Hort Innovation

Final Report

Exploring Spongospora suppressive soils in potato production

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Delivery partner:

New Zealand Institute for Plant and Food Research Limited

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Summary

This project examined characteristics of field soils that were putatively suppressive to the potato yield- and quality-limiting diseases caused by *Spongospora subterranea* (tuber powdery scab and root hyperplasia). The project aimed to determine if specific soil physical, chemical and/or biological factors were associated with disease suppression, and assess transferability of suppression to non-suppressive soils. The project objective was to provide knowledge to assist possible management of *Spongospora* diseases through manipulation of soil factors. The study focused on field soils from the South Auckland/Waikato region of New Zealand, including Pukekohe, a location where a previous long-term field trial demonstrated very low amounts of powdery scab developing in susceptible potatoes, despite 9 years of continuous potato cropping.

Research in the project was carried out in three phases, during 2017/2018 (**Phase 1**), 2018/2019 (**Phase 2**), and 2019/2020 (**Phase 3**; project extension). In each phase, a large greenhouse pot experiment was completed. Different field soils were assessed for their respective potato pathogen populations (amounts of DNA) and physicochemical profiles. The soils were then placed into large capacity pots and removed to a greenhouse, where the pots were planted with potato seed tubers (cv. 'Agria'; very susceptible to powdery scab). Experimental treatments included without (experimental controls) or with *S. subterranea* inoculum applied at seed tuber planting. Assessments were made of the soil microbiological profiles, and incidence and severity of *Spongospora* root galling and tuber powdery scab for the resulting potato plants.

The **Phase 1** experiment included 12 field soils with different characteristics and crop histories, aiming to determine if some fields were more suppressive of *Spongospora* diseases than others, and if this was the case, to develop knowledge of the soil physicochemical or microbiological characteristics associated with *Spongospora* disease suppression. This experiment confirmed that some soils were more suppressive than others to root galling and tuber powdery scab, but disease suppression was not related to their specific physicochemical factors or microbial classification profiles.

The **Phase 2** experiment focused on six field soils each with similar physicochemical characteristics, and which were shown to have different suppressiveness to *Spongospora* diseases in **Phase 1**. Experimental treatments included heat treatment of the soils before planting, to indicate if suppressiveness was "general" (associated with total soil microbial and/or soil abiotic factors) or "specific" (associated with particular soil microorganisms), and a treatment to determine if suppression could be transferred from the six putatively suppressive soils to a conducive soil. *Spongospora* suppression was "general" in three of the soils, and transferable (possibly "specific") in the other three. Relationships were mostly weak between disease suppression, physicochemical characteristics or high level classification of soil microbial populations. An exception was that all six suppressive soils contained large amounts of manganese (Mn; 292 to 634 mg/kg), while the conducive soil contained a normal amount of this element (49 mg/kg).

Following indication from **Phase 2**, the **Phase 3** experiment used, as treatments, 12 field soils with low to very high natural Mn contents (21 to 885 mg/kg), without or with *S. subterranea* inoculation. Other treatments included pre-planting application of Mn to the soils, and foliar applications to growing potato plants. High natural soil Mn contents had little effect on powdery scab in progeny tubers, particularly where pre-planting levels of *S. subterranea* DNA were also high. Otherwise, the different natural soil Mn contents generally had little effect on incidence or severity of the disease. The exceptions were for three soils containing very low amounts of natural soil Mn (21 to 24 mg/kg), and small amounts of pre-planting *S. subterranea* DNA. In these soils, foliar and soil Mn applications reduced incidence and severity of powdery scab.

Outputs from the project include ten presentations to potato growers and industry, 11 industry magazine publications, and a paper presented at the XIth International Congress of Plant Pathology. A manuscript describing some of the research is in advanced draft form (May 2020), and an article has been prepared for publication in industry magazines (Appendix 2).

Outcomes from the project include:

- Incidence and severity of *Spongospora* diseases of potato (root galling and tuber powdery scab) shown to develop differently in different New Zealand field soils.
- Previous crop rotations did not affect development of *Spongospora* diseases in potato plants grown in soils from respective fields.
- Suppression of *Spongospora* diseases was locally specific. Soil microorganisms were probably involved in disease suppression in three of six soils tested, because their elimination or reduction by soil heat treatment increased incidence and severity of *Spongospora* root galling and tuber powdery scab. Particular bacterial and fungal genera may play roles in *Spongospora*-suppressive soils.
- Soil texture, and especially soil drainage, influenced powdery scab incidence and severity.

- New knowledge on factors affecting incidence and severity of one of Australasia's major causes of yield and quality losses in harvested potatoes, particularly that some soils possess microbial and/or physicochemical properties that suppress diseases caused by *Spongospora subterranea*.
- Heat treatment of three soils (of six tested) removed *Spongospora* suppression, indicating that micro-organisms were involved with disease suppression. This suppression was confirmed as transferable "specific" suppression when small amounts of the respective soils were added to a conducive soil. The three other soils had possible "general" (non-transferable) suppression.
- Potato plants grown in field soils containing high levels of manganese (Mn) developed less severe *Spongospora* diseases than plants grown in a disease-conducive low Mn soil. Additions of Mn at seed tuber planting to three soils with low natural Mn contents reduced powdery scab on harvested tubers. However, additions of Mn to soils with "normal" to very high natural Mn contents did not affect the disease.
- Increased awareness by potato industry operatives (Australia, New Zealand and elsewhere) of powdery scab suppressive soils, and of soil factors potentially affecting disease suppression.

This project was carried out by: Mr Peter Wright (plant pathology), Dr Craig Anderson (biogeochemistry, soil microbial ecology), Dr Denis Curtin (soil chemistry), Dr Rebekah Frampton (molecular biology, microbiology, bioinformatics), Dr Duncan Hedderley (biometrics), and Prof Richard Falloon (plant pathology, project management), with assistance from Mr Grant Morris and Ms Miriam Hall (business management).

Keywords

Potato; *Spongospora subterranea*; root hyperplasia; tuber powdery scab; disease suppression; chemical, physical and biological soil factors; soil- and foliar-applied manganese; potential disease management

Introduction

In disease suppressive soils, the underground parts of plants are protected from diseases where soilborne pathogen inoculum would normally cause infections. Different physical, chemical and/or biological soil factors can contribute to disease suppression. "General" disease suppression is associated with overall soil microbiota, and physical or chemical factors (e.g. temperature, structure, organic matter, pH, water and mineral contents, fertiliser inputs) may contribute to suppression. "Specific" suppression, on the other hand, is due to interactions between soilborne pathogens and particular antagonistic microorganisms, and this can develop after continuous mono-cropping ("disease decline").

A long-term crop rotation trial, at a site near Pukekohe, New Zealand (Wright et al., 2015), indicated that the soil did not support development of powdery scab of potatoes (caused by *Spongospora subterranea*), despite 9 years of continuous potato culture using powdery scab-susceptible cultivars. In many other regions of New Zealand and elsewhere, *Spongospora* diseases (root galling and tuber powdery scab) are well-recognised as major causes of reduced potato plant and crop productivity (Falloon et al., 2016), and as diseases that develop rapidly where periods (rotations) between potato crops are short. These diseases have also been shown to contribute to the "potato yield gap", where potato crop yields in New Zealand's major potato production region averaged 35 tonne/ha below optimum (Sinton et al., 2020).

There is evidence that *Spongospora* diseases can be affected by soil nutrients. For example, Falloon et al., (2001a; 2010) showed that high levels of boron, manganese and zinc reduced potato root infection by *S. subterranea*, and adding boron to field-grown potatoes reduced powdery scab incidence and severity (Falloon et al., 2001b). In contrast, high rates of nitrogen fertiliser (calcium ammonium nitrate) have been linked to increased severity of powdery scab on tubers (Falloon et al., 2007).

The objectives at the outset of this project were:

1. to confirm that specific soils have characteristics that suppress or encourage incidence and/or severity of *Spongospora* diseases of potato;
2. to define potential mechanisms for suppression (biological, chemical and/or physical soil characteristics);
3. to determine if *Spongospora* suppression is transferrable to non-suppressive soils; and
4. depending on knowledge advances (and in consultation with Hort Innovation), to develop appropriate extension and communications for industry on progress in the project.

The hypothesis tested was;

> that some intensive cropping soils are suppressive to the potato yield- and quality-limiting diseases caused by *Spongospora subterranea*, and that disease suppression is associated with soil chemical, physical and/or biological characteristics.

The project included three greenhouse pot experiments, where different field soils were assessed for their physical, chemical and biological characteristics, and for their suppressiveness/conduciveness to potato root hyperplasia (galling) and tuber powdery scab caused by *S. subterranea*. This report summarises the project procedures, results, outputs and outcomes, from experiments carried out in 2017/18 (**Phase 1**), 2018/19 (**Phase 2**) and 2019/20 (**Phase 3**). Key **Phase 1** and **Phase 2** results are presented in Appendix 1 of this report, and detailed outline of the **Phase 3** experiment is included as Appendix 2, to this report, as the final Milestone Report for the project.

Methods and results

Three large greenhouse experiments (see **Figure 1**) were carried out during the growing seasons of 2017/18, 2018/19 and 2019/20 (respectively, **Phase 1**, **Phase 2** and **Phase 3**, below). The experiments each used several different field soils, samples of which were assayed for potato pathogen DNA, chemical and physical characteristics, and microbial activity/diversity.

Experiment methods Field soils (approx. 20 cm depth) were collected into large pots (35 L capacity) and removed to a greenhouse. They were each planted with a disease-free cv. 'Agria' seed tuber (very susceptible to powdery scab; Genet et al., 2017). At the time of planting, half of the seed tubers (pots randomly selected) were inoculated with *S. subterranea*, and the rest were left uninoculated (experimental controls). The inoculated seed tubers each received 20 mL of *S. subterranea* sporosorus/water suspension (approx. 1.75×10^7 resting spores; Falloon et al., 2011; 2016), applied into the planning hole, before covering with soil.

Figure 1. Greenhouse pot experiment (established on 27 July 2017; **Phase 1**) to assess expression of *Spongospora* diseases in 12 different field soils (indicated by different colours on the pot rims). Each pot was planted with an 'Agria' seed tuber. Half of the planted seed tubers were inoculated with *Spongospora subterranea* sporosori with the remaining tubers left uninoculated (experimental controls). The trial included 288 pots, and each double row of pots (left to right) is an experiment replicate.

Assessments of chemical, physical and biological characteristics of field soils Each field soil used in the pot experiments was analysed, using standard protocols, for different chemical, physical and biological factors (before or during the experiments). These factors were:
Potato pathogen DNA: PreDicta® Pt tests and field sampling protocols (Anonymous, 2020a; 2020b).
Chemical parameters: pH, amounts of phosphate (Olsen P test), exchangeable cations (Ca, K, Mg and Na), mineral N (Keeney & Nelson, 1982), micronutrients, total organic matter, cold and hot water extractable C and N, particulate organic matter (Ghani et al., 2003), cation exchange capacity (CEC), and anion storage capacity.
Physical parameters (texture): sand, silt and clay contents (Gee & Or, 2002).
Soil respiration: C mineralisation.
Microbial profiles: Soil samples from the different treatments applied in the **Phase 1** and **Phase 2** greenhouse experiments (below) were collected from bulk soil before tuber planting, and from plant rhizosphere soil at 12 weeks after planting when root systems were assessed for severity of *Spongospora* root galling. The bacterial 16S rRNA and fungal internal transcribed spacer (ITS) regions were targeted for microbial profiling (Klindworth et al., 2013; Martin & Rygiewicz, 2005). Amplicon sequence data were partitioned into amplicon sequence variants (ASVs) using dada2 (Callaghan et al., 2016) and phyloseq (McMurdie and Holmes, 2013). The Silva and

UNITE databases were used to determine putative ASV identifications (Quast et al., 2013; Kõljalg et al., 2013).

Assessments of* Spongospora *diseases Severity of *Spongospora* root hyperplasia (numbers of root galls per plant) was assessed (approx. 12 weeks after planting) for half of the plants receiving each experimental treatment. Progeny tubers from the remaining plants were harvested (approx. 14 weeks after planting) and were individually assessed for incidence and severity (Falloon et al., 1995) of tuber powdery scab. Powdery scab severity was scored as the proportion (%) of tubers with greater than 5% of tuber surface area affected (>5% tuber saa) by the disease. Disease severity data were statistically analysed using appropriate standard methods.

Relationships between soil and disease parameters The Multivariate statistics package PRIMER-E v7 (Clarke and Gorley, 2015) was used to examine relationships between the soil microbiome, soil physicochemical data and *Spongospora* disease data. The following statistical procedures were used: principal component analysis (PCA), multidimensional scaling (MDS), permutational analysis of variance (PERMANOVA), distance-based linear modelling (DISTLM), and redundancy analysis (dbRDA).

Phase 1 experiment (2017/18): *Spongospora* conduciveness/suppressiveness in selected field soils

Twelve soils were collected from fields in the South Auckland/Waikato regions of New Zealand. The fields represented spectra of different cropping histories, soil types, and potential conduciveness or suppressiveness to *Spongospora* diseases. The greenhouse experiment included treatments of the 12 field soils, with or without *S. subterranea* inoculation, in 12 replicates (total of 288 pots). Severity of root galling was assessed in half of the actively growing plants (randomly selected). At plant maturity, progeny tubers from the remaining plants were assessed for incidence and severity of powdery scab.

Predicta® Pt tests (Appendix 1, Table A1.1) detected no *S. subterranea* DNA in six of the soils, small amounts (3 to 7 pg/g soil) in five soils, and 709 pg/g in one soil. The soils contrasted in physicochemical characteristics, and in microbial activity.

Root galling (Appendix 1, Table A1.2). No *Spongospora* root galls occurred on the non-inoculated plants in any of the soils. Inoculation with *S. subterranea* sporosori gave few root galls in seven of the soils (means = 0 to 2 galls/plant; possible suppression of galling), but increased numbers of root galls in five of the soils (means = 4 to 11 galls/plant).

Tuber powdery scab (Appendix 1, Table A1.2). Severity of powdery scab on progeny tubers also differed in the different field soils. Inoculation with *S. subterranea* gave very little powdery scab in six of the soils (means = 0 to 3% of tubers >5% saa; possible suppression), but increased powdery scab in the other six of the soils (means = 9 to 39% of tubers >5% saa). Suppression of both root galling and tuber powdery scab occurred for five of the soils.

Bacterial and fungal DNA extracted and analysed from samples of the 12 field soils, and from bulk and plant rhizosphere soil in the pots during the experiment, identified 1451 to 2379 bacterium ASVs and 243 to 515 fungus ASVs in the different soils. Microbial diversity analyses showed that the bacterium and fungus OTUs in the 12 soils were markedly different, and were associated with the different field locations (soil types) and cropping histories (long-term intensive vegetable cropping *vs* pasture). However, there were no clear relationships between abundance of bacteria or fungi in the different soils and the respective severities of *Spongospora* root galling on the potato plants or powdery scab on their progeny tubers. Furthermore, assessments of microbial phylogenies in the respective soils did not show any relationships between microbial community structure and incidence or severity of these diseases on plants growing in the different soils.

Phase 2 experiment (2018/19): confirmation of *Spongospora* disease suppression in field soils, and assessment of associations between soil factors and disease suppression

Six soils of one soil type (volcanic origin) but with different cropping histories were collected from fields within 10 km of each other, near Pukekohe, New Zealand. A seventh powdery scab-'conducive' field soil (the soil that gave the greatest *Spongospora* disease severities in the **Phase 1** experiment) was also collected. Treatments in the **Phase 2** experiment were applied in 12 replicates to a total of 288 pots, with the experimental treatments being the six field soils and four inoculation/soil treatments, which were:

1. no *S. subterranea* inoculum applied to untreated soil (experimental control);
2. *S. subterranea* inoculum applied to the seed tuber at planting planted into untreated soil;
3. *S. subterranea* inoculum applied at seed tuber planting, where the soils had been previously heat-treated (70°C for 1 hour), (test for 'general' disease suppression); or
4. *S. subterranea* inoculum applied at seed tuber planting, to a mixture of 1 part of each soil to 9 parts the powdery scab-'conducive' soil (test for 'specific' disease suppression).

Predicta® Pt analyses (Appendix 1, Table A1.3) of pre-planting samples of five of the six field soils showed that they contained small amounts of *S. subterranea* DNA (5 to 26 pg/g soil), while the sixth soil contained 488 pg/g of *S. subterranea* DNA. Severity of *Spongopsora* root galling in plants growing in all six soils that were not inoculated with *S. subterranea* (experimental controls) was slight (0 to 7 galls/plant), and was not related to amounts of pre-planting DNA. For powdery scab on progeny tubers, no moderately severe disease (tubers >5% saa) occurred on progeny tubers from the five low DNA and non-inoculated soils. More severe powdery scab (mean = 15% of tubers >5% saa) developed in the non-inoculated soil with 488 pg/g of pre-planting *S. subterranea* DNA.

Root galling (Appendix 1, Table A1.4). Few *Spongospora* root galls occurred on the non-inoculated plants (experimental controls) (overall mean = 2 galls/plant), and root galling was only slightly more severe (mean = 6 galls/plant) from *S. subterranea* inoculation. This indicated that all of the soils were suppressive of root galling. Heat treatment plus inoculation of three of the soils did not affect root galling (mean = 5 galls/plant), indicating possible "general" suppression of the disease in these soils. Heat treatment plus inoculation increased numbers of galls in the other three soils (means = 17, 19 and 25 galls/plant), indicating possible "specific" root galling suppression. This was confirmed when small amounts (10%) of the respective soils were added to the conducive soil. From this treatment, the three soils with possible "general" (non-transferable) suppression gave very severe root galling (means = 71, 81 and 120 galls/plant), while the soils with possible "specific" transferrable suppression gave much less severe disease (means = 27, 32 and 35 galls/plant).

Tuber powdery scab (Appendix 1, Table A1.5). The pattern of treatment effects on severity of powdery scab on progeny tubers was similar to that for root galling. From the no inoculum treatment (experimental controls) for five of the soils, no moderate to severe powdery scab occurred on the tubers, but the soil with high *S. subterranea* DNA at planting (488 pg/g), gave more severe powdery scab (mean = 15% of tubers >5% saa). The *S. subterranea*-inoculated treatment to all six soils gave low powdery scab severity (means = 0 to 5% of tubers >5% saa), indicating that all six soils suppressed the disease. Heat treatment of two of the inoculated soils slightly increased powdery scab severity (means = 7% and 18% of tubers >5% saa). (Heat treatment of these two soils also gave low severity of root galling, above). Heat treatment increased powdery scab severity in the other four soils (means = 26 to 41% of tubers >5% saa), indicating that powdery scab suppression in these soils was microbe-mediated. Similarly, when the respective soils were added to the conducive soil, two of the soils gave relatively low disease severity (means = 8% and 9% of tubers >5% saa; possible "specific", transferable suppression), compared with the other four soils (means = 19% to 23% of tuber >5% saa; possible "general", non-transferable suppression).

Previous crop rotation did not affect powdery scab severity in the inoculated soils. Bacteria and fungi known to be antagonistic to soilborne plant pathogens and/or promoters of plant growth were present in the disease suppressive soils, and may have contributed to powdery scab suppression.

Bacterium and fungus diversity and abundance Differences were detected in diversity and abundance of bacteria and fungi in the microorganism DNA extracted from pre-planting bulk soil samples (including the 'conducive' soil), and from bulk and plant rhizosphere soil samples from samples taken 12 weeks after planting.
Bacterium diversity in the pre-planting bulk soil samples was generally similar for the six soils, reflecting their close relatedness, and bacterial diversity was much greater in the six putatively suppressive soils than in the conducive soil. Bacterium diversity was greater in pre-planting bulk samples than at 12 weeks post-planting.
Bacterium phylum abundance was also similar in the pre-planting samples from the six soils, but was much less in the conducive soil. Heat treatment of all six treatment soils gave similar effects on abundance, decreasing relative abundance of Acidobacteria, Bacteroidetes, Gematimonadetes, Proteobacteria and Verrucomicrobia, but increasing abundance of Firmicutes and Latescibacteria.
Fungus diversity in three of the treatment soils, and in the conducive soil, was similar in the pre-planting and the 12-week post-planting samples. For the other three soils, fungus diversity was much less in the pre-planting soil samples, but diversity in all seven soils was similar at 12 weeks post-planting.
Fungus phylum abundance in pre-planting soil samples was also greater in the three same treatment soils than in the other three treatment soils and the conducive soil. These differences, however, were much less in the 12-week post-planting samples. Ascomycota and Basidiomycota were predominant in the six treatment soils and the conducive soil. Chytridiomycota were detected at low levels in all of the soils, but were more abundant

in two of the treatment soils than in the others. Heat treatment generally reduced these differences, but in one soil increased abundance of Rozellomycota.

Physicochemical parameters For the soils inoculated with *S. subterranea*, previous crop rotations did not affect incidence or severity of powdery scab. Two of the soils, both with less than 3% organic matter, gave low powdery scab severity (no tubers >5% saa), while the other four soils (3.9 to 5.3% organic matter) gave more severe disease (means = 2 to 7% tubers >5% saa). Otherwise, the six suppressive soils, and the conducive soil, had generally similar physical and chemical profiles.

The exception was for Mn. All six of the 'suppressive' soils contained very large amounts of this element (292 to 634 mg/kg). The Mn content of the 'conducive' soil was much less (49 mg/kg), similar to amounts of this element (overall mean = 42 mg/kg) in 23 previously assayed Canterbury (New Zealand) agricultural soils (Curtin et al., 2008). This suggested that the suppressiveness to *Spongospora* diseases in the six field soils may have been related to their high Mn contents. Principal component analyses of relationships between soil physico-chemical factors and *Spongospora* diseases also indicated that Mn content in the soils had the strongest negative relationship with root galling and tuber powdery scab severity, while contents of iron, zinc, silt and organic matter had weakly positive relationships with these diseases.

Phase 3 Experiment (2019/20; project extension): Effects of soil and/or foliar-applied manganese on development of *Spongospora* diseases (see Appendix 1)

The **Phase 3** greenhouse experiment investigated effects of natural levels of soil Mn, and soil- or foliar-applied Mn, on development of powdery scab on progeny potato tubers. This was after indications from the **Phase 2** experiment, where all the *Spongospora* suppressive soils contained large amounts of Mn. This element has been associated with suppressive effects on many soilborne plant diseases, including potato diseases caused by *Phytophthora*, *Rhizoctonia*, *Streptomyces* or *Verticillium* (Thompson and Huber, 2007). Manganese has also been shown to be "mildly inhibitory" to *Spongospora* root galling in potato plants grown experimentally in sand/nutrient solution (Falloon et al., 2010).

The hypothesis tested in the **Phase 3** experiment was:

> that high natural levels of soil manganese reduce powdery scab on potato tubers, and that manganese additions (soil or soil + foliar applications) reduce this disease.

Soil samples were collected from 23 arable-vegetable cropping fields from the South Auckland, Waikato and Manawatu/Wanganui regions of New Zealand, and these were assessed for micronutrient contents. Soils from 12 of the fields (hereafter designated as Soils A to K) were selected for the experiment, based on their Mn contents (below).

The 12 selected soils represented a broad range of natural amounts of Mn (21 to 885 mg/kg; low to very high). They also contained different amounts of zinc (2.2 to 24.0 mg/kg; normal to high), and boron (0.83 to 1.83 mg/kg; normal), and had different fertility profiles, including pHs from 6.1 to 7.8, and amounts of organic matter (3.4 to 23.5%), total carbon (2.0 to 13.6%), total nitrogen (0.14 to 0.94%), phosphorus (18 to 165 mg/L) and potassium (0.26 to 1.61 me/100 g).

Six replicates were used in the experiment (total of 288 pots), with experimental treatments being the 12 soils, to which the following four inoculation/Mn application treatments were applied:

1. no *S. subterranea* inoculum applied to untreated soil (experimental control);
2. *S. subterranea* inoculum applied to untreated soil at seed tuber planting;
3. *S. subterranea* inoculum applied to soil at seed tuber planting, plus pre-planting soil application of Mn, or
4. *S. subterranea* inoculum applied at seed tuber planting, plus pre-planting soil application of Mn, plus foliar applications of Mn to growing plants.

The pre-planting soil Mn treatment was at the equivalent of 1.04 kg Mn/ha, using chelated Mn microgranules (YaraVita™ Rexolin® Mn 13). The Mn foliar treatment was at the equivalent of 1 kg Mn/ha (2 L/ha of YaraVita™ Mantrac™ Pro (50% Mn) in 200 L water), and was applied to potato plants at 6 weeks after planting, followed by three further applications at 3 to 4 week intervals.

Tubers from the plants were harvested 20 weeks after planting, and the all 'marketable' tubers (>50 g) from each plant were harvested, weighed and individually assessed for incidence and severity of powdery scab.

Pre-planting* Spongospora *DNA in the soils Predicta® Pt analyses for soil samples from the 12 fields detected no *S. subterranea* DNA in seven of the soils, low amounts (4 and 18 pg DNA/g soil) in two, and high amounts (156, 200 and 346 pg/g) in the other three.

Tuber yields Mean tuber weights of 'marketable' tubers from the different soils were considerably different, from 0.67 to 1.49 kg/plant. However, these weights were not related to any of the measured soil fertility parameters, including pH, organic matter, or macro- or micro-nutrients. In particular, the yields were not related to the different amounts of pre-planting Mn in the soils, indicating that although the Mn contents in some of the soils were very high, these did not reduce plant productivity. Furthermore, the different inoculation/Mn application treatments gave similar overall mean tuber weights, of 0.96 kg/plant (non-inoculated treatment), 0.97 kg/plant from (*S. subterranea* inoculated), 0.97 kg/plant (inoculated + soil Mn), but slightly less at 0.91 kg/plant from the inoculated + soil + foliar Mn treatment.

Powdery scab incidence No powdery scab developed on tubers from eight of the soils in the uninoculated (control) treatment, while two of the soils gave low disease incidence (means = 3 and 4% tubers affected), and the other two gave greater incidence (means = 15 and 60%). Incidence from the non-inoculated (control) treatment was associated with amounts of pre-planting *S. subterranea* DNA. Three soils, containing 18, 4 or 0 pg DNA/g, gave low incidence (respectively, 0%, 0% and 4% of tubers affected), while three other soils, containing 156, 200 or 349 pg DNA/g, gave, respectively, 3%, 15% and 60% of tubers with powdery scab.

Inoculation with *S. subterranea* increased incidence of powdery scab in all of the soils (overall mean = 17%, range 3 to 72%). Effects of Mn additions were small. The inoculation + soil Mn treatment gave, overall, slightly less incidence (mean = 15%, range 3 to 85%), and the inoculation + soil + foliar Mn treatment also only slightly reduced incidence of the disease (mean = 13%, range 0% to 80%).

Powdery scab severity No moderate to severe powdery scab developed on tubers from ten of the non-inoculated soils. For tubers from the other two soils, one soil, which contained the least amount of pre-planting Mn (21 mg/kg), produced 3% of the tubers with >5% tuber saa, and the other soil (containing 885 mg/kg Mn) gave 31% of tubers >5% saa, indicating no relationship between large amounts of pre-planting Mn and low powdery scab severity. For seven of the soils inoculated with *S. subterranea* without added Mn, no moderate to severe powdery scab developed on tubers (possible powdery scab suppression). This treatment for the other five soils gave more severe disease (means = 3 to 47% of tuber saa).

For the two lowest Mn-containing soils (with 21 and 25 mg Mn/kg), the *S. subterranea*-inoculated + soil Mn treatment and the inoculated + soil + foliar Mn treatments both reduced powdery scab severity on the harvested tubers (to 0% of tubers >5% saa), compared with the *S. subterranea*-inoculated (no added Mn) treatment. However, for the soil with the greatest amount of pre-planting Mn (885 mg/kg), applications of Mn increased the proportions of tubers affected, from 47% tubers >5% saa without added Mn, to 59% from soil-applied Mn and 54% from soil +foliar Mn.

The three soils with the lowest amounts of pre-planting Mn (21, 24 or 25 mg/kg) also contained the greatest amounts of organic matter (respectively, 23.5%, 12.2% and 13.6%). These three soils all gave means of 0% of tubers >5% saa where applications of Mn were made to the soils and to plant foliage. Where inoculation and soil and foliar Mn were applied, very little moderate to severe powdery scab developed (2% tubers >5% ssa in one soil and 0% in ten of the soils). The soil with very high pre-planting *S. subterranea* DNA (349 pg/g) gave 54% tubers >5% saa. These results suggests that soil and foliar applications of Mn may reduce powdery scab for low Mn-containing soils, that this reduction may also be associated with high amounts of soil organic matter, but that these effects do not occur where *Spongospora* inoculum levels are very high.

Other than these effects, neither pre-planting Mn content, nor soil or foliar Mn applications, affected powdery scab severity on the harvested tubers.

Knowledge transfer activities (see Outputs, below)

Seminars describing details and progress in this project were presented in Australia and New Zealand at potato grower and industry field days and industry conferences. Popular articles were published in *Potatoes Australia* and *New Zealand Grower*, and in two North American online journals. One paper was presented at an international science congress.

Outputs

Presentations to potato growers and industry representatives

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Paper presented to an international science congress

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Outcomes

- Incidence and severity of *Spongospora* diseases of potato (root galling and tuber powdery scab) shown to develop differently in different New Zealand field soils.
- New knowledge developed on factors affecting incidence and severity of one of Australasia's major causes of yield and quality losses in harvested potatoes, particularly determining that some soils possess properties that suppress diseases caused by *Spongospora subterranea*. *Spongospora* suppression was transferable from some soils to a disease-conducive soil, indicating that this suppression may be due to soil microbes. Non-transferable suppression in other soils was likely to be due to soil physicochemical factors.
- Suppression of *Spongospora* diseases was shown to be locally specific. Soil microorganisms were shown to be involved in disease suppression; when they were eliminated or reduced by soil heat treatment, powdery scab incidence and severity increased. Particular bacterial and fungal genera may play roles in *Spongospora*-suppressive soils, but these were not identified.
- Soil texture, and especially soil drainage, were shown to influence powdery scab incidence and severity.
- Potato plants grown in six field soils containing high levels of manganese (Mn) developed less severe *Spongospora* diseases than plants grown in a disease-conducive and low Mn soil. These six suppressive field soils contained amounts of Mn that were 6- to 13-fold greater than the conducive soil, and similarly greater than cropping soils where *Spongospora* diseases are severe.
- Increased awareness by potato industry operatives (Australia, New Zealand and elsewhere) of powdery scab suppressive soils, and of soil factors potentially affecting disease suppression.

Monitoring and evaluation

Programme logic

		Progress
Broad goals	▪ Develop new knowledge as a basis for procedures/methods for integrated management of *Spongospora* diseases of potato. ▪ Characterise soil biological and/or physico-chemical factors associated with suppression/enhancement of potato diseases caused by *Spongospora subterranea* (tuber powdery scab, root galling). ▪ Reduced crop losses due to *Spongospora* diseases, reduced inputs to manage these diseases, and increased profitability of potato production.	▪ Achieved. ▪ Achieved. ▪ Dependent on future knowledge, based on indications from this project, and on adoption of new disease management strategies in potato production.
End-of-project outcome	▪ New knowledge of the soil factors (biological, physical, chemical) that influence development of *Spongospora* diseases of potato. ▪ Increased industry awareness of powdery scab suppressive soils.	▪ Achieved. ▪ Achieved, through industry presentations and publications.
Intermediate outcomes	▪ Provision of knowledge to influence ongoing research within this project. ▪ Plans for new disease management/treatment approaches to enhance suppression of *Spongospora* diseases of potato (should project results indicate that effective manipulation of disease suppression may be possible).	▪ Achieved. Phase 3 of the project was based on Phase 2 results. ▪ Dependent on recommended research, and adoption of new disease management in potato production.
Activities and outputs	▪ 6-month project reports (four). ▪ Final project report. ▪ Articles (two) in grower magazines (duplicated in Australia and New Zealand grower publications). ▪ Presentations (four) to appropriate grower/industry fora. ▪ Scientific publications/conference presentations (if results warrant).	▪ Five reports completed. ▪ Completed. ▪ Six articles published in grower magazines. ▪ Ten presentations given to grower/industry groups ▪ One paper presented at an international congress, and one manuscript drafted.
Foundational activities	▪ Project team liaison to ensure timely establishment and completion of planned experiments and assessments of soil biological and physico/chemical characteristics. ▪ Provision of research data. ▪ Development of outputs (magazine articles and presentations to grower/industry forums). ▪ Contract administration. ▪ Attendance at PT16002 project meetings. ▪ Assessment of industry feedback, and modification/implementation of and indicated changes required for effective knowledge transfer.	▪ Completed. ▪ Completed. ▪ Completed. ▪ Completed. ▪ Completed. ▪ Contract variations negotiated, added tasks completed, and one grower/industry presentation evaluated.

Project monitoring

	What was monitored	How monitored	By whom	Timing
Research activities	▪ Research plans. ▪ Greenhouse experiments. ▪ Laboratory tests/analyses. ▪ Data collection. ▪ Data analyses. ▪ Reporting.	▪ PFR record keeping. ▪ Milestone reports. ▪ Research reports.	▪ Project Leader and Project Team.	▪ Throughout project, to fulfil all Milestone deadlines.
Outputs	▪ Articles for industry magazines. ▪ Presentations to industry forums. ▪ Research reports. ▪ Scientific publications/presentations.	▪ Internal PFR record keeping. ▪ Milestone reports. ▪ Annual reports.	▪ Project Leader and Project Team.	▪ Throughout project, to fulfil all Milestone deadlines.
Industry alignment	▪ Relevance of project to industry needs. ▪ Engagement of industry in research progress. ▪ Appropriateness of engagement processes.	▪ Through Project Leader & Hort Innovation linkages ▪ Participant surveys at industry forums.	▪ HIA staff and Project Leader.	▪ Phase 3 initiative progressed and contract extension negotiated. ▪ One survey completed*.

*See Appendix 3

Reporting

The table below outlines Milestone Reports and other interim reporting scheduled and completed for this project.

Reporting for PT-16002. Highlighted text indicates internal project monitoring.

Reports/activities	Due date	Achievement
Research Agreement completed.	March 2017	Completed.
Research progress assessed by project team	September 2017	Completed.
First 6-month Report submitted.	September 2017	Completed.
One article prepared for publication in Australian and New Zealand potato industry magazines.	March 2018	Completed.
Two presentations made to grower/industry groups (one each in Australia and New Zealand).	March 2018	Completed.
Research progress assessed by project team	March 2018	Completed.
Second 6-month Report submitted.	March 2018	Completed.
Research progress assessed by project team	September 2018	Completed.
Third 6-month Report submitted.	September 2018	Completed.
Research progress assessed by project team	March 2019	Completed.
Fourth 6-month Report submitted.	March 2019	Completed.
Research progress assessed by project team	October 2019	Completed.
One article prepared for publication in Australian and New Zealand potato industry magazines.	November 2019	Completed.
Two presentations made to grower/industry groups (one each in Australia and New Zealand).	November 2019	Completed.
Project variation		
Fifth 6-month Report submitted.	November 2019	Completed.
Research progress assessed by project team	March 2020	Completed.
One article prepared for publication in Australian and New Zealand potato industry magazines.	May 2020	Completed.
Final report submitted.	May 2020	Completed.

Performance evaluation

Key evaluation questions	
Process effectiveness	
▪ How effective was the project for delivery of the intended outputs?	▪ Multi-disciplinary team assembled, which provided intended outputs.
▪ How effective were the research protocols for provision of robust data, to identify differences in field soils for expression of *Spongospora* diseases in potato, and to identify soil characteristics related to these differences?	▪ Research protocols were based on extensive experience with *Spongospora* as a potato pathogen. Plant pathology, soil science, microbiometrics and statistics expertise assembled in the research team.
▪ How effective was the delivery of industry communication (research plans and results communicated to industry in presentations at appropriate forums and in articles in industry magazines), by increasing industry awareness and commitment to test the new technologies/knowledge?	▪ With industry partners, presentation venues and events were identified, and detailed presentations were given. Articles were drafted, assessed by Hort Innovation representatives, and submitted. The articles were published in grower journals in Australia and New Zealand, and in two international online journals.
Project effectiveness	
▪ How effective was the project in delivering its intended outcomes?	▪ Outcomes from the project form a strong knowledge basis for further research.
▪ To what extent have potato industry stakeholders gained relevant information/knowledge on the project operation, and the results obtained?	▪ Not assessed.
▪ To what extent have industry stakeholders developed understanding of powdery scab suppressive soils?	▪ Not assessed.
▪ Has the project provided knowledge upon which alternative options (e.g. soil chemical or biological amendments) for management of *Spongospora* diseases can be based?	▪ Yes.
▪ Has the project assisted reduction of incidence and severity of *Spongospora* diseases in Australian and New Zealand potato crops?	▪ Dependent on future (possibly field-based) research.
▪ Has the project assisted productivity gains in potato cropping through improved management of *Spongospora* diseases?	▪ Dependent on future (possibly field-based) research.
Project evaluation	
▪ How relevant was PT 16002 to the needs of the intended beneficiaries, including targeted potato growers, advisors and other industry stakeholders?	▪ *Spongospora* diseases continue to be important in potato production. This project aimed to develop new knowledge to direct future initiatives for practical management of these diseases.
▪ How well have these intended beneficiaries been engaged in the research process?	▪ Close consultation with Hort Innovation allowed development of Phase 3 of this project.
▪ To what extent were engagement processes appropriate to the beneficiary target audiences?	
▪ What is the likelihood that the outputs, new knowledge and networks generated by the project will deliver value beyond the life of the project?	▪ Depending on funding support, knowledge from the project will direct future applied research.

Recommendations

Future research

Identification and evaluation of specific soil bacteria and/or fungi associated with suppression of* Spongospora *diseases
Detailed investigation of soil populations of bacteria and fungi in *Spongospora*-suppressive soils with appropriate microbiometrical analyses. Subsequent evaluations, in controlled experiments, of inoculation of plant growth media with specific microorganisms for effects on *Spongospora* diseases. Soils from different, but selected, cropping fields could be included in these experiments.

Assessment of effects of specific soil chemical characteristics for effects of* Spongospora *diseases
Controlled glasshouse experiments, using established techniques to grow potato plants in sand/nutrient culture (Falloon et al., 2003), to evaluate specific soil factors for effects on *Spongospora* infection of potato plants. Specific factors should include the macro-elements phosphorus and potassium, and combination treatments of the micro-elements manganese, boron and zinc.

Determination of* Spongospora *suppressiveness in vegetable production areas in Australia and New Zealand
Extensive surveys and evaluations of grower experience with *Spongospora* diseases of potato, to determine if *Spongospora*-suppressive soils occur in localities other than the volcanic soils of the Pukekohe region of New Zealand. Detailed assessment of soil physicochemical and microbial characteristics should be part of these surveys.

Assessment of effects of soil factors on* S. subterranea *inoculum levels in vegetable production fields
Field trials to evaluate manipulation of identified soil factors (microbial, physical, chemical) for reducing *S. subterranea* inoculum in vegetable production fields, and alleviation of *Spongospora* diseases.

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Intellectual property, commercialisation and confidentiality

No project IP, commercialisation or confidentiality issues to report.

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Appendices

Appendix 1
Key results from the Phase 1 and Phase 2 greenhouse experiments

Phase 1

Table A1.1. Phase 1 Predicta® Pt test results. Mean amounts (pg/g of soil) of DNA of *Spongospora subterranea* and other soilborne potato pathogens in samples of 12 different cropping soils from the Pukekohe and Matamata regions, New Zealand.

Soil	Region	***Spongospora subterranea***	*Colletotrichum coccodes*	*Rhizoctonia solani*		*Verticillium dahliae*	*Streptomyces scabies*
				AG-2.1	*AG-3*		
1	Pukekohe	**5**	8	10	0	23	0
2	Pukekohe	**0**	15	1	0	3	0
3	Pukekohe	**0**	0	0	0	0	12
4*	Pukekohe	**3**	19	1	0	39	0
5	Pukekohe	**709**	35	260	24	138	293
6	Pukekohe	**20**	0	13	0	1	0
7	Pukekohe	**4**	4	0	0	13	0
8	Pukekohe	**0**	0	0	0	0	355
9	Matamata	**0**	0	269	0	5	115
10	Matamata	**0**	423	180	1	50	0
11	Matamata	**37**	814	17	0	52	0
12	Matamata	**0**	487	1156	0	10	2

*Soil with putative suppression of *Spongospora* diseases, as indicated in previous studies.

Table A1.2. Phase 1 greenhouse experiment results. Mean numbers of root galls per plant (16 October 2017), and mean marketable tuber yields and mean proportions (%) of tubers with >5% of surface area affected by powdery scab (18 December 2017), for cv. 'Agria' potato plants grown in 12 different field soils. For each soil, half of the plants were uninoculated, and the other half were inoculated with *Spongospora subterranea* at planting. Some of the soils (highlighted) contained *Spongospora* DNA prior to planting the trial (Predicta® Pt tests).

		Mean number root galls/plant		Mean percent tubers with >5% of surface area affected by powdery scab		Mean weight (g/plant) of "marketable" tubers (>50 g)	
Soil	Region	Uninoculated	Inoculated	Uninoculated	Inoculated	Uninoculated	Inoculated
1	Pukekohe	0	5	0	17	1,311	1,242
2	Pukekohe	0	17	0	18	1,297	1,180
3	Pukekohe	0	7	0	14	1,727	1,343
4*	Pukekohe	0	1	3	3	989	867
5	Pukekohe	0	11	33	39	809	1,100
6	Pukekohe	0	2	15	25	1,342	1,274
7	Pukekohe	0	0	0	2	1,196	1,159
8	Pukekohe	0	1	0	2	1,468	1,236
9	Matamata	0	4	0	9	1,502	1,356
10	Matamata	0	4	0	0	1,091	1,001
11	Matamata	0	2	0	0	874	641
12	Matamata	0	1	0	0	1,297	1,200

*Soil with putative suppression of *Spongospora* diseases, as indicated in previous studies.

Phase 2

Table A1.3. Phase 2 Predicta® Pt test results. Mean amounts (pg/g of soil) of DNA of *Spongospora subterranea* and other soilborne potato pathogens in samples of six different cropping soils from the Pukekohe and Matamata regions, New Zealand. Mean amounts (pg g^{-1} of soil) of DNA of soilborne potato pathogens in samples of the six field soils (A–F) used in the greenhouse experiment, as measured in Predicta Pt assessments.

Soil	***Spongospora subterranea***	*Colletotrichum coccodes*	*Verticillium dahliae*	*R. solani AG-2.1*	*R. solani AG-3*	*Streptomyces scabies*
A	**13**	32	4	3	0	0
B	**13**	0	5	0	0	0
C	**21**	66	52	0	0	0
D	**488**	4	26	0	0	0
E	**5**	0	7	0	0	0
F	**26**	8	6	0	0	0

Table 1.4. Phase 2 greenhouse experiment results. Mean numbers of *Spongospora* root galls (12 weeks after planting) on potato plants grown in six field soils (A–F) to which four different treatments had been applied at the time of seed tuber planting.

	Treatment			
Soil	'Not inoculated'. No *Spongospora* inoculum	'Inoculated'. *Spongospora* inoculum added	'Heat treated' Heat-treated soil, *Spongospora* inoculum added	'Conducive' 90% conducive soil + 10% Treatment soil, *Spongospora* inoculum added
A	0.0	12.2	19.3	27.0
B	0.0	2.7	4.0	71.0
C	2.2	10.8	6.8	81.4
D	0.7	2.5	16.5	31.8
E	2.0	5.2	5.2	120.2
F	4.8	3.8	24.7	34.8

Table A1.5. Phase 2 greenhouse experiment results. Mean proportions (%) of potato tubers with powdery scab severity >5% of tuber surface affected (17 weeks after planting) from potato plants grown in six field soils (A–F) to which four different treatments had been applied.

	Treatment			
Soil	'Not inoculated' No *Spongospora* inoculum	'Inoculated' *Spongospora* inoculum added	'Heat treated' Heat-treated soil, *Spongospora* inoculum added	'Conducive' 90% conducive soil + 10% Treatment soil, *Spongospora* Inoculum added
A	0	4.2	18.2	8.0
B	0	2.1	31.2	22.3
C	0	0	6.5	23.1
D	15.0	3.3	40.7	9.6
E	0	7.1	27.1	18.5
F	0	0	25.9	23.4

Appendix 2
Greenhouse experiment to measure effects of soil and/or foliar-applied manganese on development of powdery scab (Phase 3)

Soil samples were collected (July 2019) from 23 arable-vegetable cropping fields in the South Auckland, Waikato and Manawatu/Wanganui regions of the North Island of New Zealand. The fields were all cultivated and weed-free at time of sample collection. These soil samples were assessed for micronutrient contents (Hill Laboratories Ltd, Hamilton, New Zealand).

Twelve of the fields soils were selected for the Phase 3 greenhouse experiment (see below). **Table A2.1** summarises the micronutrient analysis data from these soils. The soils covered a broad spectrum of natural manganese (Mn) contents, from low to very high, based on Hill Laboratories 'medium range'. The soils also contained different amounts of zinc and boron, elements that have been shown in previous research to affect *Spongospora subterranea* infection of potato.

Table A2.1. Amounts of manganese, boron, zinc, copper, cobalt and iron in pre-planting samples of 12 cropping soils from different regions in the North Island of New Zealand, used in the Phase 3 greenhouse trial. Hill Laboratories general soil 'medium ranges' are shown.

Soil	District	**Mn (mg/kg)**	B (mg/L)	Zn (mg/kg)	Cu (mg/kg)	Co (mg/kg)	Fe (mg/kg)
1	Pukekohe	**21**	0.90	13.2	22.4	<0.2	262
2	Raetihi	**25**	1.22	5.7	5.8	0.2	236
3	Waiuku	**24**	1.45	24.0	5.4	<0.2	183
4	Glenbrook	**76**	0.83	5.9	3.6	0.5	279
5	Waiuku	**135**	1.23	3.2	5.6	0.9	196
6	Bombay	**214**	1.22	3.7	4.1	0.5	216
7	Onewhero	**225**	0.89	4.9	7.9	0.4	159
8	Matamata	**271**	1.06	20.6	5.7	0.8	196
9	Bombay	**421**	1.27	4.1	5.5	1.6	135
10	Matamata	**450**	1.83	30.2	10.1	1.0	156
11	Pukekohe	**630**	1.24	10.8	7.3	0.9	113
12	Pukekawa	**885**	0.84	4.8	5.7	2.0	227
Hill Laboratories 'medium range'		**50–400**	1.0–2.0	2–10	1.0–5.0	2.0–4.0	

Samples of the 12 selected soils were also assessed for their fertility profiles, and for soilborne potato pathogens (including *S. subterranea*) using Predicta® Pt tests. **Tables A2.2 and A2.3** summarise the results obtained from these analyses. The pHs ranged from 6.1 to 7.8, and the soils had different contents of organic matter (3.4 to 23.5%), total carbon (2.0 to 13.6%), total nitrogen (0.14 to 0.94%), phosphorus (Olsen P, 18 to 165 mg/L), and potassium (0.26 to 1.61 me/100 g). The soils also contained different amounts of DNA of potential potato pathogens, including *S. subterranea*.

Table A2.2. Soil description, organic matter contents and chemical parameters for 12 cropping soils from different regions in the North Island of New Zealand (see **Table A1.1**), used in the Phase 3 greenhouse trial. Hill Laboratories 'medium ranges' are shown.

Soil	Soil description	Organic matter (%)	pH	Total C (%)	Total N (%)	Olsen P (mg/L)	K (me/100g)
1	Poorly drained loamy peat	23.5	6.7	13.6	0.94	58	0.92
2	Well drained loam	13.6	6.1	7.9	0.64	18	0.26
3	Well drained clay loam	12.2	7.0	7.2	0.60	32	1.61
4	Well drained clay loam	9.3	6.2	5.4	0.44	27	1.39
5	Imperfectly drained clay	6.9	6.8	4.0	0.35	53	1.26
6	Imperfectly drained clay	3.4	7.8	2.0	0.14	165	1.12
7	Well drained clay	10.9	6.5	6.3	0.61	33	0.92
8	Well drained loam over sandy loam	7.8	6.7	4.5	0.47	84	1.52
9	Moderately well drained clay	4.0	7.2	2.3	0.19	48	1.09
10	Moderately well drained loam over sandy loam	8.6	6.5	5.0	0.50	57	1.41
11	Imperfectly drained clay	5.8	7.4	3.4	0.29	100	1.33
12	Moderately well drained clay	4.3	6.3	2.5	0.21	57	0.86
	Hill Laboratories 'medium range'	7–17			0.3–0.6	30–60	0.5–1.0

Table A2.3. Mean amounts of DNA (pg/g soil) of *Spongospora subterranea*, *Colletotrichum coccodes*, *Verticillium dahlia*e, *Rhizoctonia solani* (AG-2.1 and AG-3), and *Streptomyces scabies* in pre-planting samples from 12 cropping soils from different regions in the North Island of New Zealand (see **Table A1.1**). Data provided from the Predicta® Pt soil testing service, The South Australian Research and Development Institute.

Soil	Mean amounts of DNA (pg/g soil)					
	S. subterranea	*C. coccodes*	*V. dahliae*	*R. solani* AG-2.1	*R. solani* AG-3	*S. scabies*
1	**200**	214	24	12	0	6
2	**4**	32	0	1	0	0
3	**0**	140	2	154	0	13
4	**0**	0	1	0	0	0
5	**0**	75	0	0	0	0
6	**0**	138	8	0	0	0
7	**0**	1220	3	288	0	0
8	**0**	15	0	0	0	22
9	**18**	27	6	0	0	0
10	**0**	118	33	0	0	0
11	**156**	670	190	0	1	1
12	**349**	5	11	0	0	0

The aims of the greenhouse experiment were:

1. determine if different natural soil amounts of Mn affected potato growth, and development of powdery scab;
2. assess effects of soil or foliar applications of Mn on potato plants, and development of powdery scab; and
3. determine effects of soil chemical and physical factors on Mn-*Spongospora* relationships.

Methods

For each of the 12 selected fields, 24 polythene pots (35 L capacity) were filled with soil, then taken to a research greenhouse (set with auto-venting at 15°C). The 288 pots (see **Figure 1**) were arranged in a randomized block experimental design (with six replicates), with the experimental treatments being the 12 soils, and four *S. subterranea*/Mn treatments. The *S. subterranea*/Mn treatments were:

1. No inoculation ('No Mn + not inoculated'; experimental control).
2. *Spongospora subterranea* inoculation ('No Mn + inoculated').
3. Mn pre-plant soil application + *S. subterranea* inoculation ('Soil Mn + inoculated').
4. Mn soil application + Mn foliar applications + *S. subterranea* inoculation ('Soil Mn + Foliar Mn + inoculated').

The inoculation (including nil) and Mn treatments were applied either at the time of seed tuber planting (inoculation and soil Mn applications), or during plant growth (foliar Mn applications).

On 15 August 2019, each pot was planted with a disease-free seed tuber (cv. 'Agria'; very susceptible to powdery scab). *Spongospora subterranea* inoculum (20 mL of sporosorus suspension in water + Tween 20; approx. 1.75×10^7 resting spores) was applied to each inoculated pot to the soil at the bottom of the seed tuber planting hole. The excavated planting hole soil from each pot was placed in a 9 L bucket, where fertiliser (15% potassic superphosphate (N = 0, P = 6.3, K = 15, S = 7.4, Ca = 15.4) at 1.5 t/ha) was mixed with the soil. Manganese microgranules were added for the Mn soil treatments. This ensured that each seed tuber was surrounded by the base fertilisers and Mn microgranules. The soil Mn treatment was at the equivalent of 1.04 kg Mn/ha, using chelated manganese microgranules (YaraVita™ Rexolin® Mn 13). The first foliar treatment of Mn was applied to potato plants at 6 weeks after planting, and this was followed by three further applications at 3 to 4 week intervals. The foliar applications were at the equivalent 1 kg Mn/ha (2 L/ha of YaraVita™ Mantrac™ Pro (50% Mn) in 200 L/ha of water). The Mn products and advice on application were provided by Yara Fertilizers (New Zealand) Ltd.

Crop management **Figure A2.1** shows the greenhouse experiment at two stages of growth. The pots were irrigated to field capacity immediately following planting, then not again until early plant emergence. From that point, the pots were irrigated (25 mm) once each week until tuber initiation (5 October 2019), then twice a week (20 mm each time) until onset of plant senescence (22 December: 1 week before harvest). Nitrogen (equiv. 250 kg/ha) was applied as calcium ammonium nitrate (27% N) in equal split applications at 6 weeks and 9 weeks after planting. The pots were hand-weeded, and insecticides were applied as required during the growing season.

Nutrient analyses of plant foliage On 6 November 2019, two recently-matured compound leaves were removed from near the top of each plant. The leaves were combined into one sample for each Soil × Mn treatment, and sent to Hill Laboratories for analyses of plant nutrient elements, including Mn (**Table A2.4**).

Table A2.4. Pre-planting soil Mn concentrations (mg/kg), and leaf Mn concentrations (mg/kg) for growing potato plants, for 12 field soils to which different Mn treatments were applied.

		Mn concentrations in leaves (mg/kg)		
		Treatments		
Soil	**Pre-planting soil Mn**	No Mn applications	Pre-plant soil Mn application	Pre-plant soil + foliar Mn applications
1	21	90	153	1580
2	25	210	117	2500
3	24	106	35	2700
4	76	166	151	3100
5	135	148	81	3500
6	214	98	85	2000
7	225	178	150	2100
8	271	178	200	2200
9	421	139	109	3500
10	450	220	250	2700
11	630	126	121	5000
12	885	210	230	3900

Figure A2.1. The greenhouse pot trial, on 2 October 2019 (top) and 22 October (bottom). Pot rim colours indicate the 12 different field soils, and the coloured ribbons indicate the four Mn treatments.

Manganese deficiency symptoms in leaves normally occur where Mn leaf concentrations are less than 20 mg/kg, and are particularly severe at less than 10 mg/kg. Healthy plant leaves normally contain 50 to 200 mg/kg of Mn, although levels up to 1,500 mg/kg have been recorded where Mn-containing fungicides (e.g. mancozeb), or Mn foliar fertilisers (as used in the present study) have been applied.

None of the plants in the experiment were deficient in Mn, with the Mn concentrations in the foliage from the plants receiving no added Mn ranging from 90 to 220 mg/kg for the 12 soils. Soil applications of Mn had the greatest increasing effect for Soil 1 (which had the lowest amount of 'natural' Mn), increasing the concentration of Mn from 90 mg/kg to 153 mg/kg. For the other 11 soils, foliar applications gave little, or mixed, responses. In most cases the soil applications reduced the amounts of Mn in foliage. The foliar Mn applications greatly increased the amounts of Mn in assayed the leaf samples.

Assessments during plant growth The plants were inspected every 2 weeks for unusual foliar symptoms, particularly for Mn toxicity as indicated by formation of pale green leaves and necrotic spots on petioles and stem. Plant heights and overall plant health and vigour were recorded. No differences in plant vigour or health were observed between the four Mn treatments for each of the 12 soils, and no symptoms of Mn toxicity were seen in the plants, including those that received foliar Mn applications.

Tuber yields Tubers were harvested on 5 January 2020. The tubers were size-graded into 'marketable' (>50 g) and 'reject' (<50 g), and the number and weight of 'marketable' tubers from each pot were recorded. There were no appreciable differences in tuber yields between the four Mn treatments for each of the 12 soils.

Incidence and severity of powdery scab The washed tubers (**Figure A2.2**) were assessed for incidence and severity of powdery scab. Severity for each tuber was assessed using a 0–10 disease severity score (**Figure A2.3**). A disease score of >1 (**Figure A2.3**; 5% tuber surface affected) is unacceptable for washed potatoes, fresh-market potatoes.

Figure A1.2. Left. Washed tubers from the 288 pots in the experiment. Right. Tubers with severe powdery scab, from one pot.

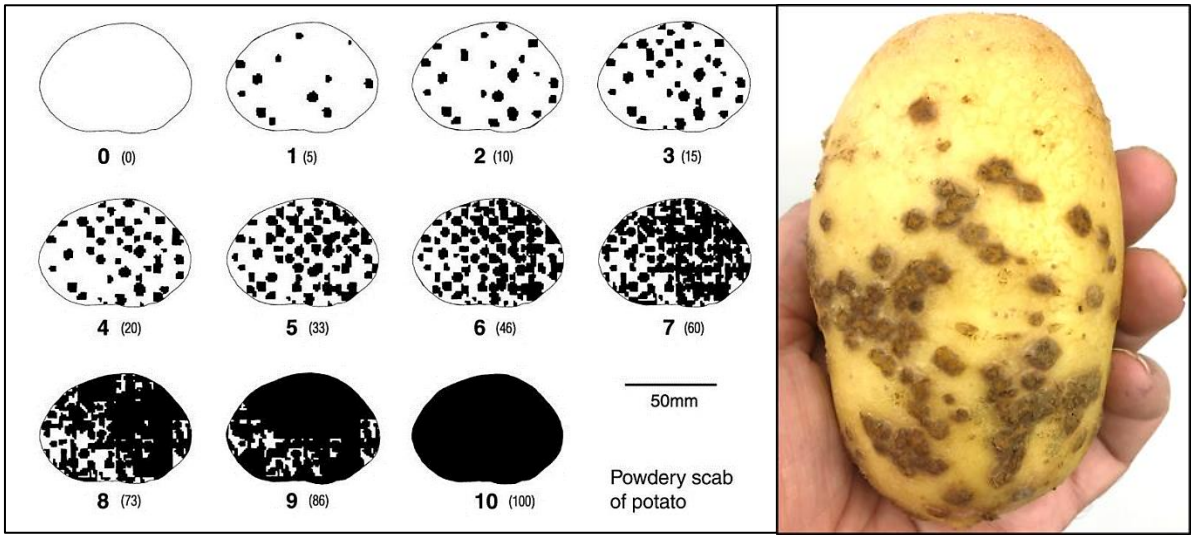


Figure A1.3. Left, powdery scab severity key (Falloon et al., 1995. *NZ J Crop & Hort Sci* 23, 31-37). Disease scores and proportions (%) of tuber surface affected are indicated. Right, a washed tuber with severity score 4.

Results

Powdery scab on harvested tubers

For the 'No Mn + not inoculated' treatment, only four soils (Soils 1, 4, 11 and 12) produced tubers with powdery scab at harvest (**Table A1.5**). This indicated that these soils contained resident *S. subterranea* inoculum, as was indicated (except for Soil 4) from the pre-planting Predicta® Pt tests (**Table A1.3**). The tubers harvested from Soil 12 had much more severe powdery scab than tubers from the other 11 soils (**Table A1.5**). Soil 12 also gave the greatest incidence of powdery scab on tubers (**Table A1.6**), and greatest proportions of tubers with powdery scab scores of >1 (**Table A1.7**).

Compared with the 'No Mn + not inoculated' treatment, the 'No Mn + inoculated' treatment increased the mean severity scores in all 12 soils (**Table A1.5**), and mean proportions of tubers with powdery scab (**Table A1.6**). Based on mean proportions of tubers with powdery scab score >1 (**Table A1.7**), the 'No Mn + inoculated' treatment of the most powdery scab-conducive soils were Soil 12 (47% tubers with severe disease), Soil 4 (10%),

Soil 1 (6%), Soil 2 (3%), and Soil 5 (3%).

The other *S. subterranea*-inoculated soils all gave no tubers with powdery scab score >1, and although they all had 3% to 11% tubers with minor (1% to 4%) powdery scab (score <1) (**Table A1.6**), these soils suppressed the disease.

Table A1.5. Mean powdery scab scores for tubers harvested from 12 field soils receiving different manganese and *Spongospora subterranea* inoculation treatments.

	Treatment			
Soil	No Mn + not inoculated	No Mn + *S. subterranea* inoculated	Soil Mn + *S. subterranea* Inoculated	Soil Mn + foliar Mn + *S. subterranea* inoculated
1	0.18	0.36	0.06	0.06
2	0.00	0.24	0.08	0.00
3	0.00	0.09	0.06	0.09
4	0.04	0.44	0.52	0.16
5	0.00	0.03	0.03	0.00
6	0.00	0.13	0.14	0.12
7	0.00	0.05	0.07	0.08
8	0.00	0.10	0.03	0.03
9	0.00	0.08	0.06	0.11
10	0.00	0.03	0.06	0.06
11	0.03	0.11	0.06	0.06
12	1.41	1.88	2.16	1.90

Table A1.6. Mean incidence (percent) of tubers with powdery scab, for tubers harvested from 12 field soils receiving different manganese and *Spongospora subterranea* inoculation treatments.

	Treatment			
Soil	No Mn + not inoculated	No Mn + *S. subterranea* inoculated	Soil Mn + *S. subterranea* inoculated	Soil Mn + foliar Mn + *S. subterranea* inoculated'
1	15.2	27.6	6.0	5.7
2	0.0	17.8	7.5	0.0
3	0.0	8.9	5.4	9.0
4	4.2	27.4	28.8	11.4
5	0.0	2.8	2.8	0.0
6	0.0	10.7	11.7	11.5
7	0.0	4.6	7.4	7.5
8	0.0	9.7	3.3	3.3
9	0.0	8.3	6.3	11.1
10	0.0	2.8	4.2	6.1
11	3.3	11.1	5.7	5.7
12	59.7	71.6	85.4	80.2

Table A1.7. Mean severity (percent) of tubers with powdery scab score greater than 1 (>5% tuber surface area affected), for tubers harvested from 12 field soils receiving different manganese and *Spongospora subterranea* inoculation treatments.

	Mn application and *S. subterranea* inoculation treatments			
Soil	No Mn + not inoculated	No Mn + *S. subterranea* inoculated	Soil Mn + *S. subterranea* inoculated	Soil Mn + foliar Mn + *S. subterranea* inoculated
1	3.3	5.7	0.0	0.0
2	0.0	3.3	0.0	0.0
3	0.0	0.0	0.0	0.0
4	0.0	10.0	13.0	2.1
5	0.0	0.0	0.0	0.0
6	0.0	2.8	2.1	0.0
7	0.0	0.0	0.0	0.0
8	0.0	0.0	0.0	0.0
9	0.0	0.0	0.0	0.0
10	0.0	0.0	4.2	0.0
11	0.0	0.0	0.0	0.0
12	30.9	47.0	59.3	53.7

Relationships between powdery scab severity and soil factors

The pre-planting soil Mn treatment, and the soil + foliar Mn treatment both reduced proportions of tubers with powdery scab score >1, for the *S. subterranea*-inoculated plants grown in the two low Mn Soils 1 and 2 (**Table A1.8**). Soils 4 and 6 had reduced proportions of tubers with this disease severity from the soil + foliar Mn treatment. However, for Soil 12, which had the greatest amount of pre-planting Mn, additional applications of this element increased the proportion of harvested tubers with powdery scab score >1. These results indicate that the soil + foliar Mn treatments reduced the disease in the low Mn soils (especially for the soils with less than 50 mg Mn/kg), but not for the soils with greater amounts of resident Mn.

Table A1.8. Mean severity (percent) of tubers with powdery scab score greater than 1 (>5% tuber surface area affected), for tubers harvested from 12 field soils with different pre-planting soil Mn concentrations, to which different manganese treatments were applied.

Soil	**Pre-planting soil Mn (mg/kg)**	**No added Mn**	**Soil applied Mn**	**Soil + foliar applied Mn**
1	21	**5.7**	0.0	0.0
2	25	**3.3**	0.0	0.0
3	24	0.0	0.0	0.0
4	76	**10.0**	**13.0**	**2.1**
5	135	0.0	0.0	0.0
6	214	**2.8**	**2.1**	0.0
7	225	0.0	0.0	0.0
8	271	0.0	0.0	0.0
9	421	0.0	0.0	0.0
10	450	0.0	**4.2**	0.0
11	630	0.0	0.0	0.0
12	885	**47.0**	**59.3**	**53.7**

Organic matter content and total carbon and nitrogen did not affect powdery scab severity. However, soil pH less than 6.4 influenced the disease, as Soils 2, 4 and 12 with pHs less than 6.4 had 3% (Soil 2), 10% (Soil 4 and 47% (Soil 12) of tubers with powdery scab scores >1 (**Table A1.9**). Phosphorous and potassium levels below 'medium range' in Soils 2 and 4 may also have increased susceptibility of tubers to powdery scab.

Table A1.9. Mean proportions (%) of harvested potato tubers with powdery scab score >1 (5% tuber surface affected), amounts of pre-planting *Spongospora subterranea* DNA (pg/g of soil), and results from micronutrient and fertility parameters (including arbitrary Mn-B-Zn score) for 12 field soils, from the **'No Mn + not inoculated'** treatment. Low soil concentrations of micronutrients are: Mn <100 mg/kg, B <1 mg/kg, and Zn = <10 mg/kg.

Soil	Percent tubers with score >1	Pre-planting *S. subterranea* DNA	Mn (mg/kg)	B (mg/kg)	Zn (mg/kg)	Mn-B-Zn Score (out of 3)	pH	Olsen P (mg/L)	K me/100g)
1	**5.7**	200	21	0.90	13.2	**1**	6.7	58	0.92
2	**3.3**	4	25	1.22	5.7	**1**	6.1	18	0.26
3	0.0	0	24	1.45	24.0	**2**	7.0	32	1.61
4	**10.0**	0	76	0.83	5.9	**0**	6.2	27	1.39
5	0.0	0	135	1.23	3.2	**2**	6.8	53	1.26
6	**2.8**	0	214	1.22	3.7	**2**	7.8	165	1.12
7	0.0	0	225	0.89	4.9	**1**	6.5	33	0.92
8	0.0	0	271	1.06	20.6	**3**	6.7	84	1.52
9	0.0	18	421	1.27	4.1	**2**	7.2	48	1.09
10	0.0	0	450	1.83	30.2	**3**	6.5	57	1.41
11	0.0	156	630	1.24	10.8	**3**	7.4	100	1.33
12	**47.0**	349	885	0.84	4.8	**1**	6.3	57	0.86
Hill Laboratories 'medium range'			50–400	1.0–2.0	2–10			30–60	0.5–1.0

The micronutrients manganese, zinc and boron have been shown previously to affect *S. subterranea* infection of potato. These elements affected powdery scab severity in this experiment. **Table A1.9** summarises the pre-planting concentrations of Mn, boron (B) and zinc (Zn) in the 12 soils. The amounts of pre-planting *S subterranea* DNA are also presented, along with the proportions of harvested tubers with powdery scab scores >1 (5% tuber surface covered by scabs). 'Desirable' concentrations (mg/kg) for each element have been categorised, as: Mn, >100 mg/kg; B, >1.00 mg/kg; and Zn, >8.0 mg/kg (see **Table A1.1** for Hill Laboratories 'medium range' for these elements). **Table A1.9** shows that soils with Mn-B-Zn scores of 0 to 1 favoured/predisposed potato tubers to powdery scab. Mn-B-Zn scores of 2, and especially 3, gave less powdery scab, even where pre-planting *S. subterranea* DNA was present (Soil 11). It was also important that for the soil P and K concentrations to be within the recommended 'medium' ranges.

Tuber yields

Mean yields of 'marketable' tubers ranged from 0.52 to 1.60 kg/plant (**Table A1.10**). Soil 8 gave the greatest yields (overall mean = 1.49 kg/plant), while Soils 11, 5, 2 and 10 gave the lowest yields (overall means = 0.64 to 0.66 kg/plant). To convert these per plant yields to yield/ha, a multiplication factor of ×44,400 can be applied, assuming, for potato crops, 0.3 m between plants within rows and 0.75 m between rows. Using this multiplication factor, per plant tuber yields were: for Soil 8, equivalent to 66.2 tonnes/ha; and for Soils 11, 5, 2, and 10, equivalent to 28.4 to 29.7 tonnes/ha.

The low yielding Soil 2 (overall mean = 0.66 kg/plant) had very low pre-planting Mn content (25 mg/kg), while the similarly low-yielding Soil 11 had very high pre-planting Mn (630 mg/kg). The greatest plant yield (overall mean = 1.49 kg/plant) was from Soil 8, which contained an intermediate amount of Mn (271 mg/kg). Comparing pre-planting soil Mn levels in the 12 soils with resulting plant tuber yields strongly indicated that the yields were not affected by soil Mn.

The different Mn/*S. subterranea* inoculation treatments gave, overall, very similar mean plant yields (0.91 to 0.96 kg/plant). This indicated that neither *S. subterranea* inoculation nor soil-applied Mn affected plant yields. The 'Soil Mn + foliar Mn + inoculation' treatment gave the lowest overall mean tuber yield (0.91 kg/plant).

Table A1.10. Mean weights (kg) of 'marketable' potato tubers (>50 g) harvested from potato plants grown in 12 field soils containing different pre-planting amounts of manganese, to which different *Spongospora subterranea* inoculation and manganese application treatments were applied.

Soil	Pre-planting soil Mn (mg/kg)	Mn application and *S. subterranea* inoculation treatments				Overall mean
		No Mn + not inoculated	No Mn + inoculated	Soil Mn + inoculated	Soil Mn + foliar Mn + inoculated	
1	**21**	1.03	1.14	1.22	0.93	**1.08**
2	**25**	0.64	0.62	0.76	0.62	**0.66**
3	**24**	1.14	0.98	0.75	1.00	**0.97**
4	**76**	1.41	1.33	1.38	1.09	**1.30**
5	**135**	0.53	0.76	0.71	0.59	**0.65**
6	**214**	1.22	1.03	1.07	0.97	**1.07**
7	**225**	1.05	1.13	1.13	0.99	**1.07**
8	**271**	1.60	1.54	1.49	1.33	**1.49**
9	**421**	0.74	0.76	0.86	1.00	**0.84**
10	**450**	0.54	0.61	0.72	0.79	**0.67**
11	**630**	0.65	0.80	0.52	0.60	**0.64**
12	**885**	0.93	0.94	1.06	0.96	**0.98**
	Overall mean	**0.96**	**0.97**	**0.97**	**0.91**	

Conclusions

The aims of the experiment were:

- to determine if different amounts of natural soil Mn affected potato growth, and development of powdery scab;
- to assess effects of soil or foliar applications of Mn on potato plants and development of powdery scab; and
- to determine effects of soil chemical and physical factors on *S. subterranea*/Mn relationships.

The experiment provided information addressing the following questions:

Does the amount of natural Mn in field soil affect development of powdery scab?
Low soil Mn concentrations (less than 100 mg/kg) gave more severe powdery scab than greater amounts of this element (see **Tables A1.8** and **A1.9**).

Do soil or potato foliar applications of Mn affect development of powdery scab?
Soil applications Addition of Mn-containing microgranules to the soil at planting reduced powdery scab, but only for soils containing very low amounts of natural Mn (less than 30 mg/kg) (see **Table A1.8**).
Foliar applications Foliar Mn applications, in addition to soil Mn applications, reduced powdery scab in some of the field soils, but not in others (see **Table A1.8**).

*Do soil chemical and physical factors affect Mn/*S. subterranea *relationships?*
Soil pH, and contents of phosphorus, potassium, boron and zinc probably affected the Mn/*S. subterranea* relationship. However, soil contents of organic matter, total carbon and total nitrogen had little effect on this micronutrient/pathogen relationship.

Do soil and/or foliar applications affect potato plant growth of tuber yields?
At the rates tested in this experiment, adding Mn to 12 different field soils before seed tuber planting did not adversely affect yields of harvested tubers (see **Table A1.10**). Pre-planting soil application plus four foliar applications also did not adversely affect tuber yields.

Appendix 3

Popular article for submission to *Potatoes Australia* and *NZ Grower*

Research on soils that suppress *Spongospora* root galling and powdery scab of potato

Powdery scab is an economically important disease of potato crops in Australia, New Zealand and worldwide, because the lesions on tubers diminish the quality and marketability of seed, fresh market and processing potatoes (**Figure A2.1**). *Spongospora subterranea* (the powdery scab pathogen) also invades potato underground stems (**Figure A2.2**), stolons and roots, forming galls. These infections obstruct water and nutrient uptake, reducing tuber growth and decreasing crop yields.

Spongospora diseases begin early in crop growth, with release of zoospores from resting spores in contaminated soils, and from infected seed tubers. The zoospores infect potato roots, where the pathogen multiplies, and produces many more zoospores that further infect roots and developing tubers. Later in the season, very many thick-walled survival structures (sporosori, each containing many resting spores), are produced in root galls, and in scab lesions on mature tubers. The resting spores can survive in soil for several years, making it difficult to control powdery scab. Disease management recommendations include long crop rotations between potato crops, pre-planting assessment of *Spongospora* in soils, and use of disease-tolerant cultivars, pathogen-free seed tubers and appropriate crop management.

Disease suppressive soils give low amounts of diseases, even where pathogen inoculum levels are high. Naturally suppressive soils have been reported for several soilborne plant diseases. Soil physical and nutrient properties, and/or microbial populations, can contribute to disease suppression. Soil sterilisation makes soils with microbe-mediated suppression disease-conducive. A long-term crop rotation trial in the Pukekohe vegetable growing region near Auckland, New Zealand, indicated that the trial site was powdery scab-suppressive, even when successive potato crops were grown for 9 years. This inferred that identifying, maintaining and enhancing suppressive soils could be a sustainable strategy for managing *Spongospora* diseases.

A research project was instigated with goals to determine if different field soils had different powdery scab suppressive capabilities, and to identify factors involved in disease suppression. The project included scientists with expertise in plant pathology, soil science, molecular biology and microbial bioinformatics, working at The New Zealand Institute for Plant and Food Research Limited. Three large greenhouse experiments (e.g. **Figure A2.3**) were conducted during the growing seasons of 2017/18 (Phase 1), 2018/19 (Phase 2) and 2019/20 (Phase 3).

Each experiment included different field soils (up to 12), which were collected in July of each year and transported to a research greenhouse. Samples of the soils were analysed for fertility parameters and soil microbial activity, and for different potato pathogens (including *S. subterranea*), using the Predicta® Pt service (South Australia Research and Development Institute). Microbial profiles of the soils were also determined, using gene sequencing of extracted bacterial and fungal DNA.

In each experiment, cv. 'Agria' seed tubers (very susceptible to powdery scab and root galling) were individually planted into large pots (Figure 3). Half of the pots were inoculated at planting with *S. subterranea*, and the others were left uninoculated (experimental controls). The pots were watered regularly to give conditions suitable for *Spongospora* infection. At 2 to 3 months after planting, half of the plants (in **Phase 1** and **2**; below) were assessed for severity of root galling, and microbial DNA from rhizosphere soil was also analysed. At maturity (in **Phases 1, 2** and **3**), tubers from the remaining plants were assessed for incidence and severity of powdery scab, and tuber yields were determined.

The **Phase 1** experiment showed that *Spongospora* inoculation of six of the soils increased severity of powdery scab on harvested tubers, but inoculation gave much less disease in the other six soils. Three soils with low clay contents (<30%) gave very little powdery scab in harvested tubers, which was likely because of their free-draining nature. Soil pH and nutrient concentrations were not associated with powdery scab incidence or severity. With the exception of one disease-conducive soil already infested with *S. subterranea* and containing high organic matter (OM), soil OM levels were not related to powdery scab suppression. The effect of crop rotations on powdery scab was variable, but one soil with three previous potato crops in the last 5 years was disease suppressive.

Although pre-plant soil microbial communities were affected by soil type and cropping history, there were no obvious relationships between *Spongospora* diseases (root galling or powdery scab) and microbial communities of the 12 soils.

The **Phase 2** experiment focused on six Pukekohe soils, of one soil type and all from within a 10 km zone. The experiment aimed to determine if powdery scab suppression was 'general' or 'specific' (transferable; involving specific microorganisms), and was associated with soil physical, chemical or biological factors (including particular bacterial or fungal groups). Four treatments involving the six soils were applied, including: no added *S. subterranea* inoculum (experimental control); *S. subterranea* inoculation; inoculation + heat treatment; and a treatment to test transfer of suppression. This treatment, also with *S. subterranea* inoculation, used the six soils added separately to a known conducive soil (from **Phase 1**), at 10% of each soil to 90% of the conducive soil.

Previous crop rotation did not affect *Spongospora* diseases on the plants or harvested tubers. All six soils displayed some level of microbe-mediated disease suppression, since heat treatment increased severity of powdery scab on the harvested tubers. Three of the soils were more suppressive than the others. Two soils gave possible 'specific' *Spongospora* suppression, with less disease where they were added to the conducive soil. One soil that contained pre-planting *S. subterranea* DNA gave low powdery scab severity, indicating disease suppression. All six "suppressive" soils contained high levels manganese (Mn; 292–674 mg/kg), which were much greater than for the conducive soil (49 mg Mn/kg), suggesting a possible link between Mn content and disease suppression. Bacteria and fungi reported as being antagonistic to soilborne plant pathogens and/or promoters of plant growth were present in the disease suppressive soils, and may have contributed to powdery scab suppression.

The **Phase 3** experiment assessed effects of different natural soil levels of Mn, or soil- or foliage-applied Mn, on development of powdery scab on potato tubers, to test the relationship with Mn indicated in **Phase 2**. Twelve field soils, with natural Mn contents from low to very high (21 to 885 mg Mn/kg) were selected after micronutrient determinations of soil samples from 23 different fields. Four treatments were applied, including: no Mn applications and no *S. subterranea* inoculum at planting (experimental control) ('No Mn + not inoculated'); no Mn applications and added *S. subterranea* inoculum ('No Mn + inoculated'); Mn pre-plant soil application and added *S. subterranea* inoculum ('Soil Mn + inoculated'); and Mn soil application, Mn foliar applications, *S, subterranea* inoculation ('Soil Mn + foliar Mn + inoculated').

The pre-planting soil application of Mn was at the equivalent of 1.04 kg Mn/ha, using chelated Mn microgranules (YaraVita™ Rexolin® Mn 13 EDTA). The first foliar Mn treatment was applied to potato plants 6 weeks after planting, and was followed by three further applications at 3 to 4 week intervals. Each foliar application was at the equivalent 1 kg Mn/ha (2 L/ha of YaraVita™ Mantrac Pro™ (50% Mn) in 200 L/ha of water).

Five of the soils contained resident *S. subterranea* DNA. For the 'No Mn + not inoculated' treatment, powdery scab was severe in two soils and moderate in three others. The 'No Mn + inoculated' treatment to the other seven soils gave much less powdery scab, indicating suppression of the disease. The 'Soil Mn + inoculated' and 'Soil Mn + foliar Mn + inoculated' treatments both reduced powdery scab severity in two soils with low natural levels of Mn (21, 24 or 25 mg Mn/kg). However, for one soil which had the greatest amount of pre-planting *S. subterranea* DNA and the greatest amount of natural Mn (885 mg/kg), powdery scab was very severe on the harvested tubers. These results indicated that Mn soil and foliar applications to low Mn soils may reduce powdery scab, but not where natural *S. subterranea* inoculum levels are very high. The three very low Mn soils (<26 mg/kg) had the greatest OM contents (12 to 23.5%).

Conclusions

This project has demonstrated that some New Zealand field soils are suppressive to powdery scab. Abiotic soil factors (texture, OM content, pH, nutrient chemicals) influenced incidence and severity of *Spongospora* root galling and tuber powdery scab on potato plants. Applications of Mn to soil and potato foliage reduced powdery scab in naturally low Mn soils. Soil microorganisms were also likely to be involved in suppression of *Spongospora* diseases, because heat treatments of suppressive soils increased these diseases (eliminated or reduced disease suppression). This research has broadened understanding of the nature and possible causes of powdery scab suppression in field soils, and may contribute to future management strategies for the intractable quality- and yield-limiting diseases caused by *Spongospora subterranea*.

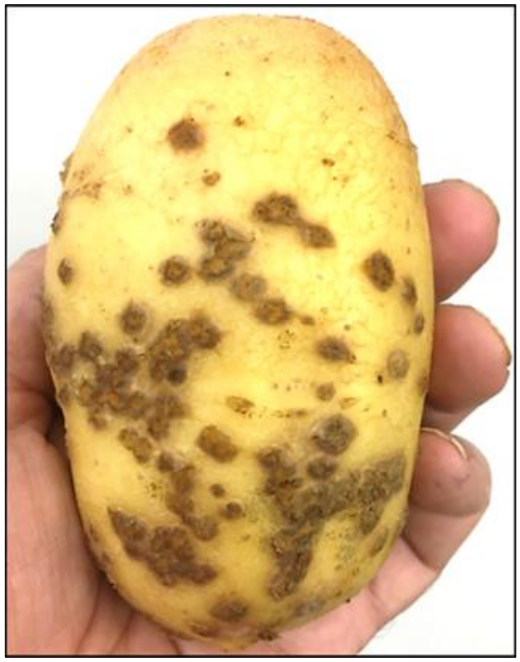


Figure A2.1. Powdery scab on an 'Agria' tuber from one of the experiments in this study (image, Peter Wright).

Figure A2.2. *Spongospora* galls on the underground stem of a field-grown potato plant (image courtesy Richard Falloon).

Figure A2.3. Greenhouse pot experiment, September 2018 (image courtesy Peter Wright).

Appendix 4

Evaluation of Potato Industry Pest and Disease Forum, Hort Connections 2019, Melbourne, Australia, 26 June, 2019.

[Provided by Dr Kristen Stirling, RCMG Consulting, Level 1 East, 1100-1102 Toorak Road, Camberwell, Victoria 3124]

RMCG

July 2019

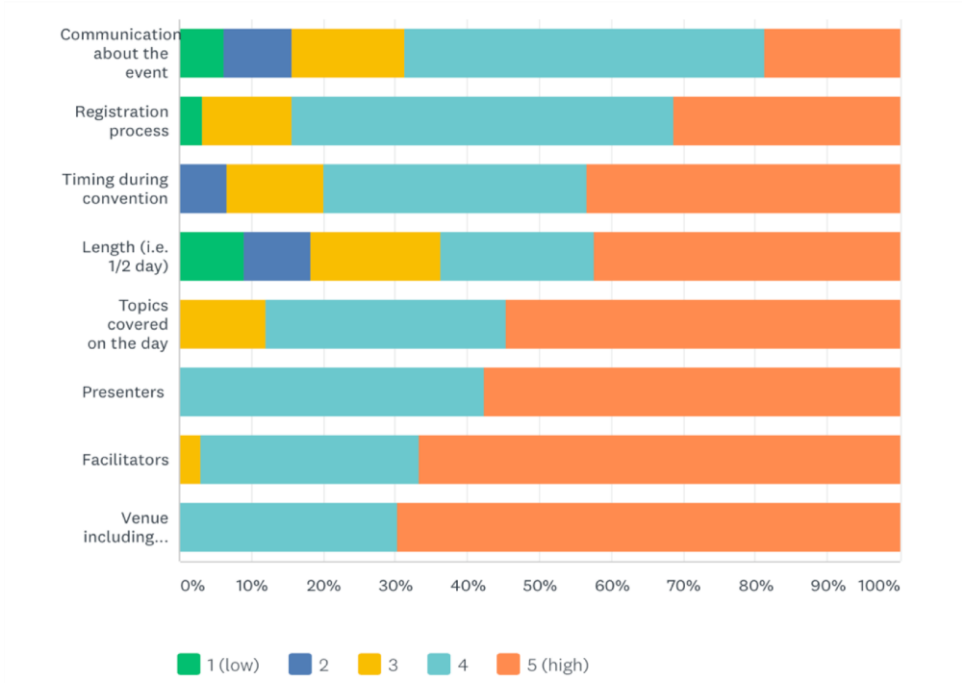
Introduction

The Potato Industry R&D Forum attracted a range of growers, service providers, and industry representatives eager to hear the most up to date information on a range of pest and disease related R&D and practical implications for the potato industry. The forum included presentations from international speakers, researchers, agronomists and growers on their experiences and R&D used to combat pests, diseases and biosecurity threats.

Of the 83 people that attended the event, 29 returned evaluation forms, providing an insight into how to run future events.

1.1 HOW DID YOU ENJOY THE FORUM?

Figure 1-1: Ratings by attendees of 2019 forum



Communication about the event

The majority of attendees were happy with communication about the event with 69% of responses rating communication as 4 or above. However, one attendee felt it should have been better communicated. The issue raised is largely due neither AUSVEG nor the Convention Centre advertising the event inside the building or in the program for the Convention (as it was held as a separate event). Other verbal feedback mentioned that the Forum was the only speaker event of value to the potato industry, apart from the trade exhibition.

Registration Process
Most attendees were satisfied with the registration process for the event with 84% of responses rating it 4 and above. One attendee provided the following comment:
"We had been at Hort Connections, we knew this event was on however there was no information on where or when. We discovered it by word of mouth and therefore arrived halfway through the session".
Unfortunately, the Convention Centre did not advertise where the Forum was being held on their notice boards inside the building and AUSVEG did not put the event into their program (booklet or app) as it was a parallel, separate event. This has been a learning and we will organise communication and registration differently for the 2020 event.

Timing during convention
80% of responses rated the timing of the Forum during the Convention as 4 or above.

Length of forum
64% of responses rated the length of the event as 4 or above. While many felt that the timing/length of the Forum was right, a number of attendees felt that a longer, up to one day Forum would have provided more time for Q&A.
"It felt very rushed, needed to have another session after lunch say an extra hour or so as the presentations felt very rushed and I feel there was many more questions/comments in the room that they didn't have time to answer".

Topics covered on the day
Most attendees were happy with the topics covered with 88% of responses rating topics covered on the day as 4 or above. Comments included:
- *Great range of topics and information.* *Great event - very informative!*

Presenters
100% of attendees rated the presenters as 4 or above. Comments included:
- *Great way to present current research to industry.*
- *Left me wanting more - loved listening - will have a lot to look up.*

Facilitators
Attendees were satisfied with the facilitators of the event with 97% of responses rating them as 4 or above. Comments included:
- *First time attending and found it very informative. Good breadth of audience from different parts of the industry to give very good feedback.*
- *Well run forum with a good cross section throughout the industry.*

Venue and catering
The majority of attendees were happy with a 100% of responses rating venue including catering as 4 or above.

1.2 WHAT OR WHOM SHOULD THE 2020 FORUM INCLUDE?

Feedback from attendees on the 2020 Forum is provided in Table 1-2.

Table 1-1: Comments related to 2019 Forum

SUBJECT	COMMENT
Format	▪ Great. ▪ Levy payers meeting to provide complete overview for potatoes working for whole of industry day. ▪ Same or similar. (2) ▪ More Q and A. ▪ Presentations. (2) ▪ Mix presenters – mini workshops to encourage networking. ▪ Good to have short presentations and time for questions. To break it up a bit you could consider having participants sitting at tables and then could debrief after each session in relation to what does this R&D mean to me and my region. ▪ Round tables.

Location	▪ Great. (3) ▪ At Hort Connections but try to include Potato SIAP group. ▪ At this conference or closely related time (4). ▪ SA. (2) ▪ Regional location and field visits to complement topics. ▪ Somewhere warm. ▪ City is central. (2) ▪ Can be anywhere. ▪ Melbourne. ▪ Tasmania / Victoria / WA. ▪ Possibly near trial / demo site.
People	▪ Great. ▪ More students. ▪ Dr Steve Johnson. ▪ Joseph Zady – Plant Pathologist.
Timing and length	▪ 9.00am to 2.00pm. ▪ Similar timing. ▪ More time for pest soil and pest discussion. ▪ Same time of year. ▪ Full day. (5) ▪ Half day. ▪ 15 minutes including questions. ▪ Good. ▪ Could be a bit longer, after lunch session. Good to have short presentations and time for questions. To break it up a bit you could consider having participants sitting at tables and then could debrief after each session in relation to what does this R&D mean to me and my region. ▪ I would have valued more time with presenters to explore more insights - e.g. Frank - wealth of knowledge - time limited. What knowledge hasn't been extended from previous investments? Need more time for R&D prioritisation. Great to network, thank you!

Topics	▪ More of the same, but more time per speaker. ▪ Seed certification and more agronomy activity. ▪ Water use efficiency of potato cultivation. ▪ Seed health PVY and PLRV. ▪ Innate gene or plant for disease. ▪ Blackleg. (4) ▪ Grower case studies, both seed and more. ▪ TPP, soil health (cover crop and lowering diseases). ▪ Soil biota interactions. ▪ Relevant issues. ▪ Precision agriculture for diseases. ▪ Disease control. ▪ Industry outlook collate sources of info for growers (blackleg). ▪ Biological - the future – solutions. ▪ Succession planning for farmers and researchers. ▪ Fungicide resistance/IPM weed control. ▪ Potential future research topics, not just current ones. ▪ Continue to build and maybe broaden topics if possible.
Other comments	▪ Great. ▪ Excellent program! ▪ Drone technology. ▪ Great talk and panel discussion. ▪ Workshop could complement farm visit. ▪ Good start. ▪ I don't know why and did not like the split up of states. I don't think it's a useful split of the room. We are already split geographically. ▪ Happy to think about these and discuss. ▪ If this event had nothing to do with Hort Connections, then more direct contact would have been great. We were under the impression it was a part of Hort Connections but there was nothing on the program on the app. ▪ I did not think having Chad Hutchison promoting a product was appropriate, it should be about funded research in Australia and not about selling stuff. ▪ Could work on getting the specifics of pests and diseases that require R&D rather than just black spot or root rot.

The feedback provided by attendees will be used to design the 2020 Forum. A range of topics mentioned in the feedback do not fit within the Pest and Disease R&D Coordination Program as they are not part of levy funded pest and disease R&D. However, there may be an opportunity to cover these topics in cooperation with the upcoming Potato Communication and Extension Program. Some topics may require R&D and thus could be communicated to Hort Innovation as an R&D need.

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