affected plants can wilt under stress, although the fungus usually does not cause serious damage to potato plants (Harrison 1963).

Very little is known about the ecology of this fungus in Australia. It is reported to survive in soil for up to 8 years as sclerotia in crop debris (Dillard and Cobb 1998). Studies in the USA found the number of propagules of *C. coccodes* in soil to be related to the history of potato cropping (Barkdoll and Davis 1992). The fungus was common in ground with a history of potatoes but undetectable in virgin ground. In Victoria, the disease was relatively common in potatoes grown after 8 years of pasture in a study where disease-free minitubers were planted as seed (de Boer 1997). The fungus can colonise roots and stems of many plant species besides potato and tomato and these hosts may also play an important role in its survival in the period between potato crops (Raid and Pennypacker 1987). Weed hosts include the nightshades, shepherds purse and fat hen. Legume crops planted in rotation may support a higher incidence of black dot than grasses (Celetti *et al.* 1989a; b; Celetti *et al.* 1990), and soybeans were found to have higher levels than either clover or grasses.

In trials conducted in Victoria and South Australia, the incidence and severity of black dot did not vary significantly with rotation (de Boer 2003a). The number of propagules of *C. coccodes* in soil at Langhorne Creek and Woodside in South Australia did not correlate well with disease incidence on tubers at harvest. It was also found by Celetti *et al.* (1989) that the incidence of black dot on rotation crops (clover, ryegrass and winter wheat) did not affect the levels of the disease on the following potato crop. Surveys of Russet Burbank crops in Victoria did not find a relationship between the duration of the break from potatoes (1-2 years or 5-6 years). It has been reported that that the numbers of black dot micro sclerotia in the soil decreased with increasing rotation length (Bollen *et al.* 1989).

Our experience suggests that inoculum introduced on the seed piece plays a major role in the epidemiology of this disease, nullifying any potential benefits of crop rotation. The challenge is to break the cycle. This will require an understanding of the relative importance of seed verses soil-borne inoculum, the survival of *C. coccodes* in Australian soils and the relationship between the fungus and other crop species in rotations, particularly pasture species. The ability of the fungus to survive for several years in a dormant state in soil makes it a particularly difficult candidate for management.

3.2.5 Pink Rot (*Phytophthora erythroseptica*)

Pink rot occurs in most major cropping areas of Australia. It is generally caused by the fungus *Phytophthora erythroseptica*. The disease occurs in patches and growers have observed that the disease can reappear in the same patches of the field after a 4 year rotation with pasture. Warm, wet soils favour disease development, especially areas of poor drainage following rain or irrigation. The pathogen can survive in the soil for three or more years as resistant zoospores or grow on decaying plant matter and the roots of various crop and pasture species (Horne *et al.* 2002).

Potato infecting strains of *P. erythroseptica* has been reported as pathogens of tomatoes, spinach and tulips (Lambert and Salas 2001). *P. erythroseptica* has also been reported as a pathogen of lupins (Trapero-Casas *et al.* 2000) and some species of clover (Pratt 1981). In pot tests, potato-infecting strains of *P. erythroseptica* have been recovered from 17 of 90 non-solanaceous plants tested, including wheat and rye (Lambert and Salas 2001).

There is a paucity of published research on the effects of rotation and rotation crops on the persistence of the pathogen. The effects of rotation was studies in a trial in Canada, which included a 2-year rotation of spring barley and potato (cv. Russet Burbank) and a 3-year rotation of barley, red clover and potato under conventional and minimum tillage (Peters *et al.* 2005). Soil and potato tubers were sampled 6-7 years after the field trial began. Pink rot disease development in inoculated tubers was less pronounced in potatoes from a 3-year rotation than the 2-year rotational soil. Potato plants grown in greenhouse trials in 3-year rotational soils were less diseased than those grown in 2-year rotational soils following inoculation. This suggested that potato plants grown in soils managed under a 3-year rotation are intrinsically more resistant to pathogen attack than those managed under a 2-year rotation. Peters *et al.* (2005) concluded that crop rotation provides benefits beyond those normally associated with pathogen population decline in the absence of a susceptible host. They hypothesised that beneficial soil micro flora and tuber endophytes are involved in disease suppression.

3.2.6 Common Scab (*Streptomyces* spp.)

Common scab (*Streptomyces* spp.) has been a persistent problem for Russet Burbank producers on the north coast of Tasmania and has been an increasingly serious problem for growers on pasture potato systems in southern Victoria over the past decade.

Surprisingly little is known about this disease in the Australian environment. Australian isolates of *Streptomyces* spp. are currently being characterised (Wiechel *et al.* 2003). Wilson *et al.* (1999) examined the effects of seed-borne inoculum seed and soil chemical treatments on common scab. Current research is focused on managing the pathogen with organic soil amendments under the Processing Potato R&D Program.

Streptomyces survives in decaying organic matter and possibly on the roots of living plants (Loria 1994). Hosts of *Streptomyces*, other than potatoes, include a range of root crops such as red and sugar beet, carrot, parsnip, radish, rutabaga and turnip (Loria *et al.* 1997). Ransom *et al.* (1994) and Wilson *et al.*(1994) reviewed the published literature on the effects of rotation on common scab. They reported conflicting results on the effects of crop rotation and green manuring on common scab due to the complex nature of the pathogen interactions with the inherent variability of different cropping systems. In a recent review conducted in the United Kingdom (S.J. Wale, unpublished), it was concluded that there are conflicting reports

in the literature of the effects of crop rotation on common scab incidence, probably due the greater effects of environmental variation. There was little strong evidence on which crops reduce or increase common scab risk, and there was some evidence that increased cropping with potatoes does not increase the risk of common scab.

Anecdotal observations from the United Kingdom suggest that a pasture phase aggravates common scab. Common scab was said to be more troublesome for two to three years after land had been ploughed out of permanent grass although no explanation for this effect was provided (Brenchley and Wilcox 1979).

Recent studies in Maine found that canola and rapeseed in 2-year and 3-year rotations reduced the incidence of common scab by up to 25% relative to continuous potatoes and other rotation crops (Larkin 2006). A red clover rotation also increased disease. The addition of a winter rye cover crop to 2-year rotations resulted in further reductions of common scab across all rotation crops. The combined effect of a canola or rapeseed rotation with a fall cover crop resulted in disease reductions of 30% of common scab. Substituting ryegrass for clover as an under seeded cover crop with barley also reduced scab severity.

A very promising area of research for the management of common scab is the use of organic soil amendments. Various types of amendments, such as blood and bone meal and fermented swine manure, have been shown to reduce the incidence and severity of common scab and other pathogens (Lazarovits 2001a; b).

3.2.7 Verticillium Wilt (*Verticillium dahliae*)

Verticillium wilt has been recognised as a disease of potatoes in Australia for some time (Harrison 1967; Sampson 1980). *Verticillium dahliae* infects roots and stems and inhabits the vascular system of plants causing early senescence of stems or whole plants and is a major disease of potatoes worldwide.

V. dahliae forms a complex with *Pratylenchus* spp., namely *P. penetrans*, to cause the 'potato early dying' syndrome (Martin *et al.* 1982; Rowe *et al.* 1987). The nematode and fungus interact synergistically to cause early dying of potato at population densities which individually have little or no effect on potato.

Very few studies have been conducted in Australia on Verticillium wilt and on *Pratylenchus* spp. in Australia. Harding and Wicks (2007) conducted surveys of soils and plant material for the incidence of *V. dahliae* and *Pratylenchus* spp. They found the fungus and the nematode to be widespread in potato soils throughout Australia. *P. crenatus* and *P. neglectus* were the predominant nematode species but the incidence of *P. penetrans* was low. They hypothesise that densities of *V. dahliae* are at thresholds sufficient to cause damage and yield loss in potato crops in Australia. However, further research is required to determine the impact of the two major nematode species on potatoes and whether they form a synergistic relationship with *V. dahliae*.

Verticillium dahliae has a wide host range and can also survive at low levels on the roots of many crops and weeds without causing disease. Although grasses and other monocots are generally not considered to be good hosts of *Verticillium,* there is evidence that the fungus can systematically colonise barley, wheat and oats. The fungus infects over 50 species of

plants covering 23 families. Common weeds suspected as hosts include Solanaceous weeds (e.g. nightshade), fat hen (*Chenopodium album*), shepherds purse (*Capsella bursa-pastoris*) and dandelions (*Taraxacum* spp.).

Frequent cropping of potatoes maintains high populations of *Verticillium* in soil. However, the effectiveness of rotations in controlling the disease depends on the initial population densities. Many studies overseas have shown that various rotation practices can affect populations of this fungus. However, it has proven difficult to reduce population levels below the thresholds at which economic damage occurs. Microsclerotia are very persistent in soil and some studies suggest that it may take 5 to 10 years to reduce populations to moderate levels using rotations with grain crops. Generally, long rotations with grasses and legumes are least favourable to *Verticillium*.

It is imperative in Australia to avoid rotations that increase populations of this fungus. The spread of potato cropping into the warm, semi-arid regions with sandy soil may see an increase in incidence of damage by *V. dahliae* and associated nematodes.

3.2.8 Nematodes

Nematodes are often overlooked as serious pests of potato production in Australia. Root knot and root lesion nematodes cause problems for seed growers, with outbreaks occurring locally and sporadically, often in potatoes grown after pastures. Damage caused by these nematodes is likely to become more common with the shift of potato production into sandy soils in warm semi-arid regions (e.g. Riverina, Mallee and Murray River areas) since the nematodes prefer coarse textured soils and warm temperatures.

Some of the common nematodes associated with potatoes in Australia are listed in Table 3.1. The best know is the potato cyst nematode (PCN) (*Globodera rostochiensis*), first recorded in Western Australia in 1986 and in Victoria in 1991 (Guy *et al.* 1992; Marshall 1998; Stanton 1986). PCN is a quarantinable pest. It is now effectively eradicated from Western Australia and is restricted to a few infested farms in Victoria. The impact of PCN on seed potato growers is that all fields designated for certified seed potato production must be tested for the nematode prior to planting.

Nematodes are often associated with disease complexes. Root knot nematode infection predisposes plants to infection by *R. solani* (Cetas and Harrison 1963) and *V. dahliae*, the cause of early dying (Martin *et al.* 1982; Rowe *et al.* 1987), the nematode and the pathogens interacting synergistically (Rowe *et al.* 1985). In a study of the incidence and distribution of *Verticillium* and *Pratylenchus* spp., V. *dahliae* was found to be relatively common in Australian potato growing areas, although the nematode *P. crenatus* and *P. neglectus*, predominated. *P. penetrans* was rarely found (Harding and Wicks 2007). The impact of *P. crenatus*, the most commonly detected species, on potato productivity in Australia is not known.

The biology and control of both the root knot and root lesion nematodes are described in detail by Brodie *et al.* (1993). The root knot and root lesion nematodes survive as eggs in soil and in host tissue. Both nematodes have a very wide host range, attacking most major crop plants and a wide range of weed species. Frequent cropping of potatoes and other hosts can rapidly build-up populations of nematodes. Disease control through rotation is difficult. Grasses in rotations have been used to successfully control some species of the root knot nematode. Populations of *Meloidogyne* decline rapidly in the absence of a suitable host. The

effectiveness of rotations on the root knot nematode depends on the particular species of nematode. Control of *Pratylenchus*, however, has proved difficult because of the very wide host range of this nematode. The survival of *Pratylenchus* is favoured particularly by cereal grain crops, such as barley (*Hordeum vulgare*) and rye (*Secale cereale).*

Various species of *Pratylenchus* and *Meloidogyne* have been associated with damage to red clover and white clover in many countries, including Australia (Cook and Yeates 1993). Little is known of nematode problems in fodder and oilseed Brassica (Riggs and Niblack 1993). Ryegrass is generally considered not to be as good a host of *Pratylenchus* spp. than other herbage species (Cook and Yeates 1993).

Table 3.1 Some nematodes associated with potatoes in Australia

Common name	Scientific name	Symptoms
Root lesion nematode	*Pratylenchus penetrans* *P. coffeae* *P. crenatus* *P. neglectus* *P. thornei*	Patches of poor growth in crop - plants are less vigorous, turn yellow and stop growing, small pimples on the tuber skin
Root knot nematode	*Meloidogyne* spp.	Knots or warts on roots and tubers
	Meloidogyne fallax	New species reported in Australia, particularly virulent to potatoes
Potato cyst nematode (PCN)[1]	*Globodera rostochiensis*	Stunting and early senescence

[1] Found only on a small number of quarantined farms in Victoria. Eradicated from WA

Nematode numbers in soil under different treatments were recorded in rotation field trials in South Australia (de Boer 2003a). *Pratylenchus crenatus* was the most common nematode found. Populations of the nematode increased with each consecutive cropping cycle in most treatments at Langhorne Creek with seasonal peaks, but remained relatively constant at Woodside (de Boer 2003a).

In a long-term rotational study in Canada, root lesions and root knot nematodes were more prominent in red clover than in Italian ryegrass stands and this trend continued into the potato crop (Carter *et al.* 2003).

3.2.9 Rotations as part of Integrated Pest Management Systems

Rotation is a critical component of Integrated Pest Management Systems. Any benefit derived from rotations can be rapidly eroded without practising other disease management strategies. The different facets of this system would include:

- **Hygiene protocols** – To minimise the recontamination of fields through diseased seed and contaminated machinery.

- **Resistant cultivars** - Introducing resistant or less susceptible cultivars into rotations has been shown to contribute, not only to a reduction in disease incidence but also to reductions in populations of pathogens in soil. The use of resistant cultivars can also alleviate the need for very long rotations in some cases. For example, in the New Polders in the Netherlands, a yield reduction of 10% in a three-year compared to a

six-year rotation could be reduced to about 3% using a cultivar tolerant to *Verticillium dahliae* and to about 8% using a cultivar resistant to netted scab (Lamers *et al.* 1989). This strategy also applies to crops grown in rotation with potatoes. For example, it would be beneficial to use *Brassica* species with some resistance to the root lesion nematode in a rotation to help minimise nematode populations.

- **Cultural, chemical and biological control** - A range of cultural (nutrition, cultivation, irrigation), chemical and biological management strategies for soil-borne diseases, including green manure crops and organic soil amendments, that prevent disease, reduce inoculum and enhance soil suppressiveness to pathogens.

- **Weed control** - Many so called 'unspecialised' pathogens (e.g. *R. solani, C. coccodes, V. dahliae* and nematodes) have a wide host range and are able to colonise the roots and stems of many different plant species, thereby surviving the period between each potato crop. Weed control, including the control of volunteer potatoes, is essential to the effective use of rotations for disease management.

3.2.10 Conclusions

Australian research on grain cropping systems shows that the pasture phase, particularly legume based pastures, replenishes and accumulates soil nitrogen and restores soil structure after cropping. The pasture phase provides a break from potatoes and reduces the build-up of inoculum that occurs in short rotations. This 'break' period has a greater effect on disease and productivity than the specific plant species grown in the rotation.

The effectiveness of a pasture in reducing populations of plant pathogens below economic thresholds depends on the life-cycle and mode of survival of each particular organism, the choice of crops rotated with potatoes and returns of organic matter and nutrients, which stimulate growth of antagonists and competitors in soil.

Peters *et al.* (2003) summarise the relationship between pathogen and hosts in rotations as follows:

> Rotating crops with plants that are less susceptible to specific pathogens causes a decline in populations of the pathogen due to natural mortality and antagonistic activities of co-existent root zone micro-organisms (Fry 1982; Williams and Schmitthenner 1962). Rotation is most successful in limiting the impact of biotrophic pathogens requiring living host tissues, or those pathogens with low saprophytic capability (Bailey and Duczek 1996). It is least successful in reducing disease caused by pathogens with a wide host range or that produce long-lived survival structures such as sclerotia or oospores (Huisman and Ashworth 1976; Umaerus *et al.* 1989). Crop rotation tends to be a good tool for limiting the increase of soil-borne pathogen populations but tends to be less effective as a curative to reduce pathogen levels that have been allowed to reach high population densities (Fry 1982).

There is scope for manipulating rotations to manage most soil-borne pathogens. However, rotation must be practiced as part of an Integrated Pest Management System. For optimum benefits, rotation should be integrated with hygiene practises on farms, including high quality seed potatoes, the use of resistant or less susceptible cultivars, appropriate cultural, chemical

and biological control strategies that include green manure crops and organic soil amendments, and effective weed control throughout the rotation sequence.

Practices that improve soil health (e.g. green manures, organic amendments and minimum tillage) can potentially enhance the benefits of rotations. Carter and Sanderson (2001) and Peters et al.(2003) conclude from there research that the use of conservation tillage in 3-year rotation systems has the potential to maintain crop productivity, protect the soil resource, and improve soil quality. They argue that soil agro-ecosystems can be modified through rotation and conservation tillage practices to improve disease suppression by enhancing the antibiosis abilities of endophytic and root zone bacteria.

3.2.11 Objectives of the project

There is evidence that a well managed legume based pasture phase provides benefits in terms of soil nitrogen and structure and can reduce the build-up of pathogen populations in soil. Despite the use of 3-5 year rotations for seed production, diseases still cause growers significant problems. The suggests that the pasture maintain, rather than reduce inoculum of some soil-borne pathogens. The objectives of this project were to:

- Evaluate the effects of different component species of pastures, e.g. grass and clover, on disease and yield of potatoes.
- Determine whether, the different pasture species supported pathogens such as *Rhizoctonia solani*
- Use DNA tools to quantify pathogen populations in the different rotations when they became available (PT01019).

The overall aim was to establish some principals with which to better manage the pasture-potato rotation for disease control.

Fodder Brassica were included in the study because previous research had shown that the potato infecting strain of *R. solani* (AG3) could establish non-symptomatic endophytic relationships with Brassica root systems (de Boer 2003a).

4 Technical Report: The implications of pastures on the management of soil-borne diseases of seed potatoes

4.1 A study of the effects of different pasture and fodder species in rotations on disease incidence and severity and yield of potatoes

4.1.1 Introduction

The rotation is the platform on which the farmer manages farm productivity, including pests and diseases, and the economics of the farm enterprise overall. Many seed producers' grow potatoes in rotation with a pasture phase in mixed enterprises involving livestock, or they lease land from other enterprises that feature a pasture phase. The objective of this project was to study the interactions between soil-borne pathogens and specific pasture and fodder species grown in rotation with potatoes in order to establish some principles with which to better manage the rotation for disease control and productivity. More specifically, the study aimed to understand what impact the clover, grass and Brassica components each had on soil-borne pathogens, disease and yield, and whether there is an opportunity to manipulate the pasture towards improved disease management.

A trial was conducted near Ballarat in the Central Highlands Region of Victoria. This region is a major seed and processing potato production area for Victoria. Soils (ferrosol or 'kraznosem') are similar to those of the major potato production areas on the north coast of Tasmania. The diseases Rhizoctonia stem canker (*R. solani*), black scurf (*R. solani*), black dot (*C. coccodes*), silver scurf (*H. solani*) and powdery scab (*S. subterranea)* predominate in this region.

Pastures are usually a mixture of perennial ryegrass (*Lolium perrene* L.) and white clover (*Trifolium repens* L.). Fodder Brassica (*Brassica napus* L.), also referred to as fodder rape, is also commonly grown in this system, usually after the potato crop.

Lucerne (*Medicago sativa* L) is grown for pasture and hay in the drier areas of the Central Highlands, north west of Ballarat. Drier seasonal conditions in the north east over the past ten years have made growing lucerne a more attractive proposition in this part of the highlands, where once this crop was not considered suitable. Additional rotation plots that included lucerne were established adjacent to the main trial pasture plots in the second year of the project. These plots provided the opportunity for observing the establishment and growth of the crop in this district.

4.1.2 Materials and Methods

Experimental treatments and design

Ideally, the type of study undertaken here requires a long-term trial in which each different component species of the pasture (e.g. pasture or fodder legumes and fodder Brassica grown over one, two or three years) are grown in phased rotations so that potato crops that follow the 1, 2 or 3-year break are all grown in the same season. Given the short time frame of this

project, each of these rotations was established at the same time and were, therefore, out of phase.

The trial was established in the Victorian Central Highlands at Clarkes Hill, north east of Ballarat in spring of 2001. The paddock had had a general history of a pasture-potato rotation (*i.e.* 1:4 rotation, one year of potatoes for every three years of pasture). Soil type is a ferrosol, also known as a 'kraznosem' soil (Isbell 1996; McKenzie *et al.* 2004), with a pH of 5.2. Paddock history was pasture prior to one season of carrots, 2 years of pasture, one season of potatoes (2000/01) followed by a fodder Brassica crop.

The trial was a randomized block design replicated four times. Each plot was a 10m long strip 4.96 m wide, allowing for 6 rows of potatoes. Rotational sequences are outlined in Table 4.1. Rotation treatment were: continuous potatoes (1:1); a 1:2 rotation of a pasture or fodder crop with potatoes (potatoes once in two years); and a 1:3 rotation with two years of pasture or fodder crop with potatoes (potatoes once in three years). Rotation crops were: perennial ryegrass (cv. New Tetila and Victoria), fodder Brassica (fodder 'rape') (cv. Maxima), white clover (cv. Waverley) and wheat (cv. Lawson). The potato cultivar Russet Burbank, was planted as cut seed pieces that had been treated with a standard rate of pencycuron fungicide for control of seed-borne black scurf.

Table 4.1 Rotational sequences that compare the effects of 1:2 and 1:3 rotations of pasture and fodder species with potatoes on disease and yield in a field trial at Clarkes Hill, 2001-2005 (The trial site had been cropped to potatoes in the 2000/01 season).

Treatment	Crop frequency	2001-2002	2002-2003	2003-2004	2004-2005[a]
Main trial (4 reps)					
A (PPP)	3:3	potato	Potato	Potato	potato
B (GPG)	1:2	ryegrass	Potato	Ryegrass	potato
C (GGP)	1:3	ryegrass	Ryegrass	Potato	potato
D (CPC)	1:2	clover	Potato	Clover	potato
E (CCP)	1:3	clover	Clover	Potato	potato
F (WCP)	1:3	wheat	Clover	Potato	potato
G (BPB)	1:2	Brassica	Potato	Brassica	potato
Additional plots (3 reps)					
H (LLP)	1:3	-	Lucerne	lucerne	potato
I (LPL)	1:2	-	Lucerne	Potato	potato
J (BaPR)	1:2	-	Barley	Potato	potato

a - Additional season (see Section 4.4)

Additional treatments were added in 2002 as a randomized block design replicated three times and included 1:2 and 1:3 rotations of barley (cv. Parwan) and lucerne (cv. Prime) with potatoes (Table 4.1).

In the 4th season (2004/05) all plots were planted to potatoes. Plots in a 2nd cycle of ryegrass/potato, white clover/potato and fodder Brassica/potato were sampled for disease and yield assessments (Table 4.1) (see Section 4.4).

Details of various crops sown are presented in Table 4.2. In the first year of the trial, the existing fodder Brassica crop was desiccated with the herbicide glyphosate, except for treatment G in which Brassica was left standing, and treatment F in which the Brassica was

slashed in autumn of 2002, plots cultivated and sown to winter wheat. All plots were cultivated prior to sowing the pasture and fodder crops.

Potato crops were irrigated using a solid set irrigation system on the same watering schedule as adjacent commercial crops. Lucerne plots were also irrigated to ensure plant establishment. The potato crop was managed for nutrients, weed control, insect and early blight control as per adjacent commercial crops.

Figure 4.1 Pasture rotation field trial plots at Clarkes Hill in October 2002

Table 4.2 Details of cropping in the pasture rotation trial, Clarkes Hill, 2001/02 -2004/05

(P, potato; G, perennial ryegrass; C, white clover; B, fodder Brassica; W, wheat; Ba, barley; L, lucerne)

Treatment	Crop	Rate of Sowing	Date Sown	Harvested/ Rotation crop ploughed in.
2001-2002				
1:1 P	Potato cv. Russet Burbank		21/12/2001	15/5/2002
1:2 GP	Ryegrass cv. New Tetila and Victoria	8 kg / Ha	12/12/2002 and 18/4/2002	18/9/2002
1:2 CP	White clover cv. Waverley	5 kg / Ha	12/12/2002 and 18/4/2002	18/9/2002
1:2 BP	Fodder Brassica cv. Maxima	3.5 kg / Ha	26/10/2001	18/9/2002
1:3 GGP	Ryegrass cv. New Tetila and Victoria	8 kg / Ha	12/12/2002 and 18/4/2002	Left for next year
1:3 CCP	White clover cv. Waverley	5 kg / Ha	12/12/2002 and 18/4/2002	Left for next year
1:3 WCP	Wheat cv. Lawson	120 kg /Ha	18/4/2002	January 2003
2002-2003				
1:2 PP	Potato		29/11/2002	23/5/2003
1:2 GP	Potato		29/11/2002	23/5/2003
1:2 CP	Potato		29/11/2002	23/5/2003
1:2 BP	Potato		29/11/2002	23/5/2003
1:3 GGP	Ryegrass	Left from last year	Left from last year	Early Nov 2003
1:3 CCP	White clover	Left from last year	Left from last year	Early Nov 2003
1:3 WCP	White clover		March 2003	Early Nov 2003
1:2 LP	Lucerne cv. Prime	10 kg / Ha	7/11/2002 and 21/3/2003	Early Nov 2003
1:3 LLP	Lucerne	10 kg / Ha	7/11/2002 and 21/3/2003	Left for next year
1:2 BaP	Barley cv. Parwan	120 kg / Ha	7/11/2002	Early Nov 2003
2003-2004				
3:3 PPP	Potato		7/12/2003	13 May 2004
1:2 GP	Ryegrass	8 kg/ Ha	11/9/2003	
1:2 CP	White clover	5 kg/ Ha	11/9/2003	
1:2 BP	Fodder Brassica	3.5 kg/ Ha	11/9/2003	
1:3 GGP	Potato		7/12/2003	13 May 2004
1:3 CCP	Potato		7/12/2003	13 May 2004
1:3 WCP	Potato		7/12/2003	13 May 2004
1:2 LP	Potato		7/12/2003	13 May 2004
1:3 LLP	Lucerne	10 kg/ Ha	11/9/2003	
1:2 BaP	Potato		7/12/2003	13 May 2004
2004-2005				
All treatments	Potato		Early Dec 2004	Mid May 2004

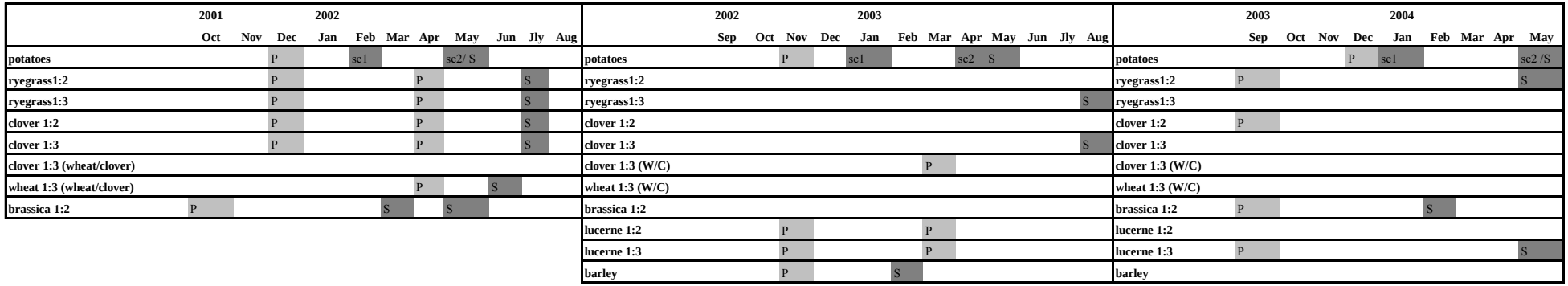

Figure 4.2 Summary of planting and sampling dates for potato, pasture, fodder and cereal crops in a field trial, Clarkes Hill

P = planted, S =sampled/harvested, sc1 = stem canker assessment number 1, sc2 = stem canker assessment number 2.

4.1.3 Plant growth, disease and yield assessments in potatoes

Pre harvest growth and Rhizoctonia disease assessment.

At approximately 6-8 weeks after planting, the total number of plants in the 2nd and 5th row of each plot was counted to determine plant emergence (% plants emerged). In the 2001/02 and 2002/03 seasons, each plant in the two rows was scored for the degree of stunting, relative to the tallest plants, on a scale of 0-4 where 0 = no stunting, 1 = slight, 2 = moderate, 3 = severe and 4 = plant not emerged. In the 2003/04 season, each plant in the two rows was scored on a scale of 0-3 where plants with no stunting, slight, moderate and severe stunting were assigned ratings of 3, 2, 1 and 0, respectively.

Potato plants were assessed at 6-8 weeks after planting for the incidence and severity of stem and stolon canker attributed to *R. solani* (cortical cankers on below ground stems and stolons). Four plants were sampled from the 2nd and 5th rows of the six-row plots (total of 8 plants/plot). Plants were rated as 0 = no symptoms of Rhizoctonia damage, 1 = less than 25%, 2 = 25-50%, 3 = greater than 50% and 4 = all sprouts and stems pruned (no emergence). When the crop had senesced, the percentage of plants in the four middle rows of each plot with symptoms of Rhizoctonia damage (late maturing and not yet fully senesced, shortened internodes and aerial tubers in the leaf axils) was recorded.

Yield and disease assessment at harvest

Potatoes from 3rd and 4th (middle) rows of each six-row plot were harvested using a two-row harvester. A sub sample of 50 tubers was taken at random from the harvested rows in each plot for disease assessments. The remaining tubers were sorted into weight categories of chats < 35 g, small 35 -110 g, medium 110 - 250 g, large 250 - 350 g, oversize > 350g and misshapen, and the numbers and weights for each category recorded.

The 50 tubers from each plot were washed and each tuber visually inspected for skin diseases and rots. The severity of silver scurf, black dot, powdery scab and common scab on each tuber was scored on a scale of 0-4, where ratings of 0 = no disease, 1 = less than 2%, 2 = 10% and 3 = 10-25% and 4 = greater than 25% of the tuber surface covered with a disease symptom. The severity of the black scurf symptom (*R. solani*) was rated on a scale of 0-3 where tubers with no sclerotia, light, moderate or heavy coverage of sclerotia were rated 0, 1, 2, and 3, respectively. Disease incidence was recorded as the percentage of tubers in each sample with disease symptoms.

Statistical analysis

Data were analysed using ANOVA, GenStat for Windows 6th Edition (Lawes Agricultural Trust, IACR-Rothamsted) and differences between treatments determined using Fisher's method of least significant difference (lsd) at the 5% significance level.

4.1.4 Results

Growth and emergence of potato plants

In the first season with rotations (2002/03), the continuous potato treatments had higher emergence but a greater incidence of smaller plants than all other treatments. Generally there were no differences in emergence and relative growth of plants in 2003/04 (Table 4.3).

Table 4.3 Emergence and growth of potatoes in the pasture field trial, Clarkes Hill (P, potato; G, perennial ryegrass; C, white clover; B, fodder Brassica; W, wheat; Ba, barley; L, lucerne)

Rotation	Emergence	Crop Growth (relative plant size)	
	(%)	Incidence (%)	Severity (0-4) (0=healthy)
2001-2002			
1:1 P	84	35	0.5
2002-2003			
2:2 PP	96 [b]	31 [b]	0.4 [b]
1:2 GP	88 [a]	20 [a]	0.2 [a]
1:2 CP	84 [a]	22 [ab]	0.2 [ab]
1:2 BP	86 [a]	27 [ab]	0.3 [ab]
F-test.	**0.011**	**0.110**	**0.117**
lsd (P=0.05)	**6.4**	**9.74**	**0.1281**
2003-2004			**(Severity 0-3, 3=healthy)**
3:3 PPP	94 [a]	-	2.6 [a]
1:3 GGP	92 [a]	-	2.6 [a]
1:3 CCP	95 [a]	-	2.7 [a]
1:3 WCP	95 [a]	-	2.7 [a]
F-test	**0.639**	**-**	**0.843**
lsd (P=0.05)	**5.618**	**-**	**0.2863**
2003-2004			
1:2 LP	92 [a]	-	2.6 [a]
1:2 BaP	91 [a]	-	2.4 [a]
F-test	**0.914**	**-**	**0.596**
lsd (P=0.05)	**38.3**		**1.70**

Numbers followed by the same letter do not differ significantly from each other

Disease

Disease results are presented in Table 4.4. Rhizoctonia stem canker (*R. solani*), and the tuber diseases black scurf (*R. solani*), black dot (*C. coccodes*), silver scurf (*H. solani*) and powdery scab (*S. subterranea)* were common each season.

The incidence and severity of black dot and silver scurf were not significantly affected by rotation treatments in 2002/04 and 2003/04.

2001/02

In the first year of the trial, 100% of plants had symptoms of Rhizoctonia damage on the stems and stolons with an average severity rating of 3.2 (on a scale of 0-5), although there were no apparent 'Rhizoctonia' patches (patches of stunted plants) in the plots during early crop growth. An average of 17% of mature plants had aerial tubers. At harvest, an average of 63% of tubers had symptoms of black scurf, 2% silver scurf and 62% black dot and 56% powdery scab.

2002/03

The incidence and severity of Rhizoctonia canker did not vary significantly between treatments ($P>0.05$), although there was a trend of more plants with symptoms in the potato-potato treatment than in the other treatments, and more plants with aerial tubers in the rotation treatments (1:2) compared with continuous potatoes. The incidence of black scurf was higher in the grass/potato and Brassica/potato treatments than the potato/potato and clover/potato treatments.

The incidence and severity of tubers with powdery scab was higher ($P\leq0.05$) (81% tubers affected) in the potato/potato treatment than in the grass, clover and Brassica/potato treatments. However, the incidence and severity of powdery scab did not differ significantly between the grass, clover and Brassica treatments.

2003/2004

The incidence and severity of Rhizoctonia canker was significantly less ($P\leq0.05$) in the continuous potato treatment than in the grass/grass, clover/clover and wheat/clover treatments. The incidence of plants with aerial tubers was higher ($P\leq0.05$) in the clover rotation than in continuous potatoes but did not differ significantly between the grass, clover and wheat/clover rotation treatments.

The severity of powdery scab was less ($P\leq0.05$) in the grass/potato treatments than the continuous potatoes, clover/potato and wheat/clover/potato treatments, and disease incidence followed a similar trend.

In the additional plots, plants with Rhizoctonia canker symptoms were more common ($P\leq0.05$) in lucerne-potato treatment than in the barley-potato treatment. In contrast, tubers with black scurf were more common ($P\leq0.05$) in the barley/potato treatment than in the lucerne/potato treatment. There were no significant differences in incidence and severity of powdery scab between these treatments.

Yield

Yield data are presented in Table 4.5. In both 2002/03 and 2003/04, the total yield was less ($P\leq0.05$) in the continuous potato treatment (PP and PPP, respectively) than where potatoes were grown in rotation. In 2002/03, marketable yields in the potato-potato treatment were significantly less ($P\leq0.05$) than in the grass/potato and Brassica/potato treatments but did not differ significantly from the clover/pasture treatment. The average number and yield of potatoes varied significantly between treatments in some size categories ($P\leq0.05$), although there were no consistent patterns linking treatment and yield. A consistent trend in numbers and yield in different size categories was a greater number of smaller tubers and a reduced number of larger tubers in the continuous potato treatments compared to the grass, clover and

Brassica rotations in 2002/03 and 2003/04 (Table 4.5, Figure 4.3, Figure 4.4). There were no significant differences in yield of potatoes when comparing the lucerne/potato and barley/potato rotation in 2003/04.

Table 4.4 Effects of different rotations of pasture and fodder species with potatoes on Rhizoctonia stem canker and on disease incidence and severity of progeny tubers (black scurf, black dot, silver scurf and powdery scab) in a field trial, Clarkes Hill Victoria

(P, potato; G, perennial ryegrass; C, white clover; B, fodder Brassica; W, wheat; Ba, barley; L, lucerne)

Rotation	Rhizoctonia damage		Tuber diseases								
	Stem canker		Plants with aerial tubers	Black scurf		Black dot		Silver scurf		Powdery scab	
	% plants affected	Severity (0-5)	% plants affected	% tubers affected	Severity (0-5)	% tubers affected	Severity (0-5)	% tubers affected	Severity (0-5)	% tubers affected	Severity (0-5)
2001/2002											
1:1 P	100.0	3.2	17.3	63.0	1.0	62.0	0.7	2.0	0.03	55.5	0.6
2002/2003											
1:1 PP	81.3 [a]	1.7 [b]	22.0[a]	58.0 [a]	1.1 [a]	93.5 [a]	2.7 [a]	44.5 [a]	0.7 [a]	81.0 [b]	1.2 [b]
1:2 GP	67.9 [a]	1.3 [ab]	37.5 [ab]	73.0 [bc]	1.8 [b]	92.0[a]	2.3 [a]	26.0 [a]	0.4 [a]	40.0 [a]	0.5 [a]
1:2 CP	49.5 [a]	0.8 [a]	49.6 [b]	62.0 [ab]	1.5 [ab]	91.5 [a]	2.4 [a]	51.0 [a]	0.8 [a]	52.0 [a]	0.6 [a]
1:2 BP	57.6 [a]	0.9 [ab]	41.7 [b]	79.5 [c]	1.8 [b]	90.5 [a]	2.5 [a]	39.5 [a]	0.6 [a]	47.5 [a]	0.5 [a]
F-test	ns	ns	0.021	0.026	ns	ns	ns	ns	ns	0.004	0.001
lsd (P=0.05)	32.44	0.879	15.94	14.09	0.69	4.95	0.52	27.56	0.57	18.73	0.27
2003-2004											
1:1PPP	25.0 [a]	0.4 [a]	6.8 [a]	66.5 [a]	1.0 [a]	84.0 [a]	1.2 [a]	14.5 [a]	0.2 [a]	87.0 [b]	1.1 [b]
1:3 GGP	54.9 [b]	1.4 [b]	12.5 [ab]	72.5 [a]	1.2 [a]	86.5 [a]	1.2 [a]	14.0 [a]	0.2 [a]	75.5 [a]	0.8 [a]
1:3 CCP	59.4 [b]	1.2 [b]	19.3 [b]	81.0 [a]	1.4 [a]	78.5 [a]	1.0 [a]	9.0 [a]	0.1 [a]	81.5 [ab]	1.0 [b]
1:3 WCP	58.5 [b]	1.1 [b]	11.3 [ab]	86.5 [a]	1.4 [a]	82.0 [a]	1.0 [a]	15.5 [a]	0.2 [a]	84.5 [b]	1.0 [b]
F-test	0.028	0.026	0.06	ns	ns	ns	ns	ns	ns	0.05	0.025
lsd (P=0.05)	23.85	0.6318	8.74	25.67	0.58	14.59	0.27	8.78	0.08	8.06	0.16
2003-2004											
1:2 LP	43.5 [b]	0.7 [a]	12.2 [a]	36.7 [a]	0.4 [a]	73.3 [a]	0.9 [a]	29.3 [a]	0.3 [a]	47.3 [a]	0.6 [a]
1:2 BaP	13.7 [a]	0.2 [a]	14.0 [a]	44.0 [b]	0.7 [a]	64.7 [a]	0.8 [a]	12.0 [a]	0.1 [a]	53.3 [a]	0.6 [a]
F-test	0.046	ns	ns	0.032	ns	Ns	ns	ns	0.098	ns	ns
lsd (P=0.05)	28.52	0.6732	8.62	5.74	0.90	20.08	0.32	28.25	0.26	99.2	1.16

ns – not significant at P≤0.05. Numbers followed by the same letters do not differ significantly from each other.

Table 4.5 Effects of different rotations of pasture and fodder species with potatoes on total and marketable yields and on the number and yield of tubers in the different size categories in a field trial, Clarkes Hill Victoria

(P, potato; G, perennial ryegrass; C, white clover; B, fodder Brassica; W, wheat; Ba, barley; L lucerne)
(Yields are in t/ha or kg/plot)

Rotation	Total yield (t/ha)	Marketable yield (t/ha)	<35 g		35 - 110 g		110 -250 g		250 - 350 g		> 350 g		Misshapen	
			Number	Wt (kg)	Number	Wt (kg)	Number	Wt (kg)	Number	Wt (kg)	Number	Wt (kg)	Number	Wt (kg)
2001-2002														
1:1 P potato	29.79	26.29			194.75	13.37	151.00	24.65	19.75	5.80	9.25	4.33		
2002-2003														
1:1 PP	37.31 [a]	31.86 [a]	67.25 [b]	1.73 [b]	236.25 [b]	16.60 [b]	187.00 [a]	28.75 [a]	22.50 [a]	6.59 [a]	3.50 [a]	1.46 [a]	29.75 [a]	6.10 [a]
1:2 GP	49.93 [b]	40.75 [b]	47.75 [a]	1.24 [ab]	199.50 [ab]	14.45 [ab]	241.00 [b]	38.30 [b]	47.50 [b]	13.68 [b]	14.00 [b]	6.21 [b]	31.00 [a]	7.51 [a]
1:2 CP	46.04 [b]	36.27 [ab]	29.75 [a]	0.76 [a]	152.75 [a]	10.94 [a]	196.00 [a]	31.74 [a]	57.75 [c]	16.44 [c]	21.25 [bc]	8.95 [bc]	25.25 [a]	6.22 [a]
1:2 BP	51.88 [b]	40.82 [b]	34.50 [a]	0.89 [ab]	158.00 [a]	12.03 [a]	249.50 [b]	40.34 [b]	47.75 [b]	14.17 [b]	22.75 [c]	10.28 [c]	28.00 [a]	6.86 [a]
F-test	0.002	0.022	0.099	ns	0.023	ns	0.006	0.005	<0.001	<0.001	0.002	<0.001	ns	ns
lsd (P=0.05)	6.159	5.934	31.96	0.925	54.80	4.408	35.4	5.809	9.36	2.159	8.35	3.271	22.18	5.128
2003-2004														
1:1 PPP	23.8 [a]	21.0 [a]	65.50 [a]	1.47 [a]	203.00 [a]	15.12 [a]	120.00 [a]	18.13 [a]	5.75 [a]	1.69 [a]	2.00 [a]	0.86 [a]	9.50 [a]	1.09 [a]
1:3 GGP	52.24 [b]	41.54 [b]	59.00 [a]	1.12 [a]	210.50 [a]	15.40 [a]	258.25 [b]	44.76 [b]	31.00 [b]	9.08 [b]	16.25 [b]	7.26 [b]	60.75 [b]	9.46 [b]
1:3 CCP	52.73 [b]	42.19 [b]	60.50 [a]	1.13 [a]	204.25 [a]	15.42 [a]	271.50 [b]	45.71 [b]	30.75 [b]	9.19 [b]	17.50 [b]	8.06 [b]	52.75 [ab]	8.37 [b]
1:3 WCP	48.43 [b]	40.33 [b]	67.25 [a]	1.30 [a]	268.75 [a]	20.00 [a]	242.50 [b]	39.85 [b]	25.25 [b]	7.37 [b]	10.75 [b]	4.87 [b]	53.25 [ab]	7.33 [ab]
F-test	< 0.001	0.008	ns	ns	ns	ns	0.023	0.002	0.007	0.005	0.002	0.003	ns	0.098
lsd (P=0.05)	10.30	11.92	29.04	0.6109	93.8	7.11	97.3	12.15	13.56	3.825	6.81	3.267	46.71	7.13
2003-2004														
1:2 LP	45.75 [a]	38.69 [a]	24.17 [a]	0.56 [a]	202.33 [a]	15.86 [a]	230.67 [a]	40.76 [a]	25.83 [a]	7.88 [a]	16.83 [a]	7.79 [a]	12.50 [a]	3.41 [a]
1:2 BaP	36.02 [a]	31.98 [a]	39.17 [a]	0.84 [a]	188.67 [a]	13.79 [a]	197.00 [a]	33.14 [a]	21.83 [a]	6.37 [a]	8.83 [a]	3.80 [a]	14.17 [a]	2.10 [a]
F-test	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
lsd (P=0.05)	17.41	20.0	25.94	0.6995	151.2	10.76	173.8	24.48	22.08	6.194	15.11	7.239	29.95	7.155

ns – not significant at P≤0.05. Numbers followed by the same letters do not differ significantly from each other

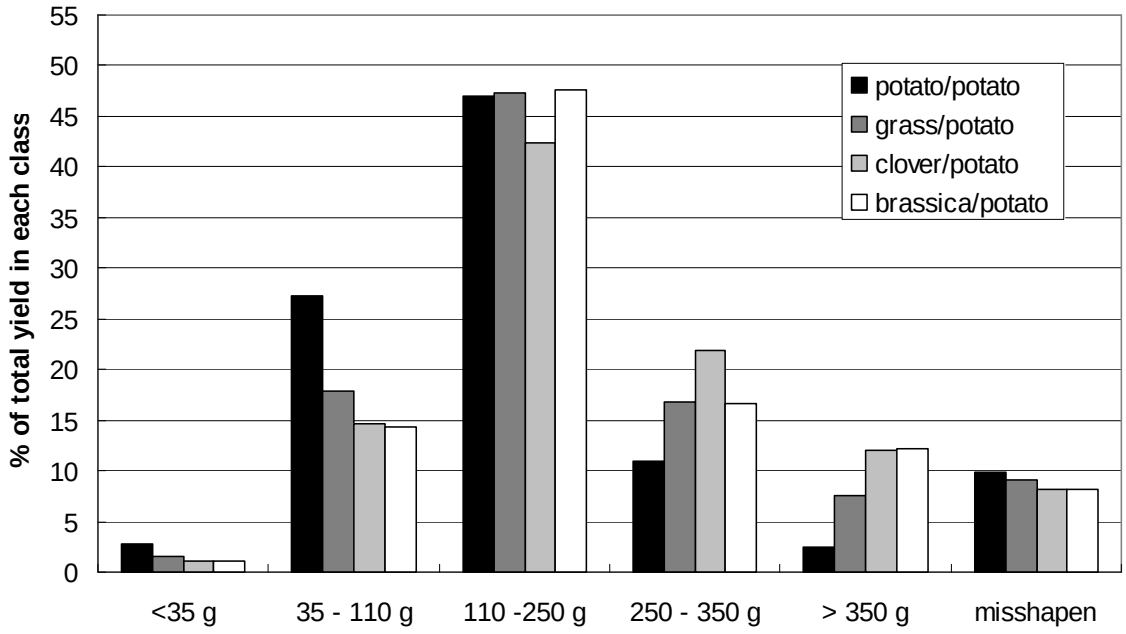

Figure 4.3 Effects of different rotations of pasture and fodder species with potatoes on the proportion of tubers by weight in the different size categories in a field trial, Clarkes Hill Victoria, 2002/03.

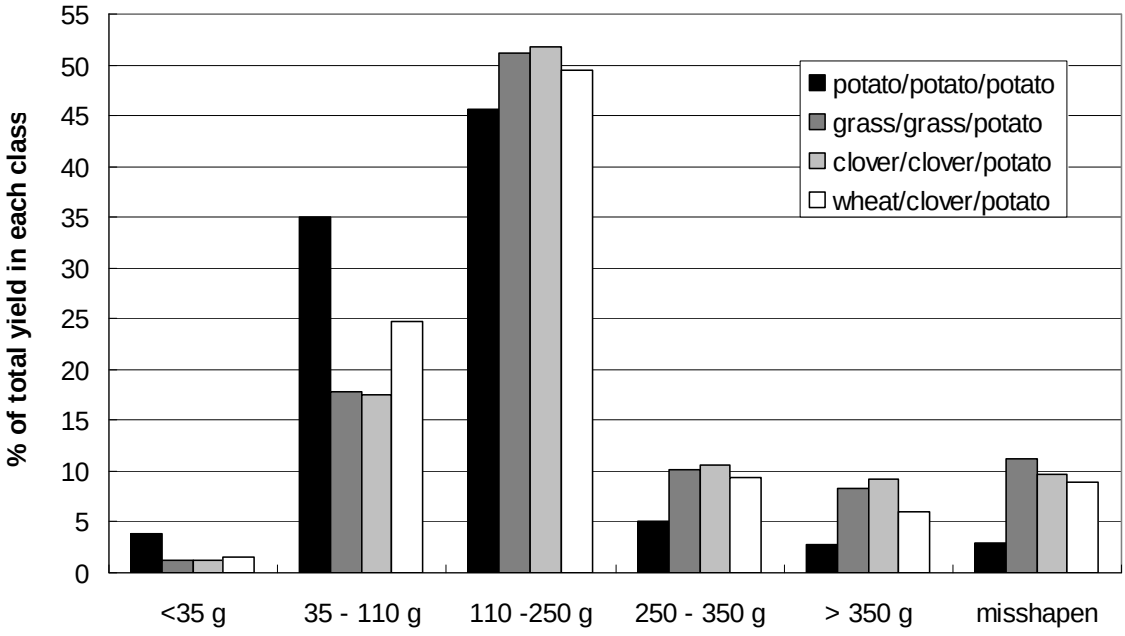

Figure 4.4 Effects of different rotations of pasture and fodder species with potatoes on the proportion of tubers by weight in the different size categories in a field trial, Clarkes Hill Victoria, 2003/04.

4.2 *Rhizoctonia solani* on the root systems of pasture and fodder crops in different rotations

4.2.1 Introduction

Rhizoctonia solani is a versatile soil-inhabiting fungus adapted to survive under a diverse range of conditions (Anderson 1982; Ogoshi 1987). The fungus survives as sclerotia or thick walled hyphae in plant debris and in soil (Papavizas *et al.* 1975). Under favourable conditions the fungus grows actively in soil, colonising the stems, stolons and roots of potatoes. Although the fungus is pathogenic on its preferred host, it can also colonise the root systems of 'non-hosts' (de Boer 2003a). Because of these characteristics, the population and activity of this fungus can be affected by the various pasture species and other crops grown in rotation with potatoes.

In tandem with the rotation trial at Clarkes Hill, a study was conducted of *R. solani* in the various pasture and fodder crops in the pasture trial (Table 4.1) to investigate whether the fungus predominated in a particular crop or rotation.

4.2.2 Materials and Methods

Plants were sampled from the pasture/fodder and cereal plots once they had reached maturity (generally in winter months for clover, ryegrass and lucerne and autumn months for Brassica). Eight individual plants of the Brassica, lucerne and clover, or ryegrass and cereal plants within 6, 10 cm diameters areas, were sampled from each replicate plot in a W or zigzag pattern across the plot. Brassica plots were sampled before mowing and again at 2 months after mowing (simulated grazing) to test the hypothesis that grazing stimulates colonisation of roots by *R. solani*.

Samples were lightly washed to remove soil. Specimens were examined for typical symptoms of damage on crowns, tap roots and fine roots and for the presence of melanised fungal hyphae and sclerotia.

Fragments of diseased Brassica and clover tissue or sclerotia were washed and plated onto 2% water agar. If *R. solani* was present after 12-24 hrs, it was transferred onto potato dextrose agar (PDA). Cultures were maintained and stored on 10% PDA at 4°C.

The finer roots of clover and grass were plated out for evidence of colonisation by *R. solani*. For grass roots two sub samples of 28 random, 5-10mm segments of roots were taken from each sample and plated as described previously. For clover, 14 pieces were plated from each of 8 plants per plot. The roots of wheat and barley were similarly plated but root systems were not examined microscopically.

Multinucleate *R. solani* isolates, characteristically associated with disease in potato, clover and Brassica, were distinguished from binucleate isolates using the method of Bandoni (1979). Isolates were not tested for AG group.

4.2.3 Results

Table 4.6 shows three seasons of data of the association of *R. solani* with various pasture and fodder plants. There was evidence of *R. solani* on the roots of clover, ryegrass, fodder Brassica and lucerne.

Sclerotia were found on the roots of 6.3 to 15.6% of clover plants over the seasons sampled, and 7.5 to 30% of Brassica plants (Table 4.6). Sclerotia were not observed on the roots of ryegrass or lucerne. Lesions consistent with infection by *R. solani* were evident on the roots of clover plants. Melanised hyphae were found on the roots systems of clover, Brassica, ryegrass and lucerne. Mown Brassica plants had no less sclerotia on the roots but a greater incidence of *R. solani* hyphae, compared with non-moved Brassica.

Over the 3 year period, 59 isolates were collected from Brassica, 20 from clover and 9 from grass. Isolates from sclerotia and lesions on Brassica and clover roots were multinucleate *R. solani* with characteristics typical of those associated with these crops. Multinucleate isolates from grass were more typical of those affecting cereals. The fungus was not isolated from the roots of wheat or barley or lucerne. Binucleate *R. solani* were occasionally isolated from the roots of ryegrass.

Table 4.6 Incidence of pasture and fodder plants from different rotations with lesions or epiphytic survival structures of *Rhizoctonia solani* (crop sampled in bold)

Year	Pasture Rotation	% plants with sclerotia	% plants with lesions	% plants with hyphae
2001-02	E **clover**/clover	6.3	12.5	28.1
2001-02	D **clover**/potato	15.6	6.3	46.9
2002-03	E clover/**clover**	6.3	46.9	40.6
2003-04	D clover/potato/**clover**	ns	ns	ns
2001-02	C **ryegrass**/ryegrass	0	0	29.2
2001-02	B **ryegrass**/potato	0	0	33.3
2002-03	C ryegrass/**ryegrass**	0	0	33.3
2003-04	B ryegrass/potato/**ryegrass**	0	0	33.3
2001-02	**G Brassica**/potato (before mowing)	25.0	na	45
2001-02	**F Brassica**/wheat/clover (cover before wheat sown)	20.0	na	47.5
2001-02	**G Brassica** (after mowing)	30.0	na	75.0
2003-04	G Brassica/potato/**Brassica** (before mowing)	7.5	5	32.5
2003-04	H lucerne/**lucerne**	0	0	33.3

ns – not sampled due to poor condition of plots (drought conditions)
na – not assessed

4.3 Quantification of pathogens from soil in pasture/fodder rotations

Soil samples were taken from field trial plots each season prior to planting potatoes and stored at 4°C with the objective of quantifying pathogen populations through bioassays or DNA analysis when techniques were refined.

4.3.1 *Rhizoctonia solani* (Rhizoctonia stem canker and black scurf)

Quantifying *R. solani* inoculum from soil in different pasture treatments - beet seed bioassay

Materials and Methods

A 'red beet seed assay' method (Kyritsis 2003; Papavizas *et al.* 1975) was used to measure relative quantities of inoculum of *R. solani* in field plots.

Soil samples were taken from each replicate plot of field trial treatments A (1:1 potatoes), C (1:3 ryegrass/ryegrass/potato), E (1:3 clover/clover/potato) and F (1:3 wheat/clover/potato), prior to sowing potatoes in the spring of 2003. Ten trowels of soil were taken from each plot to a depth of 15 cm in a W or zigzag pattern across the plot and combined to make a bulk sample of approximately 1kg per plot. The soil was then air dried at room temperature and stored at 4°C. Sub-samples of 250 g from each plot were passed through a 2mm sieve to remove coarse debris. Soil moisture was then adjusted to 20% (w:w) with sterile distilled water and the sample sub-divided into four 9cm Petri dishes, sealed with Parafilm and incubated at 25° for 64 hours.

Red beet seeds (*Beta vulgaris* L.) (cv. "Detroit Dark Red") were soaked overnight in sterile distilled water, autoclaved at 121°C for one hour on each of three consecutive days and air dried in a laminar flow cabinet. Ten sterilised beet seeds were placed in the soil evenly distributed in each dish (*i.e.* 40 seeds/replicate/treatment). Dishes were then re-sealed and incubated at 25°C for 48 hours, after which the soil from each dish was washed through a 2 mm sieve to retrieve the beet seeds.

Beet seeds were washed in running tap water for five minutes, blotted dry, plated onto a modified Ko and Hora's media (Ceresini 1999; Ko and Hora 1971) and incubated at 25°C. Plates were examined after 24 and 48 hours for evidence of *R. solani*. The percentage of seeds colonised by *R. solani* was recorded.

Results

Average seed colonisation rates varied from 3.9 to 15.9% from soil from the different pasture treatments (Figure 4.5). Significantly more ($P \leq 0.05$) seeds were colonised in soil from clover/clover/potato treatments (15.9%) than from the ryegrass/ryegrass/potato, wheat/clover/potato and potato/potato/potato treatments (6.3%, 4.5% and 3.9%, respectively).

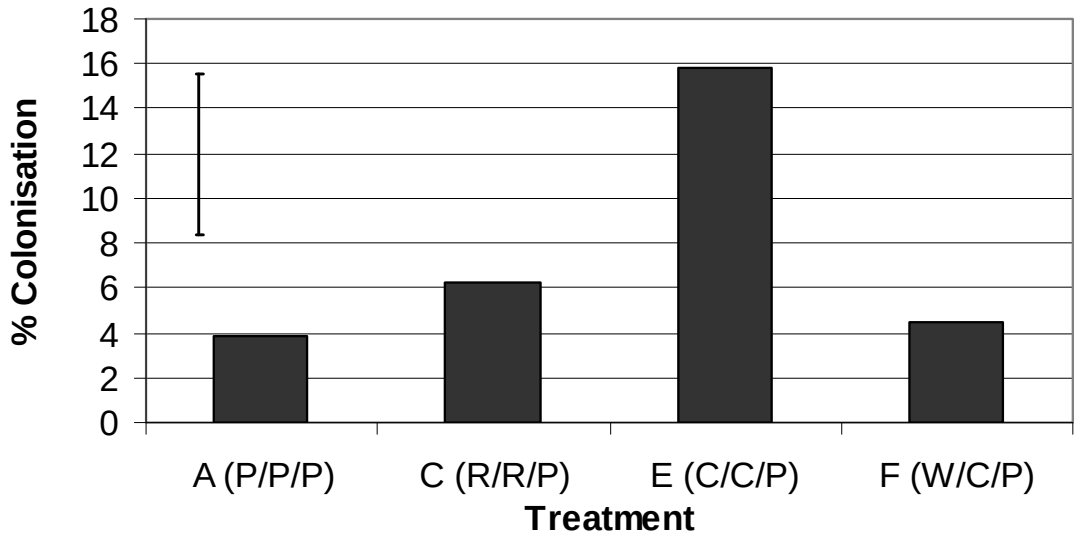

Figure 4.5 Colonisation of beet seeds by *Rhizoctonia solani* in soil taken from different rotation treatments (Soil sampled prior to sowing potatoes in treatments in 2003 A, potato/potato/potato; C, ryegrass/ryegrass/potato; E, clover/clover/potato; F, wheat/clover/potato)(Bar represents the lsd at P=0.05).

Determining anastomosis grouping (AGs) of *R. solani* isolates from a beet seed bioassay using a PCR technique

The results of the beet seed bioassay in Section 4.3.1 indicated a relatively high rate of colonisation in the clover/clover/potato treatments. The beet seed bioassay was repeated on soil from this treatment to collect isolates of *R. solani* for AG determination using a PCR technique.

Materials and Methods

A beet seed bioassay was conducted on a soil sample collected from four replicate trial plots, Treatment E, clover/clover/potato (Table 4.1), sampled just prior to sowing potatoes the spring of 2003. A sub-sample of soil from each of the four plots was combined to give one bulked sample and the bioassay conducted on three sub samples. Soils were tested undiluted or diluted 25% and 50% with sterile sand. The bioassay was conducted as described previously in Section 4.3.1.

Isolates of *Rhizoctonia solani* taken from beet seeds were grown in V8 broth for 2 weeks, after which mycelium was removed from the broth and freeze dried. DNA was extracted from the mycelia of the different isolates and subjected to PCR primers specific for *R. solani* AG3 using the method of Lees *et al.* (2002).

Results

An average of 22% of seeds exposed to undiluted soil were colonised by *R. solani*. A total 53 isolates of *R. solani* were obtained from beet seed assays from undiluted and diluted soils and 24 of these isolates were tested using PCR primers, all of which tested positive for *R. solani* AG3, the potato infecting strain of the fungus.

4.4 Effects of 2-year pasture/fodder rotations on pathogen DNA, disease and yield of a second consecutive crop of potatoes

4.4.1 Introduction

Two different rotation sequences of one (e.g. clover/potato/clover) and two year rotations (e.g. clover/clover/potato) were established in the field trial described in Table 4.1. All plots in the trial were sown to potatoes in the 2004/05 season to investigate the effects of rotations on a second consecutive potato crop in the one year rotations of clover/potato, ryegrass/potato and potato/potato.

4.4.2 Materials and Methods

The field trial plots were maintained over the autumn and winter months of 2004 after the potato harvests in May. All plots were planted to potatoes (cut seed cv. Russet Burbank) in December 2005. Crop irrigation was with a solid set irritation system on the same schedule as commercial crops in adjacent fields. Crop management (fertiliser, weed management, insect and blight control) was managed by the potato grower, as per adjacent commercial crops.

Prior to sowing potatoes, 40 soil cores (depth 15 cm) were taken in a 'W' pattern from each replicate plot. Bulked samples from each plot were sent to the South Australian Research and Development Institute's (SARDI) Root Disease Testing Service Laboratory for Q-PCR analysis of *R. solani* AG2-1 and AG3 and *S. subterranea* populations in soil.

Rhizoctonia stem canker assessments were conducted as described in Section 4.1.3. Potatoes were harvested from the middle two rows of each plot in May 2005 and total plot yield recorded. The incidence of disease on tubers was assessed on sub samples taken at harvest as described in Section 4.1.3. Results were analysed by ANOVA (Section 4.1.3).

4.4.3 Results

The results of soil DNA tests, incidence and severity of Rhizoctonia stem canker, the incidence of tuber diseases and total yield are presented in Table 4.7.

There was significantly more ($P \leq 0.05$) DNA of *R. solani* AG2-1 in soil from the Brassica/potato rotation than in the clover/potato, ryegrass/potato and potato/potato rotations. *R. solani* AG3 DNA was more common ($P \leq 0.1$) in the continuous potato treatment than in the ryegrass, clover and Brassica rotations. DNA of *S. subterranea* was more common ($P \leq 0.05$) in the continuous potato treatments than in the ryegrass, clover and Brassica rotations.

An average of 44% of plants in the continuous potato plots were affected with Rhizoctonia stem canker. However, there were no significant differences in the incidence and severity of stem canker in potatoes between the different rotation treatments. Similarly, the incidence of tubers with black scurf, black dot and powdery scab (43, 34, 52%, respectively, in the continuous potato plots) were not affected significantly by rotation treatments.

The average total yield was 37 t/ha. There were no significant differences ($P>0.05$) in total yields when comparing the different rotations, although the total yield from the continuous potato treatment tended to be less than rotations (32 compared with 38, 40 and 38 t/ha for grass, clover and Brassica rotations, respectively). In addition, it was observed that the yield of marketable potatoes in the plots of the continuous potato treatments was relatively low compared with the ryegrass, clover and Brassica rotations (data not collected). Tubers were generally small and the plots were affected with a high incidence of soft rots.

Table 4.7 Effects of a four year rotation sequence of pasture and fodder species with potatoes on pathogen soil DNA levels for pathogens, yield, incidence of Rhizoctonia stem canker and incidence of tuber diseases on progeny tubers (black scurf, black dot and powdery scab) in a field trial, Clarkes Hill Victoria, 2004/05.

(P, potato; G, perennial ryegrass; C, white clover; B, Brassica)

Rotation Treatment & sequence	DNA in soil (pg DNA/g soil) *Rhizoctonia. solani*		*Spongospora subterranea*	Yield (t/ha)	Rhizoctonia damage Stem canker		Tuber diseases Black scurf	Black dot	Powdery scab
	AG 2-1	AG 3			% plants affected	Severity (0-5)	% tubers affected	% tubers affected	% tubers affected
2004/2005									
A (PPPP)	38 [a]	215 [a]	19,993 [a]	32.1	40.6	0.97	42.5	33.5	52.2
B (GPGP)	141 [a]	25 [b]	9,439 [b]	38.2	43.8	0.78	26.0	32.5	41.5
D (CPCP)	564 [a]	11 [b]	8,012 [b]	40.4	22.8	0.49	31.5	20.0	44.5
G (BPBP)	1,366 [b]	36 [b]	11,436 [b]	38.3	32.2	0.46	33.0	25.5	50.0
F-test	0.007	0.052	<0.001	ns	ns	ns	ns	ns	ns
lsd (P=0.05)	690.4	158.0	2706.5	6.114	30.58	0.5112	30.94	15.46	20.87

ns – not significant at P≤0.05

Numbers followed by the same letter to not differ significantly to each other

4.5 General Discussion

Diseases in the trial were common, indicating a relatively high disease pressure at this site consistent with a history of potato cropping. The disease profiles are typical of those found in the Central Highlands and other potato production areas.

Total and marketable yield was significantly less in potatoes cropped after one and two years of a pasture or fodder crop than in continuously cropped potatoes This shows a clear benefit of a break in the potato cropping cycle. However, this effect could not be attributed directly to disease incidence and severity, which, with a few exceptions, did not differ significantly between continuously cropped potatoes and potatoes in the rotations. One of the contributing factors to yield decline in continuous potatoes may have been deterioration of soil structure, which was evident (not measured) as compaction, water logging and higher incidence of tuber rotting.

There was evidence from the 2nd cycle of the two-year rotation, of lower inoculum of *R. solani* AG3, causing stem canker and black scurf, and *S. subterranea* causing powdery scab in the rotation than in continuous potatoes. This indicates that the break period serves to minimise the build-up in inoculum that occurs under continuous potatoes. However, despite the reduced inoculum levels in the rotation, disease incidence and severity did not differ significantly from that in continuous potatoes, which suggests that even the reduced inoculum levels were above the threshold required for disease.

There was no clear evidence of differences in disease incidence and severity between the different pasture crops, namely perennial ryegrass and white clover, and the fodder Brassica. Quantitative DNA of *R. solani* AG3 and *S. subterranea* did not vary significantly under the different hosts in the two-year rotation. The 3-year study of root systems of pasture crops found that fodder Brassica and white clover supported epiphytic populations of *R. solani* sclerotia and hyphae, whereas ryegrass roots did not. Although the *R. solani* from Brassica and clover was not characterised into AG groups, a previous study had found that the sclerotia isolated from Brassica roots were AG3, the potato infecting strain (de Boer 2003a). Nevertheless, it does not appear from our study that the clover and Brassica significantly affected populations of *R. solani* AG3.

A beet seed bioassay indicated a potentially higher population of *R. solani* AG3 in a clover/clover/potato treatment than in grass/grass/potato, wheat/clover/potato and continuous potato treatments. However, this does not correlate with disease incidence and severity under those treatments. The beet seed bioassay is a measure of the saprophytic activity of *R. solani* (Frank and Murphy 1977) and may potentially select for saprophytic strains to the detriment of pathogenic strains (Banville *et al.* 1996; Specht and Leach 1987). With regard to clover potentially favouring *R. solani* over grasses, there is a generally held belief that grasses and cereals are good rotation crops for control of *R. solani* (Banville et al. 1996), although there is no consistent evidence of this in the literature. Celetti *et al.* (1989b) reported a higher incidence of *R. solani* in clover stands than in Italian ryegrass and other crops. However, differences in Rhizoctonia stem canker levels in subsequent potato crops grown after the different rotations were not evident (Celetti *et al.* 1989a; b; Celetti *et al.* 1990). Larkin and Honeycutt (2006) report that potato crops rotated with clover or soybean, had higher levels of Rhizoctonia damage compared with potatoes rotated with barely and oil-seed Brassica crops.

This trial was conducted over a period of four seasons, following on from a commercial potato crop in 2000/2001. Others have reported that differences in disease and yield between

rotation treatments did not become apparent for several years into a field trial (Carter and Sanderson 2001). Although there was a trend of higher disease levels in the continuous potatoes in our trial, our data indicates that inoculum levels in the three pasture/fodder rotations was high enough to support disease levels similar to that in continuous potatoes, which had highest DNA levels. *R. solani* can multiply quickly from a low initial inoculum level (Scholte 1992). *S. subterranea* also has the potential to rapidly build up inoculum on potato roots from a low base (Harrison *et al.* 1997). The Russet Burbank variety is rated as slightly susceptible to powdery scab. The effects of changes in soil inoculum *S. subterranea* on disease on tubers tend to be more apparent on highly susceptible varieties, than on relatively tolerant cultivars.

Because the 1:2 and 1:3 rotations were not in 'phase' (i.e. potato crop sown after each rotation were not planted together the same season), a direct comparison of these treatments was not possible due to the confounding effects of season. Reports from several studies show that, in the long term, time out of potatoes is the most important factor affecting disease caused by *R. solani* (Carter and Sanderson 2001; Johnston *et al.* 1994; Peters *et al.* 2004; Scholte 1987). In a long-term trial in Canada, Rhizoctonia stem canker, stolon canker and black scurf declined over time (over a 6-year period) in a long term 3-year rotation compared with a 2 year rotation (barley/potato, and barley/red clover/potato) (Peters *et al.* 2004). The 2-year rotation was not sustainable because of diseases and a decline in tuber quality.

DNA analysis of soil in 2004 showed that populations of *R. solani* AG2-1 were higher in Brassica/potato rotation than in potato/potato, clover/potato and ryegrass/potato. AG2-1 is commonly associated with Brassica and leguminous crops in (such as clover) (Hwong *et al.* 1996; Khangura *et al.* 1999; Wong and Sivasithamparam 1985). It is not clear why the population of *R. solani* AG2-1 was relatively low in the clover rotation. More work is needed to determine whether the DNA test is specific to AG2 strains affecting clover or whether Brassica is a better host of AG2 strains than clover.

There was little measurable impact of rotation treatments on Rhizoctonia diseases of the stem and tubers in the trial. However, in 2003/04 season, the incidence and severity of stem canker was 50% less in continuous potatoes than in potatoes after a two year break. (Larkin and Honeycutt 2006) report of a higher incidence and severity in continuous potatoes than in potato in rotations and it is reported generally that a break in potatoes reduces the incidence of Rhizoctonia diseases (Carter and Sanderson 2001; Peters *et al.* 2004; Scholte 1992). One possible explanation for our result is restrictions in the growth of the pathogen in soil due to compaction and, associated with this, a reduced ability of 'outrun' antagonists, as *R. solani* is relatively poor competitive saprophyte. Another explanation is that the soil in the continuous potato treatments has become 'suppressive' to the pathogen. The higher incidence of stem canker in potatoes after lucerne compared with potatoes after barley might be due to a preference for *R. solani* for lucerne over barley roots. We observed *Rhizoctonia* hyphae colonising the roots of lucerne but not of barley.

A higher incidence of powdery scab was recorded in continuous potato treatments compared with potatoes in rotation in 2002/2003 but not in 2003/2004. *S. subterranea* is an obligate parasite of potatoes and diseased roots and tubers can release large quantities of resistant cystosori into the soil each season. Inoculum would be expected to build up with successive potato crops as indicated by the results of DNA soil tests in 2004/05. However, demonstrating good correlation between DNA data and disease incidence and severity is difficult (S Wale, Scottish Crops Research Institute, personal communication). Cystosori survive in soil for several years without a host (Harrison *et al.* 1997) and previous research

showed that rotations of 4 years or more do not reduce powdery scab (de Boer 2003a; de Boer and Theodore 1997).

There was no effect of rotation treatments on the two tuber diseases black dot and silver scurf. The pathogens causing these diseases are very common in the soils of the Central Highland (de Boer 1997; 2003b) and are also seed-borne. Both diseases are common on seed potatoes (de Boer and Wicks 1994) and planting infected seed will help maintain pathogen populations.

Our study reports the results of a relatively short term field study of the potential interactions between pasture species and the disease and yield of potatoes. Our study was not able to determine any specific advantage or disadvantage or different components of pasture and fodder crops on disease incidence, severity or yield. Long term studies in Canada, which combined different rotations with conservation tillage in practices, have proved to be an invaluable tool for studying the productivity of the potato crop in relation to soil health and disease management (Carter and Sanderson 2001; Larkin and Honeycutt 2006; Peters *et al.* 2004). Peters *et al.* (2003) conclude from their research that "the potential exists for cropping systems to be engineered to suit specific agro-ecosystems and through selection of rotation crops that minimise plant pathogen attack and reduce saprophytic survival, while stimulating functional development of communities of beneficial, disease suppressive rhizosphere and endophytic micro-organisms". The challenge is to integrate other disease management options into the rotation system. Current research under the Processing Potato R&D Program is investigating a number of disease management options, including the use of organic soil amendments and soil applied fungicides.

4.6 Conclusions

A pasture phase is an important feature of many seed potato production enterprises. Past research in Australia has shown that pastures replenish soil nitrogen and restores soil structure after cropping. In our study, the one and two year break from potatoes resulted in better yields than continuously cropped potatoes. Although there was no major effect of the break from potatoes on disease incidence and severity in our trial, the break served to reduce soil-borne inoculum of the major potato pathogens. The effect of the break was more important than the individual component pasture and fodder species.

The positive impact of the rotation on disease depended to some degree on the pathogen. *R. solani* is both a saprophyte and parasite, able to exploit its environment including the root systems of crops other than potatoes. *S. subterranea*, on the other hand, is an obligate parasite which produces long-lived resting spores and is, therefore less exposed to the effects of rotation and the environment. *H. solani* and *C. coccodes* populations are readily reintroduced by planting infected seed potatoes and this may nullify the effects of rotation on soil-borne populations of these pathogens. Other disease management strategies should to be integrated into the rotation to reduce soil-borne inoculum below disease threshold levels, such as effective seed treatments, soil-applied fungicides, green manures, soil-applied organic amendments and tillage practices.

5 Technology Transfer

Communication with growers and the industry

- *Rhizoctonia solani* anastomosis groups associated with potatoes in Victoria. Presentation at the Potato Grower Seminar organised by Central Highlands's Integrated Production Systems Group, 27 August 2002, Burkes Rd Bullarook, Victoria.
- Potato Growers Seminar - CHIPS Demonstration Farm, Bullarook, 22 August 2001
- Grower workshops, Colac, Portland and Ballarat, Victoria, 2-3 September 2002
- Grower workshops Devonport and Scottsdale, Tasmania, 7-8 October 2002
- ViCSPA Certification Workshop - Toolangi, 17 January 2002
- *Rhizoctonia solani* anastomosis groups associated with potatoes in Victoria. Presentation at the Horticulture Conference, 21-22 August 2002, DPI Knoxfield.
- Rhizoctonia stem canker and black scurf – Biology and Management. Presentation to Tasmanian potato growers, Devonport and Scotsdale October 2002
- 3rd Biennial Seed Potato Industry Workshop, Portland 2003 – Managing Rhizoctonia in potatoes
- Powdery scab of potatoes – Research in Australia. Potato Association of America Meeting, Scottsbluff Nebraska, 8 -12 August 2004
- Vegetable Growers Technical Conference, Pukekohe NZ, July 2005 – Managing Rhizoctonia in Potatoes
- Potato 2005 *National Potato Conference* - Today & Beyond, Phillip Is. VIC, 19-21 September 2005 - "Looking forward to looking back" Five-year review of outcomes of pathology research.
- Vegetable Growers Technical Conference, Pukekohe NZ, March 2006 – Soil-borne diseases of potatoes.
- Vegetable Growers Technical Conference, Pukekohe NZ, March 2006 – Managing Rhizoctonia in Potatoes

Publications arising from this project

- Yearly reports for Potato Australia

- Petkowski, J.E., de Boer, R. F., 2001. *Rhizoctonia solani* anastomosis group AG3 and AG2-1 as pathogens of potato and other crops in potato production systems. Proceedings of the Second Australasian Soilborne Diseases Symposium, 5-8 March, Lorne, Victoria, Australia.
- Petkowski, J.E., Czerniakowski, B., de Boer, R. F. 2002. Rhizoctonia solani anastomosis groups associated with potatoes in Victoria, Australia. Horticulture Conference, 'Working with plants: How do you fit into the future?', 21-22 August 2002, Institute for Horticultural Development, Knoxfield, Victoria.
- Petkowski, J.E., de Boer, R.F. 2002. Know your enemy – rotation trials help unravel the Rhizoctonia story. *Potato Australia* 13:40-42.
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- de Boer, R. F. and Petkowski, J. E. 2004. *Rhizoctonia solani* AG 2-1 and 2-2 implicated as the cause of a wilt disease of potatoes in Australia. Proceedings of the 3rd Australasian Soilborne Diseases Symposium, 8-11 February, Rowland Flat, South Australia. pp 192-193.
- de Boer, RF 2004. Powdery scab of potatoes – Research in Australia. Proceedings of the Potato Association of America Meeting, Scottsbluff Nebraska, 8 -12 August 2004

6 Recommendations

Australian research has shown that a pasture phase provides clear benefits after cropping. Pastures have been shown to replenish soil nitrogen and restore soil structure. Our study shows that the pasture phase improves yield and provides a break in the disease life cycle by reducing the build-up of inoculum of the major soil-borne pathogens. However, over the period of this trial, inoculum was maintained at high enough levels to cause disease in potatoes, irrespective of the pasture species. The stipulated break for certified seed growers is a minimum of 3 years. Longer rotations are often not practical unless growers have access to 'new' ground not cropped to potatoes before. Other disease management options should be integrated into the pasture rotation in order to maximise the benefits of the pasture break. These include:

- Chemical seed treatments to minimise the re-introduction of seed-borne inoculum
- Soil chemical treatments for control of Rhizoctonia stem canker, common scab and powdery scab
- Green manure crops which improve soil structure and suppressiveness to disease
- Organic soil amendments which have both direct and indirect (suppressiveness) effects on pathogens and disease
- Conservation tillage which has been shown in Canadian studies to enhance soil suppressiveness to pathogens in rotation systems.
- Selective removal of clover in favour of grasses before cropping potatoes
- Effective management of potato volunteers in the rotation.

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