THESIS

DEVELOPMENT AND TESTING OF A GREENHOUSE ASSAY FOR SCREENING POTATO GERMPLASM FOR SUSCEPTIBILITY TO *Spongospora subterranea* f. sp. *subterranea* (POWDERY SCAB)

Submitted by

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Steve,
Thanks for being on my committee. Your help & support were greatly appreciated throughout the entire process. Also, I greatly appreciate you working on getting me an increase for my salary! Thankyou! Andrew

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WE HEREBY RECOMMEND THAT THE THESIS PREPARED UNDER OUR SUPERVISION BY ANDREW HOUSER ENTITLED **DEVELOPMENT AND TESTING OF A GREENHOUSE ASSAY FOR SCREENING POTATO GERMPLASM FOR SUSCEPTIBILITY TO *Spongospora subterranea* f.sp. *subterranea* (POWDERY SCAB)** BE ACCEPTED AS FULFILLING IN PART REQUIREMENTS FOR THE DEGREE OF MASTERS OF SCIENCE.

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ABSTRACT OF THESIS

DEVELOPMENT AND TESTING OF A GREENHOUSE ASSAY FOR SCREENING POTATO GERMPLASM FOR SUSCEPTIBILITY TO *Spongospora subterranea* f. sp. *subterranea* (POWDERY SCAB)

Potato resistance to powdery scab, caused by the protist *Spongospora subterranea* f. sp. *subterranea* (*S.s.s.*), has become extremely important in recent years due to the increased damage caused by this disease. Variable field conditions and the environmental sensitivity of the *S.s.s.* infection process in naturally infested fields have created difficulties in consistently evaluating potato germplasm resistance. A greenhouse assay for evaluating potato germplasm resistance which is consistent and compatible with field results was developed.

Two soil types, three inoculum levels, two inoculum sources and two soil moisture regimes were evaluated in a greenhouse for powdery scab severity using four potato cultivars. Soil temperature was maintained at an ideal range for powdery scab development as reported in current literature. Greenhouse results were then compared with three years of field data collected from field trials conducted in the San Luis Valley (SLV). Cultivars were evaluated for root galling, tuber lesion incidence, and severity. Soil with 50% sand, an inoculum level of one sporeball per gram of soil, and relatively high soil moisture (0-10 cbars) had the most consistent symptom expression when compared with field results.

This combination of factors was then tested in a greenhouse using fourteen potato cultivars which varied in levels of powdery scab susceptibility. A relative ranking system was also developed to compare greenhouse results with known SLV field results. Results demonstrated that a greenhouse assay can be used successfully for screening advanced potato germplasm for susceptibility to powdery scab.

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Ronald Merritt Houser

Who taught me the meaning of life and the joy that can come from scientific inquiry.

To my wife and son:
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CHAPTER 1: INTRODUCTION

Powdery scab, caused by the protist *Spongospora subterranea* (Wallir.) Lagerh. f. sp. *subterranea* Tomlinson *(S.s.s.)*, is a disease that affects the roots and tubers of potato plants (*Solanum tuberosum* L.). It is found throughout the world and this pathogen causes scabby lesions on tubers and galls on roots. Powdery scab is primarily a cosmetic disease. An increasing number of newly-developed potato cultivars are susceptible to the disease (Harrison *et al.* 1997). Powdery scab is a disease that must be economically and effectively managed because of the large number of potatoes being raised for fresh market consumption.

For several years, researchers have tried to utilize chemicals for the management of powdery scab. Few have been shown to reduce severity and increase marketable yield. Cultural management strategies (e.g. irrigation timing, pH manipulation, fertilizer, etc.) have also been evaluated for controlling powdery scab, with little or no success (Harrison *et al.* 1997, Taylor *et al.* 1981, Tuncer 2002). Planting potato cultivars which express resistance to powdery scab symptom development has been shown to be the most economical means of controlling the disease (Harrison *et al.* 1997).

Screening cultivars for resistance to powdery scab has primarily been conducted in fields which are infested with the sporeballs of *S.s.s.* Due to the highly sensitive nature of the *S.s.s.* infection process, symptom expression in field environments can be inconsistent from year to year (Falloon *et al.* 2003). *S.s.s.* prefers low soil temperature and high soil moisture for infection and symptom development (Wale 2001).

In recent years, more emphasis has been given to breeding new cultivars for disease resistance (Douches 2005). Much of this screening does not start until late in the breeding process (8^{th} or 9^{th} year) due to restrictions in labor, amount and durability of seed, and time (Holm 2005). As a result, a new cultivar which may be highly susceptible to powdery scab would not be evaluated for resistance until after much time, money, and labor have been put into that cultivar's

development. An efficient growing environment needs to be established for the timely and consistent screening of cultivars for resistance to powdery scab.

In a potato breeding program, a greenhouse is used during the initial stages of the development of a new cultivar because the environment can be effectively controlled and maintained with minimal effort (Holm 2005). Several studies have utilized a greenhouse to evaluate *S.s.s.* infectivity and symptom expression (de Boer 1991, Jellis *et al.* 1987, Mertz *et al.* 2004). This project is designed to determine the appropriate range of variables necessary for favorable powdery scab development. Variables examined include soil temperature, soil moisture, inoculum levels, and cultivar response. After determination of these variables, the resulting greenhouse model will be validated by comparing several cultivars with varying levels of powdery scab susceptibility against San Luis Valley field trial results.

CHAPTER 2: LITERATURE REVIEW

POTATO HISTORY AND IMPORTANCE

Where Potatoes are Grown:

The potato (*Solanum tuberosum* L.) was discovered by the western world in the mid-1500's and is thought to have been brought to Europe around 1570. Spain was the first European country to receive the potato, which was eventually disseminated throughout Europe. It was not initially recognized as an important part of the diet due to superstitions and the fact that potatoes were thought to be poisonous. Also, due to its similarity to Nightshade, some felt that the potato might be a narcotic (Brown 1993).

Potatoes gradually became more popular in Europe due to their high yields when compared with other crops, such as wheat or barley. Potatoes were a great benefit to the people of Ireland, and between the years 1790 and 1845, the population in Ireland increased from 1.5 million to 9 million. This was due, in part, to increased potato crop production coupled with high yields, which increased production while using the same amount of land. Tenants could use potatoes as a food source for the family since meat and grain were used as rent to pay English landlords. The large consumption of potatoes came to an end in 1845 with the widespread occurrence of the late blight fungus (*Phytophthora infestans* (Mont) de Bary), which resulted in the Irish Potato Famine. This catastrophe resulted in the death of one million people and the emigration of two million people from Ireland, mainly to America (Brown 1993).

As a result of the loss of potatoes due to the late blight epidemic, efforts were made to find or develop potato cultivars with resistance to late blight. Since that time, potato breeders have been looking for resistance to late blight (in addition to other potato pests) using the gene pools of different cultivars. Many of the current marketable cultivars, as well as some native cultivars, have effective disease resistance (Brown 1993).

Potatoes are grown world-wide and in a very diverse range of environments (Burton 1983). China is currently the number one producer of potatoes, followed by Russia, India, The United States, Ukraine, and Germany (National Potato Council 2008). Potatoes rank fourth in the world as a food crop of major nutritional importance, following corn (maize), wheat, and rice, while producing the most food value per hectare of production. In the United States, potatoes are grown in nearly every state. In Colorado, potatoes are mainly grown in the San Luis Valley (85%) located in the south central part of the state, with northern Colorado raising the remaining production as a summer crop (15%) (Rowe 1993). Colorado ranked fourth (2007) in the U.S. for certified seed potato production and fifth (2006) in overall potato production (National Potato Council 2008).

The San Luis Valley is a high elevation (2316 meters above sea level), flat intermountain valley, in which potatoes, alfalfa, barley, native hay, wheat, lettuce, spinach, carrots, and strawberries are grown. Because of the lack of precipitation where average annual rainfall is approximately 180 millimeters, crops are watered using primarily center pivot sprinkler irrigation, with some flood irrigation in areas near the Rio Grande River (CPAC 2007).

Warm days and cool nights during the summer months make the San Luis Valley an ideal area for growing potatoes. Cold winters also help reduce potential pest and disease problems. The potato growing season typically runs from May through August, with harvest beginning in September. The majority of the San Luis Valley potato crop goes to the fresh market or for seed production (CPAC 2007).

Potato Biology and Production:

The potato is a member of the family Solanaceae, which includes pepper, tomato, eggplant, and tobacco. Potatoes can propagate themselves in one of two ways. First, potatoes can reproduce through the production of true seed. Plants produce tomato-like berries, which contain seeds (Rowe 1993). Each seed will produce a progeny potato plant that is genetically

different from the parent plant. Generally, potato plants are only grown from true seed in breeding situations. Conversely, a potato plant can also reproduce vegetatively through its daughter tubers, which are genetically identical to the parent (clones). Each daughter tuber, when replanted, will develop into a plant that is an exact clone of the parent (Burton 1983). Potatoes are typically grown from daughter tubers when produced in commercial agricultural situations (Rowe 1993).

A potato tuber is a portion of an underground stem, called a stolon, which has enlarged. Previously, it had been thought that tuber initiation occurred at the time when flowers were produced by a potato plant. This is not always the case and is greatly dependent on the cultivar. A tuber generally forms at the tip of a stolon, but can also develop along the stolon as well. The eyes (buds) tend to be more concentrated at the apical end of the tuber, but normally occur over the entire tuber. The periderm and the corky epidermis make up the skin of a tuber, which varies in color and texture depending on the cultivar. The "flesh" of a tuber consists of a cortex, vascular ring, and the outer and inner medulla. A tuber grows through cell division, elongation, and storage of food within its cells (Commercial 1993).

A potato plant goes through five different stages during its growth and development. Stage I is referred to as sprout development. During this stage, sprouts start to develop from the eyes of a seed tuber and roots start to develop at the base of the sprouts (Rowe 1993). Stage II is referred to as vegetative growth. Leaves and stems develop above ground while roots and stolons develop below ground. Stage III is the tuber initiation stage. Tubers begin to develop in the soil at the stolon tips or along the stolon. Stage IV is the tuber bulking stage. Tubers begin to expand and become the primary site for nutrient and carbohydrate deposition. Stage V is the maturation stage. The vines turn yellow, photosynthesis slows, the vines eventually die and the skin on the tuber sets (e.g. become thicker, more resistant to skinning at harvest). After skin set, the potatoes or daughter tubers are harvested and are put in storage for use as next year's seed or for consumption (Rowe 1993).

Potato Breeding Program:

The potato is one of the most diverse food crops in the world with hundreds of cultivars available, a direct or indirect result of breeding efforts. By utilizing the genes of closely related *Solanum* species from South America and the cultivars that are currently available, the breeding of new and better cultivars is possible (Douches 2005).

In Colorado, potato breeding is conducted at the San Luis Valley Research Center, which is primarily funded by Colorado State University, support from other grants, and the Colorado potato growers' organizations. Several potato crosses are made every year which produce true potato seed. Of the true seed that is produced, 70,000 to 80,000 seeds are grown in the greenhouse and the tubers are harvested for replant in the field. Of these, only 700 to 800 selections pass the first year of testing and then move onto the next year's trials. In each subsequent year more of the potato crosses are removed from the program as a result of screening for tuber and plant quality. In years 8 through 13, after intense screening in the previous years, selected new cultivars are given to growers and researchers in many potato growing regions, where they are evaluated for growth characteristics, quality and disease susceptibility. Once a new potato cultivar has passed all the testing and evaluations and fits into a useable market niche, it is released as a named cultivar (Holm 2005).

Potato Resistance to Disease:

Since the devastating outbreak of potato late blight in Ireland in the mid-1800s, the search for ways to eliminate and manage diseases has been a major issue in potato production. Breeding potatoes for resistance to late blight began shortly after the Irish potato famine. Since the site of origin is South America, potato clones from that area were used in the development of

late blight resistant cultivars in the mid-late 1800's due to the high levels of genetic variability found in those plants (Brown 1993).

In the late 1800's, it was observed that cultivars which were propagated by tubers over several generations began to decrease in productivity. It was thought that the seed tubers had "run out" after being planted back for many years and could only be restored by planting true seed, which was not genetically identical to the original cultivar. By the early 1900's, the removal of the "run out" seed tubers was found to be a better solution, which led to the formation of certified seed programs throughout the U.S. We now know that the "run out" was a result of virus infection, such as *Potato virus Y* (PVY) (Whitworth *et al.* 2008). Since then, many other potato diseases have been discovered and breeders have bred for cultivars resistant to these diseases.

Diseases and pests have a significant impact on yield and quality. In addition to late blight and PVY, other diseases/pests have been the focus of breeding efforts such as *Potato leaf roll virus*, *Potato virus X*, bacterial ring rot, early blight, pink rot, pythium leak, powdery scab, Colorado potato beetle, various nematodes, and many others (Jansky 2000, Davidson personal communication 2008). Potato breeders currently have a gene pool that is capable of increasing resistance for many diseases, yet can keep the appropriate market characteristics and quality high for food consumption (Douches 2005). The development of resistant cultivars is the most economical, efficient, and environmentally friendly way for potato producers to control disease.

POWDERY SCAB: A SOIL-BORNE POTATO DISEASE

Introduction:

Powdery scab is a potato disease that is the result of tuber infection by the pathogen *Spongospora subterranea* f. sp. *subterranea* (*S.s.s.*). The first report of this disease was in 1842, at a scientific meeting in Germany, where it was found on local potato crops. In 1847, it was

reported in the United Kingdom and was first observed in North America in 1913. In recent years, it has been an increasing problem in most potato production areas around the world. There are some proposed reasons as to why powdery scab has become more prevalent. First, the pathogen has been introduced to previously clean potato growing areas at an alarming rate. Second, new cultivars are often put into production before their level of susceptibility is known (Harrison *et al.* 1997). In the past, experience with the older cultivars indicated which ones were susceptible and allowed growers to make better informed management decisions.

S.s.s. is considered to be a member of the order Plasmodiophorida (Qu *et al.* 2004). It has been classified both as a fungus and as a protist since it possesses properties representative of each group (Harrison *et al.* 1997). Plasmodiophorids are currently classified in the kingdom Protista by most researchers (Down *et al.* 2002).

Powdery scab can produce brown, corky lesions and galls on the roots of the potato plant which range in size from 0.1 to 1.0 cm in diameter (Christ 2001). Powdery scab also creates lesions on the tuber surface that are scab-like and contain a brown to tan colored powdery mass of cystosori (sporeballs), which gives the disease its name. In severe cases, the galls can reduce the health and vigor of the plant. *S.s.s.* also results in the degradation of tuber appearance and increases water loss through the infection sites in storage, which makes the resulting product undesirable to the consumer (Harrison *et al.* 1997). The causal agent is also a vector for *Potato mop top virus*, which results in poor plant growth and can cause spraing (brown or rust colored arcs or flecks which result from necrotic regions in the tuber flesh) to develop (Harrison *et al.* 1997, Johnson 2002).

Significance of the Disease:

The main yield losses caused by powdery scab are a result of rejection of scabby tubers to achieve potato market grade standards (Wale 2000). Powdery scab is primarily a cosmetic

concern and affects the appearance of a susceptible potato. Powdery scab tuber lesions have little effect on potato yield. The disease, if severe enough, can also result in a loss in plant vigor (Harrison *et al.* 1997, Wale 2000). If a crop has a severe outbreak of powdery scab, it may not be financially justifiable to harvest the crop (Harrison *et al.* 1997). This waste could be a result of stringent market standards, quarantines, or both.

Some countries have a zero tolerance for powdery scab, with differing standards for the levels of powdery scab on seed tubers for export (Wale 2000). In the United States, powdery scab lesions covering 5% or more of the surface of a potato, is not acceptable for seed potatoes or for sale as a U.S. No. 1 (United States standards for seed potatoes 1997, United States standards for potatoes 1997).

In addition to infected tubers being able to distribute powdery scab inoculum, spore infested soil can also be an issue in spreading the disease. Powdery scab is a major problem in soils where tuber infection has occurred in the past (Wale 2000). Also, a potato cultivar that is resistant to tuber lesion development may be highly susceptible to root galling, which could drastically increase the amount of inoculum in the soil (Falloon *et al.* 2003). Once in the soil, sporeballs can be carried by the wind during dust storms (Wale 2000), which occur regularly in the San Luis Valley (Davidson, personal communication 2008). This could result in an increase in inoculum level in fields that have not had a previous history of powdery scab, or the movement of spores into sterile greenhouse environments, making this disease difficult to contain in isolated locations.

Recently, *S.s.s.* has taken on new significance as the vector of *Potato mop top virus* (PMTV). PMTV is a member of the genus furovirus and has a single stranded RNA genome (Arif *et al.* 1995). A tuber infected with PMTV may show symptoms of spraing. Total yield may or may not be affected, with the primary loss resulting from rejection due to areas of necrotic tuber tissue which are hard to identify and sort out during routine grading of the crop. Also, an infected potato plant may not contain the virus in all of its stems, which can make it difficult to

rogue (remove from the field) (Johnson 2002). The possibility of *S.s.s.* spores vectoring PMTV significantly increases the number of potential problems due to powdery scab outbreaks in a potato crop.

Life Cycle:

The life cycle of *S.s.s.* is not yet fully known, but the essential features have been identified. The life cycle consists of two phases, a sporogenic or primary phase and a sporangial or secondary phase. The sporogenic phase results in the formation of resting spores and the sporangial phase is the secondary or repeating phase (Braselton 1995).

A single cyst (spore), which is released from a cystosorus (spore ball) that is present in the soil, will germinate and produce a single, primary zoospore. This will only occur if the soil conditions and environment are conducive to germination and liquid water is present (Harrison *et al.* 1997). A primary zoospore can then infect a tuber or root hair (Diriwachter *et al.* 1991). If a root hair is infected, the zoospore develops into a primary plasmodium, which can cleave into several zoosporangia. A primary zoosporangium will undergo mitotic divisions until eight zoospores are produced and released. These are the secondary zoospores, which can re-infect the root hairs and repeat the disease cycle (Harrison *et al.* 1997). Not all of the zoospores produced in the zoosporangium are necessarily released at the same time (Merz 1992).

The secondary zoospores can also infect the tubers, which can result in lesion development on the tubers. Tubers are most susceptible to infection within two to three weeks after tuber initiation has occurred, but can still be infected as the tuber matures. Infection of the root hair or tuber can result in the formation of resting spores, which can survive in the soil or on the tuber until conditions are suitable for germination (Harrison *et al.* 1997).

The infection agents (primary and secondary zoospores) are biflagellate with one short and one long flagellum (Merz 1997). The zoospores move in the direction of the short flagellum, with the long flagellum acting as a rotor (Merz 1992). The morphology and swimming pattern of

the primary and secondary zoospores are identical. The zoospores are thought to be attracted to the root or tuber by the excretion of host exudates and penetrate the host tissue through mechanical means. Zoospores are considered the primary target for powdery scab management due to the resistant nature of resting spores (Merz 1997).

The life cycle of *S.s.s.* has also been documented to occur on plant species other than potato. Tomato bait plants have been used for studying the release of zoospores and details of the infection process (van de Graaf *et al.* 2000, Merz 1989). Falloon showed that planting potatoes in rotation with a wheat or brassica crop resulted in a lower powdery scab incidence than following a fallow year (Falloon *et al.* 1999). Qu and Christ (2006) examined several plant families including many weed species (eastern black nightshade, pigweed, and lambsquarters) common in potato production areas and several crop species (alfalfa, soybean, oat, corn, wheat and tomato). Root infection, root gall formation, and sporeball production were evaluated. Only six of the 26 species were shown to produce root galls (yellow mustard, penlate orchard grass, oat, eastern black nightshade, tomato, and jimsonweed - the last three of which are in the family Solanaceae). This study showed that the life cycle of *S.s.s.* can occur on these six species, in addition to potato (Qu *et al.* 2006).

Powdery Scab Identification Methods:

On potatoes grown in the field, powdery scab can be visually detected on tubers (lesions) and roots (galls and brown, corky lesions). However, spore balls and zoospores can only be seen with the aid of a microscope, thus the presence of *S.s.s.* cannot always be identified visually. Also, an infection site that may produce zoosporangia but does not result in the production of root galls or lesions cannot be seen by the naked eye. Zoosporangia can be stained and then observed under a microscope (Merz 1989). However, staining zoosporangia is not species specific to *S.s.s.* since zoosporangia of several species will also take up stain and can result in a false positive (van de Graaf *et al.* 2000).

In addition, determining the presence of non-lesion and/or non-gall forming infections is difficult since *S.s.s.* is an obligate parasite. It is difficult to isolate the pathogen since it cannot be grown as a pure culture (Harrison *et al.* 1993). To quantify levels of *S.s.s.* in soil and to determine whether infection has occurred without visually identifying lesions or galls, more sensitive methods need to be utilized for identification.

In 1983, a method was developed for quantifying *S.s.s.* in field soil using bait plants. In addition, Harrison developed an antiserum for the identification of *S.s.s.* in the spore ball stage. The ELISA (Enzyme Linked Immuno Sorbent Assay) test he developed could detect spore balls, but could not detect the plasmodial stage. This was because the epitopes, which give rise to the antibodies, were specific to the spore ball walls. This ELISA, however, was more specific to *S.s.s.* and was much more sensitive than other known identification methods (Harrison *et al.* 1993).

In 1998, Bulman and Marshall isolated DNA primers to use conventional PCR (Polymerase Chain Reaction) to detect tuber infections caused by *S.s.s.* The ribosomal internal transcribed spacer (ITS) was examined and primers were designed from these DNA sequences. The primers Spo8 and Spo9 (which amplified a DNA product of 390 base pairs (bp)) were developed during this study (Bulman *et al.* 1998). Since Bulman and Marshall's initial work, there has been much progress in the use of PCR technology for *S.s.s.* identification and the development of several different and more specific primer pairs (Bell *et al.* 1999, Qu *et al.* 2001, Qu *et al.* 2004). In 2004, Qu and Christ discovered two distinct isolates of *S.s.s.* by using PCR techniques (Qu *et al.* 2004).

A real time PCR technique was developed by Ward that has decreased the amount of time necessary to identify *S.s.s.* in infected tubers and has increased sensitivity over ELISA and conventional PCR techniques (Ward *et al.* 2004). In addition, a competitive PCR technique was used to quantify *S.s.s.* levels in the soil. The ratio of the amount of amplified competitive DNA product (541 bp) to the *S.s.s.* DNA product (434 bp) was calculated. A standard curve was then

created with ratios of *S.s.s.* spore ball DNA and several levels of competitive DNA. The number of spore balls per gram of soil was then determined based on the curve (Qu *et al.* 2006). The development of ELISA and PCR techniques in recent years has greatly improved the accuracy and sensitivity of *S.s.s.* identification and quantification.

Conditions Favorable for Infection by *S.s.s.* and for the Disease Cycle:

Introduction:

A wide range of environmental factors can determine the severity of a powdery scab outbreak. Since 1918, it has been consistently demonstrated that powdery scab development requires cool, wet soil conditions (Ramsey 1918, Harrison *et al.* 1997). Potatoes are most susceptible to the disease when soil temperatures are in the 11-18°C range (Wale 2001). In addition to soil moisture and temperature, the inoculum level in the soil and on seed tubers can play an important role in powdery scab development. Research in Scotland indicates that as little as 0.5 spores/gram of soil is considered an infective dose (van de Graaf *et al.* 2000). The condition of the spore balls and the components of the soil can also play a major role in *S.s.s.* infection (Harrison *et al.* 1997).

Soil Moisture/Irrigation:

For the primary and secondary zoospores to travel and infect a host, they must be in close proximity to the infection site and free water must be present. Several studies have shown that infection and lesion development cannot occur without free water present in the soil (Harrison *et al.* 1997). One study on water was conducted by Tuncer who looked at different irrigation levels for the duration of a growing season. Potato plots that received low levels of irrigation water (400 mm) expressed less powdery scab symptoms than plots that received higher levels of irrigation water (650 mm) (Tuncer 2002).

Another study showed that by altering irrigation patterns at various times during plant development, powdery scab severity could be reduced. When irrigation water was withheld

seven days prior to tuber set and until 21 days after tuber set, powdery scab severity was reduced. However, disease incidence was not affected and a reduction in total yield was also a result of withholding irrigation water during tuber set (Taylor *et al.* 1986).

Different soil matrix potentials were also evaluated and it was found that a matrix potential of -0.0001 bars (near saturation) had the highest index for zoosporangia presence in root hairs in the potato cultivar Sebago. As the saturation level was decreased, the disease index for zoosporangia in the roots decreased proportionally. Infection of roots was favored by high soil moisture content (de Boer 2000).

In Scotland, a study was conducted that looked at fluctuating soil moisture levels. Fluctuating soil moisture was less conducive to powdery scab development than soil that remained in constant moisture. However, in a study conducted by Burnett, there was no significant difference in powdery scab severity between fluctuating and constant soil moisture (van de Graaf *et al.* 2005). Also, de Boer did not see a difference in powdery scab severity in poorly verses well drained soil in a greenhouse environment. This was possibly a result of an increased amount of irrigation in the well drained soil (de Boer 1991). Coarse textured soils that have good drainage can also be conducive to powdery scab development in high altitude areas that are dependent on irrigation. In this situation, temperature also plays a major role in disease expression (Zink *et al.* 2004).

Soil Temperature:

Powdery scab development is favored by cool soil temperatures. In addition, zoospores of *S.s.s.* can be released rapidly from the resting spores at temperatures ranging from 12-15°C (Harrison *et al.* 1997).

In Scotland, a study was conducted by van de Graaf to compare three different temperature regimes (9, 12, and 17°C) in a greenhouse environment. Powdery scab levels were high at each temperature regime, but the 12°C regime produced the most severe disease expression (van de Graaf *et al.* 2005). In a study conducted by de Boer, using the potato cultivar

Kennebec, four temperature regimes were examined. The greatest incidence and severity of powdery scab occurred at 12.5°C, intermediate levels of disease occurred at 15°C, low levels occurred at 17.5 and 20.0°C, and no powdery scab was observed at 10°C (de Boer 2000). Soil temperatures in the 12-15°C range appear to be the most conducive to powdery scab development.

In Queensland, Australia, susceptible potato cultivars grown during the winter are much more prone to developing powdery scab than cultivars grown in the summer. This is primarily due to the cooler growing conditions during the winter (average monthly soil temp of 17.2°C and 18.2°C for July and August), which is more favorable to powdery scab development (Hughes 1980).

Some research has been conducted to assess the effectiveness of delaying the potato planting date to reduce the amount of time tubers are exposed to cool, wet soils; powdery scab was reduced by delaying planting in Pennsylvania (Christ 1989). However, Zink *et al.* (2004) observed the opposite effect when planting was delayed in the San Luis Valley, Colorado; the differences in these two studies is likely a result of more favorable soil conditions later in the season in the San Luis Valley, which is a high mountain valley with coarse textured soils. This may suggest that in addition to soil temperature, soil texture could also play a role in powdery scab development.

Inoculum Levels:

The amount of powdery scab inoculum also plays a role in the infection of potatoes by *S.s.s.* and in the expression of powdery scab symptoms. A study conducted in Switzerland used tomatoes as bait plants with varying concentrations of cystosori in a nutrient solution. There was a correlation between the number of root infections observed and the number of cystosori present in the nutrient solution (Merz 1989). Since this study measured only root infection rather than root galling or tuber lesion development, conclusions about powdery scab symptom development verses inoculum level could not be made.

In a study conducted by Christ (1989), dried potato skins with powdery scab lesions were added to field soil prior to planting. Six cultivars were then planted in the inoculated soil and compared with the same six cultivars planted in naturally infested soil. The final results were not conclusive in terms of more disease expression in the potatoes grown in the inoculated soil. This may have been due to high levels of inoculum in the naturally infested soil. By adding more inoculum to the soil, the levels of powdery scab observed did not increase (Christ 1989). It appears that there is an upper limit to the amount of inoculum present in the soil which will be expressed as an increase in powdery scab symptoms.

In a study conducted by van de Graff *et al.* (2005), three different field soils (sand, loam, and clay) were evaluated for powdery scab development in a greenhouse setting. To each soil type, four inoculum amounts were added (0, 5, 15, and 50 cystosori/gram of soil). The control treatments (0 cystosori/gram of soil) resulted in significantly less powdery scab than in the inoculated treatments across the three soil types. However, the 5, 15, and 50 cystosori/gram of soil treatments showed no significant difference in the amount of disease development. This study showed that under favorable conditions, a small amount of inoculum (5 cystosori/gram) can cause as much damage to a susceptible potato crop as higher levels.

The initial amount of inoculum in the soil may play a less important role in overall disease development than previously thought (Harrison *et al.* 1997). Inoculum levels as low as 3 cystosori/gram of soil were as capable of causing infection as higher inoculum levels. High infection rates from low inoculum levels can be explained since each cystosorus contains hundreds of resting spores and once an infection occurs, secondary infections can rapidly propagate *S.s.s.* (van de Graaf *et al.* 2005). Powdery scab development in roots and on tubers may be affected more by other factors (Harrison *et al.* 1997). However, based on the findings of Merz (1989), root infection rates correlate to the levels of inoculum present. The relationship between root infection, symptom development, and inoculum level dynamics in the soil need to be better understood for use in developing an appropriate powdery scab management plan.

Soil Texture and pH and their Effects on Powdery Scab Severity:

According to Wale (2001), powdery scab tends to be high in soils that have a high water holding capacity (e.g. clay soils) and where drainage is poor (due to soil texture or compaction). Powdery scab can also be severe in soils with a low water holding capacity (e.g. sandy soils) if alternating wet and dry cycles occur (Wale 2001). However, according to van de Graaf *et al.* (2005), lighter soils (sandy and loamy) are more conducive to disease development than heavier clay soils. It has also been suggested that sandier soils are more favorable to disease development due to the increased amount of irrigation required to grow potatoes (de Boer 2000). Powdery scab severity may have more to do with soil moisture within a given soil texture, rather than soil texture alone.

Soil pH can influence the viability of *S.s.s.* Alkaline soils, such as those found in the San Luis Valley, CO, tend to be more conducive to powdery scab development than acidic soils. Hughes (1980) showed that a reduction in soil pH can reduce the incidence of powdery scab. A soil pH of less than 5.5 resulted in significantly less scab than in higher pH soils. Merz (1989) looked at the effect of a range of pH (5.0-8.0) on root infection by *S.s.s.* He found no differences between the different pH levels. Diriwachter (1981) found that different pH levels (5.0-8.0) had no effect on resting spore germination. The relationship between powdery scab and pH is not fully understood. However, based on his observations, Wale (2001) suggests that lower levels of powdery scab tend to be present in more acidic soils.

Viability of Cystosori (resting spores) and Zoospores:

It is known that several factors are involved in the infection of a potato plant by *S.s.s.* The condition of the cystosori and the relative susceptibility to infection of the host, however, are two key factors which must be considered (Diriwachter *et al.* 1991, Harrison *et al.* 1997, Merz 1989).

Cystosori produced by *S.s.s.* have been documented to remain viable in soil for up to six years (Harrison *et al.* 1997, Christ 2001), and it has been suggested that the cystosori can cause disease for 20 years or longer (Miller 2001). Resting spores can also remain viable after passing through the digestive tract of farm animals (Harrison *et al.* 1997).

Research suggests that if the resting spores are kept in moist soil for an extended period of time, the spores lose the ability to germinate. If the soil then becomes dry, there is evidence that the resting spores can regain the ability to germinate when favorable conditions exist (Merz 1989). In addition to the condition of the cystosori, the susceptibility of the host plant affects infectivity and symptom expression.

Taylor showed that powdery scab was more severe in a greenhouse when inoculum was added to the soil at planting or at emergence, rather than later in the season (Taylor *et al.* 1986). Diriwachter *et al.* (1991) conducted an experiment which looked at the timing of infection in the potato cultivar Kennebec. When inoculum was added to the soil at tuber initiation, infection occurred and when inoculum was added 20 to 25 days after tuber initiation, no infections occurred. The susceptible cultivar did not develop powdery scab symptoms when inoculum was withheld during the two to three week period that tuber initiation occurs (Diriwachter *et al.* 1991).

Certain areas of a growing potato are more susceptible to infection than others. The rose or bud end of a developing tuber is more susceptible to infection than the stem or heel end (Wale 2001). This may be a result of exudates being released into the soil from the dividing, filling cells. As a tuber grows, the filling cells divide in the meristematic region of the potato, which is typically at the rose end. A larger number of lesions may develop on the rose end as a result of zoospores being attracted to these filling cell exudates (Diriwachter *et al.* 1991).

Management of Powdery Scab:

Introduction:

The limited ability to manage powdery scab has been an ongoing problem for potato producers and researchers since its discovery over 150 years ago. Chemicals have been evaluated with only a few showing any effectiveness at reducing the disease (Harrison *et al.* 1997). The chemicals that reduce symptoms can also be cost prohibitive. Different cultural management practices have been evaluated with some success. These include irrigation, soil pH, soil fertility, inoculum load in the soil and on seed, crop rotation and the use of trap crops (Harrison *et al.* 1997, Falloon *et al.* 1999, Qu *et al.* 2006). Effective cultural management practices typically only reduce disease severity rather than eliminate it. Cultivars which are resistant to powdery scab symptom development are also used. This is the best option at limiting powdery scab (Harrison *et al.* 1997). However, potato cultivars which express resistance do not necessarily have the characteristics (e.g. skin type, shape, color, and flavor) that the consumer demands.

Control using Fungicides:

Potatoes have been treated with many different chemicals since the early part of the twentieth century for control of powdery scab. Formalin, copper sulfate, and sulfur were some of the first chemicals used that showed some measure of control. Research has shown that mercuric chloride showed some control and zinc sulfide has worked well when disease pressure was low (Harrison *et al.* 1997). Braithwaite *et al.* (1994) found that several chemicals (including fluazinam) applied to seed tubers worked well in suppressing the disease. Several trials in the early 1990's showed that applications of up to 3kg/ha of fluazinam reduced disease incidence and severity (Mertz 2000). Research conducted in Pakistan showed that the use of Boric acid or elemental sulfur as a pre-plant seed treatment reduced disease levels, but it resulted in a decrease in soil pH that may be too low for successful potato growth (Jan *et al.* 2002). In the San Luis Valley (Colorado), it has been shown that the chemical Omega (a.i.-fluazinam) has had some success in controlling powdery scab (Zink *et al.* 2004). Also, treatment of whole seed with a 2%

solution of Formaldehyde prior to cutting has been used to reduce tuber-borne powdery scab spore viability in California (Davidson personal communication 2008).

There have been varying levels of success with different chemicals in different areas of the world during the last 30 to 40 years. However, most chemicals work best when disease pressure is low (Harrison *et al.* 1997). A good chemical control has not yet been developed that can deal with high levels of powdery scab. A more effective method for managing powdery scab needs to be developed.

Control using Cultural Management Strategies:

There have been several studies looking at different environmental factors that could have an effect on controlling powdery scab. One important management strategy that could be utilized to control powdery scab is irrigation timing in regions dependent on irrigation. Since soil moisture plays a major role in powdery scab development, irrigation management could be a useful tool. In a study conducted by Taylor *et al.* (1981), three irrigation regimes were looked at in both greenhouse and field settings. The potato cultivar Kennebec was used for this study. One regime was standard irrigation, the second regime was withholding irrigation until tuber set (TS), and the third regime was withholding irrigation until three weeks after TS. In both the greenhouse and field, powdery scab incidence was reduced when irrigation water was withheld until TS and three weeks after TS. In another study conducted by Taylor *et al.* (1986), several irrigation regimes were examined which involved withholding irrigation before, during, and after TS. Withholding irrigation during TS consistently reduced powdery scab severity, but had little effect on incidence. Tuncer (2002) showed that lower total irrigation applied during a growing season (400-650 mm) resulted in lower powdery scab incidence. Also, van de Graaf *et al.* (2002, 2005) showed that powdery scab is more severe in soils that are kept in constant moisture than in soil where the moisture fluctuates. These studies show that irrigation management is a tool that can be used to decrease powdery scab severity where irrigation is necessary to grow a potato crop.

In fields where powdery scab inoculum levels are low or nonexistent, planting seed that is scab free is a good option for managing the disease. Without planting, harvesting, and evaluating a susceptible potato cultivar in a particular field, determining the inoculum level in the soil has been virtually impossible until recently. A PCR technique was developed by Qu *et al.* (2006) for quantifying the level of inoculum in naturally infested soils and correlating that to powdery scab severity. Soil was collected and analyzed from across the U.S. and Europe, and a competitive PCR assay was used for quantifying inoculum levels. The results were compared with powdery scab incidence on tubers grown in the fields from which the analyzed soil was collected. Field soils with high, medium, low, or zero inoculum level resulted in tubers with the corresponding disease incidence from the San Luis Valley. For example, a low inoculum level resulted in low tuber disease incidence (Qu *et al.* 2006 #2). By using a competitive PCR assay, field inoculum levels can be estimated without planting susceptible cultivars, which would increase soil inoculum levels if inoculum was present.

The manipulation of soil pH has also been evaluated in managing powdery scab (Hughes 1980). Sulfur and lime treatments were used to create a range of soil pHs (from 3.7 to 7.1). The disease levels increased as lime content in the soil increased. As sulfur levels increased, disease levels decreased. However, in the first experiment, the treatment with the lowest level of sulfur increased the disease severity more than any other treatment. This might be a result of a plant response to the sulfur. Powdery scab expression may be greater in soils with a higher pH.

Fertilizer amounts may also have an effect on powdery scab severity. In a study conducted by Tuncer (2002), nitrogen (N) levels were increased from 150 to 600 kg/ha using the potato cultivar Marfona (which is highly susceptible to powdery scab) in the country of Turkey. Generally, powdery scab severity was greater at the higher N levels (Tuncer 2002). More research needs to be conducted with lower levels of N (less than 150 kg/ha) to determine whether decreasing N levels can reduce disease levels without sacrificing economical yield potential.

In addition to pH, crop rotation has been shown to help reduce powdery scab severity in the field. In a study conducted by Falloon *et al.* (1999), a crop of potatoes was rotated with fallow ground and several different crops (wheat, brassica, and ryegrass/clover). Potatoes planted in the wheat and brassica rotations had a lower percentage of powdery scab incidence than the fallow and ryegrass/clover rotations. It was also reported that a three year rotation with ryegrass/clover did not reduce incidence or severity of powdery scab (de Boer 2000). Some cover crops may help reduce powdery scab when used as a trap crop in the field (Falloon *et al.* 1999, Qu *et al.* 2006).

There are several plant species that *S.s.s.* has been known to infect. Some of the represented genera include: *Lycopersicon, Trifoliium, Triticum, Hordeum, Avena, Lolium, Linum, Medicao*, and *Brassica*. However, de Boer (2000) found that only the zoosporangial stage was produced in these species and it is unclear whether or not infection would take place in the field. A study was also conducted by Qu *et al.* (2006) which evaluated a wide range of plant species as possible hosts for *S.s.s.* in the United States. Twenty six species, including ten plant families, were screened in the lab for possible infection. Of the 26 species, 16 developed zoosporangia in the roots (nine families), six developed root galls (three families), and three developed spore balls (three families). The families that produced root galls and spore balls were Brassicaceae, Poaceae, and Solanaceae, and the families that only produced zoosporangia were Amaranthaceae, Asteraceae, Chenopodiaceae, Cyperaceae, Fabaceae, Malvaceae, and Polygonaceae. Species that only develop zoosporangia may have an impact in reducing soil inoculum since resting spore balls, which propagate the organism, are not produced. However, field tests need to be conducted before a conclusion can be made (Qu *et al.* 2006).

Control using Resistant Cultivars:

All potato cultivars are thought to be susceptible to powdery scab to some degree (Harrison *et al.* 1997). The current market reliance on cultivars that are highly susceptible to powdery scab is a major factor contributing to the increased difficulties in controlling powdery scab (Christ 2005). Some cultivars are more susceptible to lesion development than to root gall development and vice-versa. It is unclear what regulates the susceptibility in root hairs or on tubers. A cultivar may be very resistant to powdery scab lesion development but be very susceptible to root galling. The tubers can appear asymptomatic, while root galls develop on the roots, which results in an increase in the amount of powdery scab inoculum in the soil (Harrison *et al.* 1997).

In a ten year study conducted by Falloon *et al.* (2003), 99 potato cultivars were evaluated for resistance. It was found that nearly all of the cultivars that were "very resistant" to lesion development in the field were also resistant to root infection. However, Falloon also thought that this may not be true in every situation. According to Zink *et al.* (2004), tuber resistance is not correlated to root resistance. Also, potato cultivars with smooth skin have generally been more susceptible to powdery scab lesion development than cultivars with a russeted skin (Miller 2001). The mechanism for resistance in certain cultivars has not been determined (Harrison *et al.* 1997); there is little known about what causes powdery scab development in different cultivars and what the connection is between lesion and root gall development.

Evaluation of Powdery Scab Using a Greenhouse

Research has been conducted on powdery scab development in field and greenhouse environments. Field studies are important, but due to the dynamic environment, field trial results can result in inconsistent conditions and data. Research conducted in the San Luis Valley has shown that powdery scab incidence and severity can vary from year to year for susceptible

cultivars. For example, in 2006, the percent incidence of powdery scab was 70.7% in control plots for the susceptible potato clone DT6063-1R (Cherry Red). In 2007, percent incidence was only 44.4% for the same cultivar in grower cooperator fields in the San Luis Valley (Davidson 2008). This is due, in part, to the availability of fields infested with *S.s.s.* for research, different irrigation practices among grower cooperators, and seasonal temperature and precipitation variability from year to year.

To consistently evaluate cultivars for susceptibility to powdery scab, a consistent location and environment needs to be developed. In a greenhouse, the environmental conditions can be controlled and soil can be mixed and inoculated at a level which is conducive to reproducible powdery scab symptom development. The dependence on grower cooperators and the weather for screening potato germplasm for powdery scab could be eliminated. Also, the evaluation of cultivars for powdery scab susceptibility would not be dependent on outside environmental conditions. In temperate regions, a greenhouse would allow for cultivar evaluations to be conducted during all seasons and not just during the summer or spring.

In addition, the initial stages of a potato breeding program usually occur in a greenhouse environment (Holm 2005). When evaluating new potato germplasm, as well as previously recognized potato cultivars, for powdery scab susceptibility, a greenhouse would be an ideal choice. Favorable environmental parameters within a greenhouse setting need to be determined for powdery scab development before reliable evaluation of potato germplasm can be accomplished.

Most powdery scab research has been conducted in the field, although several studies have occurred in a greenhouse. Anderson *et al.* (2002) conducted a study in a greenhouse environment that looked at the relationship between PMTV and *S.s.s.* in several plant families, and Falloon *et al.* (2003) and de Boer (1991) have looked at cultivar susceptibility in the greenhouse (Falloon *et al.* 2003; de Boer 1991). Diriwachter studied the infection biology of *S.s.s.* in the greenhouse. Taylor *et al.* (1986) and van de Graff *et al.* (2002) looked at irrigation

regimes and their impact on *S.s.s.* infection in the greenhouse. These studies show that a greenhouse can be used for detecting and evaluating powdery scab on potato.

PURPOSE AND SCOPE OF PROJECT

This project was designed to develop a greenhouse assay (e.g. evaluate proper soil temperature, inoculum level, soil moisture) that will consistently result in powdery scab development in susceptible potato cultivars. Additionally, greenhouse results should correlate with disease expression that is seen in the field. A cultivar found susceptible or resistant in field trials should show a similar disease expression in the greenhouse. Previous powdery scab research conducted in greenhouse settings was used to develop a suitable greenhouse assay.

The focus and primary objective of this project was to determine the environmental greenhouse settings which produce comparable results found in field trials conducted in the San Luis Valley. A greenhouse assay was developed in which this comparison can be made and it could be used on a much larger scale. A universal standard could foreseeably be developed for powdery scab that is not dependent on disease readings which vary in different potato production areas. Also, a potato breeding program could utilize this greenhouse assay to screen new cultivars for powdery scab susceptibility and resistance much earlier in the breeding process. A greenhouse environment was chosen for this project to ensure more consistent powdery scab development in susceptible and moderately susceptible potato cultivars and to assist in clearly identifying resistant cultivars.

CHAPTER 3: MATERIALS AND METHODS

Phase 1: Trials – Inoculum Level x Soil Type x Cultivar, Inoculum Source x Soil Type, Soil Moisture x Inoculum Level x Cultivar

Acquiring Inoculum:

Inoculum was obtained from powdery scab lesions on Rio Colorado (NDC5281-2R) tubers grown in field plots from the 2005 growing season. Lesions containing sporeballs (sb) were scraped off infected tubers onto a sterile surface using a knife. A mortar and pestle was then used to thoroughly break up the lesions. The concentration of spore balls present in the ground material was determined using an enclosed digital scale. Ground material equaling 0.0007 g and 0.0015 g was measured and five ml of distilled water was added to each measured amount. Fifty microliters of this suspension, after mixing, was placed on a microscope slide with cover slip. The number of sporeballs was determined under 100x magnification. The total number of spore balls per gram of inoculum was then calculated using an average of three replicated counts. On average, 0.001 g of ground material contained 2,725 sporeballs. A similar method for obtaining inoculum was used by van de Graaf (2005).

Soil preparation and inoculum addition:

Two soil mixes were used in phase #1. Soil type #1 (S1) was the standard soil used in the potato breeding program at the San Luis Valley Research Center (S1 = 1:1:1 - vermiculite: peat moss: sand). Soil type #2 (S2) was a soil with 50% sand (S2 = 1:1:2 - vermiculite: peat moss: sand). Each soil type was mixed using an electric cement mixer. Different amounts of inoculum were then added to and mixed in the soil.

The amount of inoculum that was added to the soil was based on the weight of soil in each batch. S1 soil was lighter than S2 soil because it had a lower sand content. In order to have uniform inoculum addition between the different soil types, an average dry weight (average of S1 and S2 equaling approximately 1700 g of soil/pot, pot size = 15x15x15 cm) was used to determine the amount of inoculum to add to each soil batch, depending on the projected inoculum

level (1, 5, or 10 sb/g of soil). Two hundred ml of distilled water was put into the spray bottle with the appropriate amount of inoculum and sprayed on the soil as it was being mixed. Once no more solution could be sprayed, the remaining liquid in the bottle was swirled and poured into the mixing soil. Another 200 ml of water was added to the bottle and the rinsate was also added to the mixing soil.

In trial 1, four inoculum levels were evaluated: UC = Untreated Control, I1 = 1, I2 = 5 and I3 = 10 sb/g of soil. A total of eight soil mixtures were evaluated (2 soil types x 4 inoculum levels). The batches of soil were mixed in the following order:

1. UC,S1; 2. UC,S2; 3. I1,S1; 4. I1,S2; 5. I2,S1; 6. I2,S2; 7. I3,S1; 8. I3,S2

The soil was mixed in this order to reduce any sporeball contamination from soil with a higher inoculum level into a soil with a lower inoculum level or untreated control. Also, between mixing each batch of soil, the cement mixer and wheelbarrow were rinsed with water to remove any excess soil and reduce sporeball contamination.

For the inoculum source trial (trial 2), Rio Colorado seed with high levels of powdery scab lesions were obtained. The S1 and S2 soil types were used with three inoculum source treatments (untreated control, soil-borne inoculum, and seedpiece inoculum). The average number of lesions per infected tuber eyeball (eye region with one sprout) was between 10 and 15. Each lesion was estimated to have between 400 and 1000 sporeballs (Davidson personal communication) resulting in a total inoculum load of approximately 7500 sb/eyeball. When comparing seedpiece inoculum load to the soil-borne inoculum, it was decided to approximate the levels found on the seedpiece. Thus, 5 sb/g of soil was mixed into the soil to equal approximately 7500 sb/pot.

For the soil moisture trial (trial 3), the I1S2 soil was used with two irrigation treatments (3.8 liters per hour (lph) throughout the plant growth, and 3.8 lph to 1.9 lph prior to tuber initiation and during early bulking, and then back to 3.8 lph for later bulking).

Preparing Seed:

Certified seed potatoes of the cultivars Rio Grande Russet, Russet Burbank, DT6063-1R (Cherry Red), Rio Colorado (NDC5281-2R), and non-certified, scabby Rio Colorado seed were held at room temperature (20°C) for 21 days to break dormancy and initiate sprouting. A melon scoop was then used to remove individual potato eye regions from the seed tubers. Each potato eyeball contained one sprout (eye). Due to differences between cultivars, sprout length varied at the time of melon balling. Depending on the size of the seed tubers, one or more eyeballs could be taken from each tuber.

Between tubers, the melon scoop was dipped in a dilute sodium hypochlorite solution (10 ml chlorox + 90 ml sterilized/double distilled water) to reduce disease contamination between sampling. The eyeballs were placed in black flats and placed between paper towels moistened with sterilized/double distilled water to maintain appropriate humidity for suberization and healing of the cut seed. Between sampling each potato cultivar, a fresh batch of the sodium hypochlorite solution was used to disinfect the melon scoop to reduce any disease transfer between cultivars. The potato eyeballs were allowed five to seven days to suberize prior to planting to further reduce the risk of disease development (e.g. seed decay).

Planting Seed:

Only one batch of soil was used for planting at a time. Each pot was filled with soil to about 2.5 cm from the top of the pot, to allow a drip tube to fit near the top of each pot. The pot size was 15 x 15 cm; soil was damp at the time of planting. A hole 6.4 cm deep and 2.5 cm in diameter was created in the center of each soil filled pot in order to create a site that was uniform for the placement of each eyeball. Each eyeball was then placed (eye facing up) in a pot. The eyeballs were arranged in such a way that the eyeballs planted for replication I had the largest sprouts, while later replications had shorter sprouts to decrease the variability within replications. Once an eyeball was planted, the pot was labeled appropriately and additional soil from the same batch of soil was added to cover the eyeball. Immediately after planting, each pot was watered to

saturation using approximately 500 ml of water. One extra pot per bench was planted, and a soil moisture and soil temperature sensor was placed in that pot.

Irrigation:

Soil moisture was monitored carefully to make conditions more favorable for powdery scab development. For the inoculum level x soil type x cultivar trial, the pots were irrigated from the planting date until 35 days after planting (DAP) using a garden hose with a soaker nozzle attached. The pots were watered a total of five times using a total of 2,150 ml of water during this time period. A drip system was installed at 36 DAP, just prior to tuber initiation, to precisely measure the amount of water applied to each pot. On average, the drip system applied water every 1-2 days at approximately 160 ml/pot. After the drip system installation, soil moisture was kept within the 5 to 15 cbar range for the duration of plant growth.

For the inoculum source trial, pots were watered from the planting date until 35 DAP (crop #1) and 52 DAP (crop #2) with a garden hose a total of 5 times for a total of 2,150 ml of water (crop #1) and 16 times applying 4,400 ml of water (crop #2), respectively. At 36 DAP, the drip system was installed for crop #1. Crop #2 had the drip system installed at 53 DAP. Crop #1 averaged a water event through the drip system every 1½ days, irrigating an average of 160 ml/pot. Crop #2 was irrigated every 3 days on average, delivering approximately 300 ml/pot. After the drip system installation, soil moisture was kept within the 5 to 15 cbar range.

The soil moisture trial had treatments requiring an alternate irrigation using either 3.8 lph emitters or 1.9 lph emitters. Pots were watered with a garden hose a total of six times during the early growth of each crop applying a total of 1,820 ml of water (crop #1) and 1,690 ml of water (crop #2). At 28 DAP, a drip system was installed for both crops. For crop #1, in the half irrigation regime, the 3.8 lph emitters were exchanged with the 1.9 lph emitters from 46 DAP until 81 DAP when the 3.8 lph emitters were reinstalled. For crop #2, this exchange took place from 49 DAP until 81 DAP. During the growth of both crops, irrigation through the drip system took place every 2 days and the system delivered approximately 260 ml/pot for the 3.8 lph emitter

and 100 - 110 ml/pot for the 1.9 lph emitter. Soil moisture during the drip phase was kept between 5 and 15 cbars for the 3.8 lph pots and between 10 and 25 cbars during the 1.9 lph emitter time frame. The emitter exchange for the half irrigation regime corresponded with the time frame just prior to tuber initiation and through early tuber bulking.

The drip system consisted of a 1.9 cm diameter tube, with 0.64 cm rubber tubes running off of the main line which each had an emitter attached to the end of the tube. Drip T's, each with four branches, were evenly set up along the entire length of the 1.9 cm main line. The 0.64 cm tubes were fed through a 0.64 cm hole that was drilled in the top portion of each of the pots. A 3.8 lph emitter was then connected to the 0.64 cm tube and placed inside each pot. Each drip T irrigated four pots. There were a total of up to 16 T's per bench which could water a total of 64 pots. One extra T was also added to the mainline to measure the amount of water that was being applied to each pot when all 16 T's were being utilized. A 0.64 cm tube with a 3.8 lph emitter was set up on the extra T. Thus, the amount of water was able to be precisely measured for each irrigation event. Irrigation was controlled by a battery operated timer connected to the water source. This approach allowed for control of both the timing and duration of each irrigation event.

Fertilizing:

No starter fertilizer was used in any trial; however, each pot was fertilized individually with the same amount of fertilizer. A fertilizer device (Hozon Brass Siphon Mixer with backflow preventer, Earth City, MO) was connected at the water source. A rubber hose pulled the fertilizer solution through the Hozon device. A garden hose was then attached to the Hozon device and as water was pushed though the hose, fertilizer solution was pumped through the hose with the water. The fertilizer used was the water soluble "Peter's Blossom Booster" (Earth City, MO) with an analysis of 10:30:20. The solution was pumped through the garden hose at a ratio of 1:16 (1 part fertilizer solution: 16 parts water). A soaker nozzle was attached to the end of the garden hose and each pot was watered with the diluted fertilizer solution for approximately 2 to 3

seconds resulting in an application average of 300 to 350 ml of fertilizer solution per pot or between 200-300 ppm nitrogen per pot for each application. For all of the trials, the fertility program was started at around 30 DAP and was continued until 95 DAP. Fertilizer was applied to each pot on a 7 to 10 day schedule during this time frame.

Managing Greenhouse Temperature:

Environmental conditions were controlled in the greenhouse by using an environmental sensor located in the center of each bay which monitored the air temperature. This sensor took readings and communicated with the Wadsworth Envirostep Control Panel (Arvada, CO). The computer then made adjustments (e.g. to turn on the exhaust fan, open the pad vent, activate the heater, start the pump for the swamp cooler) to maintain the appropriate temperature. The temperature in the bay was initially set to 18-24°C with a 1.5° differential at 1 DAP for each of the trials. The temperature range was lowered to 11.5-15°C with a 1.5° differential at 36 DAP for the duration of the plant growth for the inoculum x soil type x cultivar trial and the inoculum source x soil type trial (both crops). The temperature was lowered at 46 DAP for the soil moisture x inoculum level x cultivar trial (both crops). This regime moved soil temperatures into a range more favorable for powdery scab development on the roots and on tubers just prior to tuber initiation.

Four Watchdog weather stations (Plainfield, IL) monitored the soil temperatures and soil moisture for the inoculum level x soil type x cultivar trial. Two sensors were set up in pots with S1 soil type and two in S2 soil type representing each bench in the greenhouse. Each of the cultivars being evaluated was also represented. For the soil moisture x inoculum level x cultivar trial, two weather stations were set up for each crop, one monitored soil conditions for the 3.8 lph emitter irrigation setup and the other one for the 3.8 lph + 1.9 lph emitter setup. Only one weather station was used for the inoculum source x soil type trial for each crop.

To help control insect damage on the plants, insecticides were applied at timely intervals. A Fulex Nicotine Fumigator (Earth City, MO) was set off at 81 DAP and Attain (Earth City, MO)

was used at 89 DAP for the inoculum level x soil type x cultivar trial. For the other two trials, a nicotine bomb was set off at 26 DAP and 41 DAP.

Harvesting and disease readings (roots and tubers):

At approximately 70 DAP, the roots and plant structure from one pot of a susceptible clone/cultivar during each of the trials was examined. The presence of root galls was confirmed at this time. The tubers present on the plant were the size of marbles or smaller. At approximately 115-120 DAP, all pots were harvested. For each pot, the remaining stems were removed with scissors and the roots and tubers were removed from the soil and evaluated for powdery scab severity. Tubers were evaluated at harvest for symptoms and roots were evaluated at harvest and up to one week after harvest for galls.

Tubers were scored for powdery scab severity based on the percent coverage of the tuber by powdery scab lesions. A rating of one to five was given to every tuber that expressed symptoms (Rating: 1 = one lesion up to two percent coverage, 2 = 2.1 to 5.0 percent, 3 = 5.1 to 10.0 percent, 4 = 10.1 to 25 percent, 5 = greater than 25 percent). A standard powdery scab scoring table was used to help determine the rating for each tuber (Fig. 1). The weight of each tuber was determined to arrive at the total tuber weight for the scabby and healthy tubers for each plant. Each symptomatic tuber was rated for severity of the powdery scab symptoms. Then, the total percent of infected tubers per plant was multiplied by the average tuber severity rating per plant to obtain the severity index. The treatment severity index was then calculated by averaging all of the per plant severity indices within a given treatment. This closely approximated the methods used during the SLV field trials to calculate severity indices.

Each set of roots was washed with water to remove any excess soil and placed in a zipper seal bag. The roots were then either examined for the presence of galls or placed in a cooler until symptom evaluations could be made. The roots were weighed (fresh weight) and examined using a dissecting microscope. Roots were given a symptom rating of 0 - 4 based on root gall presence

and severity (Rating: 0 = no root galls observed, 1 = 1 to 3 small or 1 large gall, 2 = 4 to 10 small or 2 to 3 large galls, 3 = 11 to 20 small or 4 to 8 large galls, 4 = > 30 root galls observed).

San Luis Valley Field trials:

Field trials took place over several years in locations which contained naturally infested soil. Sporeball counts were not taken during the field trials, but powdery scab symptoms were prolific in all years on untreated control tubers of susceptible clones/cultivars. The greenhouse trials were structured in such a manner as to mimic ideal field conditions for powdery scab symptom development. All field trials were conducted in cooperating potato growers' fields and production practices (e.g. fertility, planting date, cultivation, harvest) were within normal parameters for potato production in the San Luis Valley.

Statistical Analysis and Experimental Design:

A randomized complete block design was used for each greenhouse trial with six replications per treatment. The first trial included 32 total treatments evaluating the variables of cultivar (4), soil type (2), and inoculum level (4). The second trial included 6 total treatments evaluating inoculum source (3) and soil type (2), while the third trial included 16 treatments examining inoculum level (2), soil moisture (2) and cultivar (4). A least significant difference (LSD) analysis at a 95% confidence interval was utilized to separate treatments for each trial. Agricultural Research Management (ARM) revision 6.1.13 fourth edition (Brookings, SD) was the statistical program used for the analysis. The treatment means for trial 1 were also compared to three years of field data from the growing seasons 2002 - 2006, depending on which years the given clones/cultivars were screened in the field.

Phase 2: Greenhouse Model Validation

Phase 2 used the same inoculum source as in Phase 1 at a rate of 1 sb/g of soil. The soil types used were UCS2 and I1S2. All of the protocols for soil mixes and inoculum placement were the same as described previously. Seed potatoes of fourteen clones/cultivars with varying reactions to powdery scab obtained from the SLV Research Center and the 2006 SLV powdery scab field clonal evaluations were utilized (Tables 1 & 2). Two separate crops with six replications per crop were planted. Planting, irrigation, fertilization, managing the greenhouse environment, and harvest were the same as described earlier for the trial on inoculum level x soil moisture level. Fertilizer was applied at 22 DAP every 10 to 12 days until 96 DAP. The temperature in each bay was initially set to 18-24°C with a 1.5° differential at planting. At 43 DAP, the temperature range was lowered to 11.5-15°C with a 1.5° differential. The early irrigation and drip system installation/watering using only the 3.8 lph emitters was the same as described for the soil moisture trial. Analysis was the same as described for Phase 1 with each crop in Phase 2 having 28 total treatments with the variables of cultivar (14) and inoculum level (2).

CHAPTER 4 – PHASE 1 (RESULTS AND DISCUSSION)

Preface:

The primary objective of Phase 1 of this study was to evaluate the key factors necessary for the greenhouse expression of powdery scab symptoms on potato roots and tubers. The findings from Phase 1 were then used to develop a greenhouse assay for screening potato germplasm for resistance to powdery scab to replace the use of variable field trials. The factors of soil inoculum level, soil type, soil moisture, and inoculum source were evaluated against four potato cultivars: DT6063-1R, Rio Colorado, Rio Grande Russet, and Russet Burbank. These cultivars differ in root and tuber susceptibility to powdery scab.

Data was collected and analyzed from three trials to evaluate these key factors and cultivars. Trial 1 data was compared to three years of San Luis Valley (SLV) field data for each cultivar to determine which inoculum levels and soil types gave results which most closely matched results from the field. In trial 2, inoculum source and soil type were evaluated to determine which source of inoculum produced the most consistent and accurate powdery scab levels. In trial 3, two irrigation regimes were evaluated (high soil moisture level vs. half the soil moisture during the tuber set stage of the potato plant) to determine which regime resulted in the highest and most consistent disease expression. The results from these trials indicated that a soil type with 50% sand, an inoculum level of 1 sporeball (sb)/g of soil (mixed in soil), and a high soil moisture (averaging between 5 to 10 cbars) during and after tuber set produced the most consistent greenhouse results compared to three years of field data.

Trial 1: Inoculum Level x Soil Type x Cultivar

Root Galling:

In trial 1, the variables of inoculum level, soil type, and potato cultivar were evaluated. Each potato cultivar differed in susceptibility to powdery scab root gall and tuber lesion development.

For root galling, soil type 2 (S2 = 50% sand) gave more consistent disease levels across all three inoculum levels when evaluating the four potato cultivars (Figs. 2.1 – 3.2) than soil type 1 (S1 = 33.3% sand). For the cultivar DT6063-1R planted in S2, when evaluating the three inoculum levels, there were no significant differences when the inoculum levels were compared with the three years of field data. When evaluating S1 and the cultivar DT6063-1R, certain root gall severity readings were significantly different when compared with the three years of field data for the I1 and I3 inoculum levels.

For soil type 1, all four cultivars were not significantly different within each cultivar for the three inoculum levels. For cultivars DT6063-1R, Rio Colorado, and Russet Burbank the different inoculum levels also produced similar results within each cultivar for S2. However, in the cultivar Rio Grande Russet, 1 sb/g was the only inoculum level that was not significantly different from the three years of field data for both soil types (Fig. 3.1).

Based on results taken from the four potato cultivars, a soil type with 50% sand and an inoculum level of 1 sb/g is the most ideal combination for obtaining accurate powdery scab root gall data when compared with SLV field results. In general, higher inoculum levels did not increase root gall severity for soil type 1 when comparing against the three inoculum levels. Overall, however, S1 soils resulted in lower root gall severity when compared with the S2 soils. High variability existed between different years of field results in the cultivars Rio Colorado and Russet Burbank. This variability in field results due to environment illustrates the benefits of using a greenhouse or more controllable situation for accurate screening of potato cultivars when evaluating root gall severity.

% Tuber Incidence:

For the cultivar DT6063-1R, there was no significant difference in % tuber incidence among soil type or inoculum level (Fig. 4.1). All three years of field data (avg. 40% tuber

incidence) were significantly lower than the greenhouse data (avg. 85% tuber incidence) for soil type and inoculum level.

For the cultivar Rio Colorado, all three years of field data were the same across all three inoculum levels for soil type 1 and for the 1 sb/g and 5 sb/g inoculum levels in soil type 2 when looking at % tuber incidence (Fig. 4.2). The inoculum level of 10 sb/g in soil type 2 was significantly different than the 2006 field data.

For the cultivars Rio Grande Russet and Russet Burbank, the % tuber incidence was not high enough to detect any significant difference between soil type, inoculum level, and field results because of tuber resistance to powdery scab lesion development in these cultivars. This resistance has been demonstrated through field trials to be common to russet skinned cultivars. The results obtained from the two cultivars susceptible to tuber lesion development show that there are no major differences between the soil type and inoculum level in % tuber incidence, which may be due to low tuber yields and high uniformity of inoculum in the soil in the 15 cm pots used in the greenhouse when compared with the plants grown in the field.

Severity Index:

The powdery scab severity index for the cultivar DT6063-1R when planted in soil type 2 resulted in similar disease readings across all three inoculum levels (Fig. 5.1). Only the 1 sb/g inoculum level was not significantly different than all three years of field data. For soil type 1, the 5 sb/g inoculum level was significantly different from the other two inoculum levels and all three levels of inoculum were significantly different from the three years of field data.

In the cultivar Rio Colorado, the three inoculum levels in soil type 1 were not significantly different from each other or from the three years of field data (Fig. 5.2). However, the years of field data for 2005 and 2006 were significantly different from each other. For soil type 2, the inoculum levels of 1 sb/g and 10 sb/g were not significantly different from each other or from the 2003 and 2005 field data, but a difference was observed when compared with the

2006 data. There was no significant difference between the 5 sb/g and the 2003 and 2006 years of field data.

The consistency of severity index across soil type and inoculum level in trial 1 varied between the two cultivars susceptible to tuber lesion development. For DT6063-1R, soil type 2 and an inoculum level of 1 sb/g was the best combination for matching field results. Soil type 2 also produced the most consistent results across all three inoculum levels. Soil type 1 was not consistent across inoculum levels and did not match field data. For Rio Colorado, the results show that soil type 1 was more consistent across inoculum levels and all inoculum levels were not significantly different from the three years of field data. Soil type 2 was the same in two of the inoculum levels and two of the years of field data. There was some variability between the two soil types for the potato cultivars DT6063-1R and Rio Colorado, but overall, soil type 2 produced the most consistent results when compared with the field data.

Overall Summary:

The specific environmental requirements necessary for powdery scab development were maintained in the greenhouse for trial 1. Soil temperature was kept in the range of 11-18°C after tuber set (the stage in a potato plant's development when daughter tubers begin to form) based on findings in the literature (Wale 2001) and on the average soil temperature in San Luis Valley fields after tuber set (Figs. 30 and 31). Tuber set typically occurs between 40 to 50 days after planting (DAP), which is when temperatures in the greenhouse were lowered into this favorable temperature range.

In addition to temperature, soil moisture was kept at a relatively high level (< 10 cbars) on average (Fig. 27). This was based primarily on the current literature available for powdery scab symptom expression (Harrison *et al.* 1997) because soil moisture data was not available for SLV field conditions. The soil temperature and soil moisture levels in the greenhouse were kept at a favorable range for powdery scab infection and symptom development. This was verified in

the greenhouse since *S.s.s.* disease infection and powdery scab expression (root galling and lesion severity) were relatively high and comparable with field results for the four cultivars in trial 1. Ideal soil moisture levels were explored further in trial 3.

Overall observations point to a more consistent powdery scab symptom development when using the S2 soil type with the 1 sb/g inoculum level. To justify one soil type /inoculum level over another, it is not appropriate to look at just one powdery scab symptom. Instead, all three symptoms (root gall severity, % tuber incidence, and severity index) need to be examined and compared. Soil type S2 demonstrated more consistency within the inoculum levels used and more closely matched the various field results. While there were no significant differences in the % tuber incidence, there were differences in root gall severity and severity index. These differences and the match with the field justify the use of S2. Additionally, it should be noted that van de Graaf *et al.* (2005) indicated that when comparing soil types, a soil with higher sand content tends to be more conducive to powdery scab development than less sandy soils when soil moisture is high. This may be due to a large pore size in sandy soils which would allow the *S.s.s.* zoospores to move more freely to infect the host tissue.

For inoculum level, the 1 sb/g resulted in more consistent powdery scab expression when comparing with field results for the cultivars evaluated in this trial. Also, as higher levels of inoculum (5 and 10 sb/g) were added to the soil, disease levels did not tend to increase. This agrees with research conducted by van de Graaf *et al.* (2000), which showed that once the optimum inoculum level was reached, *S.s.s.* infection levels did not increase when inoculum levels increased. Therefore, when evaluating both of the potato cultivars susceptible to powdery scab tuber lesion development, soil type 2 and the 1 sb/g inoculum level was the combination which produced the most consistent symptom expression within the greenhouse environment and compared most favorable with field data.

Trial 2: Inoculum Source x Soil Type

Inoculum Source:

In trial 2, the variables of inoculum source and soil type were evaluated using the susceptible potato cultivar Rio Colorado. Across both crops and both soil types, the scab infested seed resulted in lower powdery scab severity (root galls and tuber lesions) than clean seed planted in inoculated soil with 5 sb/g (Figs. 6, 7, and 8).

Root gall severity was significantly higher in the S2 inoculated soil than the scab infested seed and was the same for the S1 soil in crop 1. For crop 2, the S1 inoculated soil was significantly higher than the scab infested seed, with a lower overall severity for the S2 inoculated soil than crop 1. For both crops where differences were observed, the inoculated soils had a higher disease severity than soils which were inoculated only with scab infested seed.

For % tuber incidence, both inoculum sources across both soil types were not significantly different for crop 1 (nearly 100%). In crop 2, the scab infested seed source was significantly lower than the inoculated soil for both soil types. For powdery scab severity index, the inoculated soil was significantly higher than the scab infested seed source in soil type 2 for crop 1 and was higher for both soil types in crop 2.

Overall, the severity index was higher in the plants which started from scab-free seed grown in inoculated soil than in plants started from scab infested seed. Only in % tuber incidence (crop 1) was there no significant difference between the two inoculum sources. This may have been due to the small pot size and the ability for zoospores to travel the short distance to infect the tubers during tuber set. Since severity index was lower in the scab infested seed source pots, the total number of infections producing tuber symptoms was lower in these pots. One could speculate that as tuber set and development continued in the seed source pots, the number of sporeballs adjacent to the developing roots and tubers continued to decrease as tubers and roots grew away from the infected seed piece. The number of secondary infections would have been less as the tubers developed away from the seed piece. The clean seed planted in inoculated soil

had an opposite effect because sporeballs were mixed uniformly throughout the soil which would have kept the roots and tubers adjacent to sporeballs throughout tuber set and maturity. This could explain why % tuber incidence was high for both inoculum sources but severity index was higher in the 5 sb/g inoculated soil.

Watering regime between crops:

There are distinct differences between the two crops in trial 2 which need to be addressed. Overall, disease levels were higher in crop 1 than in crop 2 (Figs. 6, 7, and 8). Also, soil type 1 produced higher powdery scab disease values than soil type 2 for crop 2, but not in crop 1. One possible reason for these differences is the irrigation regime used for each crop. The drip system for both crops was set up with 3.8 liters per hour (lph) emitters but the crops were grown nearly a year apart. The water pressure available between crops was different due to differences in the total number of treatments utilized and water consumption throughout the greenhouse during the growth and maturing of each crop. The watering regime for crop 1 averaged one irrigation event every two days during and after tuber set, resulting in approximately 160 ml of water per pot per irrigation event. In crop 2, plants were irrigated every 3 days resulting in 300 ml of water per pot per irrigation event. The differences between irrigation regimes more than likely had an effect on disease severity. Both irrigation regimes provided the same total amount of water for each crop; however, the crop 2 regime allowed the soil to dry more between irrigations (Fig. 28). This reduction of available free water in the soil should have reduced the number of zoospores infecting both roots and tubers as based on the literature. This provides a reasonable explanation for the lower disease levels in crop 2 when compared with crop 1.

These results suggest that adjusting irrigation during a potato plant's tuber set stage could be used by potato producers to decrease disease levels in the field. Based on the data from trial 2, irrigating more per irrigation event but less frequently may have an impact on reducing powdery

scab severity in the field. This would have the most impact on disease severity during tuber set since the daughter tubers are most susceptible to *S.s.s.* infections during this tuber set stage (Wale 2001).

Due to the complexity of this disease, the available literature does not always agree where soil type and soil moisture are concerned. Information presented by Wale (2001) indicates that clay soils are more conducive to powdery scab development and that sandy soils can result in high disease levels when the soil undergoes wet/dry cycles, which does not agree with the findings from trial 2 of this project. However, van de Graaf *et al.* (2002) found that sandy soils were more conducive to powdery scab development than clay soils and soils kept in constant moisture resulted in higher disease levels than in soils with fluctuating soil moisture. It appears that there are more factors affecting powdery scab severity than just soil type and soil moisture when the other conditions which are known to be necessary for powdery scab development appear to be in the favorable range.

Managing irrigation timings for the control of powdery scab may be a good strategy to utilize in the field. However, due to inconsistencies in the literature, more evaluations of differing irrigation regimes during tuber set need to be conducted in the San Luis Valley. This needs to be evaluated under field conditions and in additional greenhouse trials before recommendations on irrigation management can be made to potato producers for the management of powdery scab.

Temperature effects:

Temperature differences may have also played a role in the disease levels varying between both crops in trial 2 and between results from trials 1 and 2. In crop 1 of trial 2, the average root gall severity (3.4) and tuber severity index (420) for the cultivar Rio Colorado were higher than the severities found in trial 1 (1.8 and 175) with the 5 sb/g inoculum level (Figs. 2.2 and 5.2). In crop 1, due to limited space, the trial was planted on a bench that was closest to a

swamp cooler. This would have decreased the soil temperature and in turn increased the soil moisture due to less evaporation (Figs. 29 and 33). This could explain why crop 1 of trial 2 had higher disease levels than trial 1 for the same cultivar. Since all of crop 1 (trial 2) was located on the same bench, temperature and moisture would have been the same throughout the crop. Any differences between the two inoculum sources in crop 1 would not have been affected by varying soil temperature and moisture.

Overall Summary:

Based upon results from trial 2 and earlier discussion, a soil which is thoroughly mixed with *S.s.s.* inoculum was determined to be a better choice than using scab infested seed for obtaining accurate disease results. In addition, scab infested seed is not always an option for use in a greenhouse due to the limited availability of this seed in cultivars which are resistant to tuber lesion development (e.g. russets).

Trial 3: Soil Moisture x Inoculum Level x Cultivar

To determine the proper soil moisture level for optimum powdery scab symptom expression, two different irrigating regimes were evaluated. Plants in each regime were irrigated the same number of times with the amount of water per irrigation event differing between the two regimes (Fig. 32) and this trial was repeated in two crops. According to current literature, soil moisture plays a key role in the infection of potato roots and tubers by *S.s.s.* (Harrison *et al.* 1997). The proper soil moisture level needed to be determined before a greenhouse assay for screening potato germplasm for powdery scab could be adequately developed.

Root Gall Severity:

In the potato cultivar DT6063-1R, there was no difference between the two irrigation regimes for root galling in crop 1 (Fig. 9). In crop 2, the irrigation regime with half the water

level resulted in higher root gall severity than the high irrigation regime.

In the cultivars Rio Colorado and Russet Burbank there was no significant difference between the two irrigation regimes in root gall severity (Figs. 10 and 12). However, in the cultivar Rio Grande Russet, the high irrigation regime resulted in significantly higher levels of root galling than in the half irrigation regime for crop 1 (Fig. 11). In crop 2, no significant difference was observed between irrigation regimes.

Three potato cultivars (DT6063-1R, Rio Colorado, and Russet Burbank) did not appear to have a consistent advantage or disadvantage between the two irrigation regimes for root gall severity. However, the cultivar Rio Grande Russet showed a significant reduction in root gall severity when the half irrigation regime was utilized. This situation may have been due to the fact that the water reduction between the two irrigation regimes was insufficient to impact spore germination and root infection for the three cultivars. These cultivars might also have a higher susceptibility to root galling than Rio Grande Russet, which has been observed to be quite low in field trials. This difference in response to irrigation may provide insight into cultivar variation and resistance.

% Tuber Incidence and Severity Index:

In the cultivars DT6063-1R and Rio Colorado, there was no significant difference between irrigation regimes for % tuber incidence (Figs. 9 and 10). The level of disease was not high enough in Rio Grande Russet or Russet Burbank to analyze for tuber lesion % incidence and severity index.

Severity index levels in the cultivar DT6063-1R did not show a significant difference between irrigation regimes (Fig. 9). However, the overall means from both crops indicated a trend for higher disease severity in the high moisture regime (avg. 285) than in the low moisture regime (avg. 205). In Rio Colorado, the severity index was significantly higher in the high

moisture regime than in the low moisture regime for both crops (Fig. 10). This was also evident in Rio Grande Russet for root gall severity (see root gall discussion on pages 43 and 44).

Overall Summary:

Differences in root gall severity and severity index were observed between the two irrigation regimes in the cultivars Rio Grande Russet and Rio Colorado. While % tuber incidence was not different, there was a trend in Rio Colorado toward higher incidence levels when the high soil moisture regime was used. A high soil moisture irrigation regime (< 10 cbars) during a potato plant's tuber set stage results in significantly higher powdery scab severity index levels in a greenhouse grown crop for the cultivar Rio Colorado.

Not all of the cultivars evaluated expressed a difference when grown under different irrigation regimes, but two of the evaluated cultivars had lower severities when grown under the half irrigation regime. To explain these differences, field observations with these cultivars should be explored. The cultivars Rio Colorado and Rio Grande Russet, which both showed reductions in either root galling or tuber severity index, could be reducing the levels of exudates they release into the soil because of a perceived water stress when watered under the half irrigation regime. Current literature suggests that plant exudates may be triggering the release of *S.s.s.* zoospores from the sporeball and attracting them to the plant tissue for infection. With a potential reduction in exudates, the number of *S.s.s.* infections could be reduced in these cultivars.

A perceived water stress may also trigger a resistance response in the potato cultivars with lower powdery scab severity under the half irrigation regime. This resistance response could account for lower severity levels. In the case of Rio Grande Russet, the cultivar is known for its deep root system and water scavenging ability within the soil profile. The ability of this cultivar to more fully utilize scarce water may tend to favor the potato over the pathogen in the cycle of spore release, infection, and symptom development. In any case, when looking at screening potato germplasm for susceptibility to powdery scab, the high soil moisture irrigation regime appears to be the most useful for this approach.

Factors influencing plant health:

There has been documentation showing that there can be a decrease in potato plant health as a result of powdery scab symptom expression in roots and tubers if disease levels are high (Harrison *et al.* 1997). Therefore, data on root weight, tuber number, and overall tuber weight was collected in addition to disease data for trial 3. A decrease in fresh root weight, tuber number and size was observed in inoculated soil when compared with un-inoculated soil under the high soil moisture irrigation regime. Each cultivar showed a trend towards reduced plant health in at least one of these three areas.

In the cultivar DT6063-1R, the total tuber weight across both crops was significantly lower when grown in the inoculated soil than in the un-inoculated soil, but there was no significant difference in fresh root weight or tuber number (Fig. 9). In Rio Colorado, total tuber number in crop 1 and total tuber weight in crop 2 were significantly lower in the inoculated soil (Fig. 10). In Rio Grande Russet, the fresh root weight and the total tuber number was significantly less when planted in the inoculated soil for crop 2 (Fig. 11). A decrease in total tuber number occurred in Russet Burbank when planted in inoculated soil in the half irrigation regime. The only example of plant health improving in inoculated soil, resulting in higher total tuber weight, was for the cultivar Russet Burbank in crop 2 (Fig. 12). This, however, appears to be an anomaly because the opposite trend was observed in the other three cultivars and in total tuber number for Russet Burbank.

In addition to the presence of inoculum in the soil, tuber yield can also be affected by a low amount of available water in the soil during tuber set. In the cultivar Rio Colorado, the total number of tubers in un-inoculated soil A1 versus B1 was significantly less in the lower moisture soils than in the higher moisture soils for crop 1 (Fig. 10). However, this trend was not consistently observed throughout all of the cultivars evaluated in trial 3. For a potato producer, the limiting of irrigation water during tuber set may result in a decrease in powdery scab

symptoms in susceptible potato cultivars based on results found in this trial and in current literature (Harrison *et al.* 1997). A downside to this management practice is a possible decrease in tuber yield resulting from limiting the water supply to the potato plant during tuber set, which was observed in this trial for the cultivar Rio Colorado. For this reason, limiting the total amount of water during tuber set is not recommended in all situations as a control measure for powdery scab.

The results from trial 3 indicate that susceptible potato cultivars grown in conditions favorable for adequate powdery scab symptom development tend to decrease root health and overall yield of tubers produced by the plant. This indicates another disadvantage, in addition to cosmetic tuber damage, for susceptible potato plants grown in conditions favorable for powdery scab development. This has also been suggested by the literature currently available on powdery scab (Harrison *et al.* 1997).

Phase 1 - Conclusions:

The variables of soil type, inoculum level, inoculum source, and soil moisture were evaluated in a greenhouse for determining symptom expression of powdery scab in four potato cultivars. Symptom expression was evaluated based on the combination of these variables which produced symptoms that most closely matched field data, provided the most consistent disease expression, and produced the highest disease levels when applicable. The red potato cultivars DT6063-1R and Rio Colorado are highly susceptible to root gall and tuber lesion development. Rio Grande Russet is considered resistant to tuber lesion development and has a low susceptibility to root gall development. Russet Burbank is also considered resistant to tuber lesion development and has a medium susceptibility to root gall development.

This project evaluated and tested one possible greenhouse assay for the screening of powdery scab resistant germplasm. The greenhouse assay developed in Phase 1 was found to produce the most appropriate powdery scab severity levels in a greenhouse when compared with

SLV field results. Based on the results from Phase 1, the variables that were found to be the most appropriate for evaluation of potato germplasm resistance to powdery scab in a greenhouse were: a soil type with 50% sand, an inoculum level of 1 sb/g of soil (mixed uniformly throughout the soil), and a high soil moisture (an average between 5 to 10 cbars) during and after tuber set. These variables, in addition to a soil temperature range of 11-18°C during and after tuber set, were found to produce greenhouse results that best matched SLV field data for the four cultivars evaluated and were used in Phase 2 (the testing phase) of this project. Based on the literature, different soil types and irrigation regimes could be used to produce disease levels similar to those found in Phase 1 (Wale 2001).

CHAPTER 5 – PHASE 2 (RESULTS AND DISCUSSION)

Preface:

The objective of Phase 2 was to test and evaluate the greenhouse growing conditions from Phase 1 trials which were determined to be the most consistent and accurate in developing powdery scab symptoms in potato cultivars with varying susceptibility. An important goal was to identify the greenhouse conditions needed to develop powdery scab disease results in susceptible potato cultivars that are comparable with SLV field results. In Phase 1, results from trial 1 were compared to SLV field results with good success.

In Phase 2, fourteen potato cultivars were evaluated for root gall severity, % incidence of tuber lesions, and tuber lesion severity index using the greenhouse assay developed in Phase 1. The fourteen cultivars were selected based on varying levels of susceptibility to powdery scab development on roots and tubers (Tables 1 and 2). Disease data from each cultivar was then compared with three years of SLV field data. Since all fourteen cultivars were not evaluated in the field during the same years, data for each cultivar was used from the three most recent years when comparing greenhouse data to field data.

Results from Phase 2 indicate greenhouse screening of cultivars with varying levels of susceptibility to powdery scab can be more consistent and accurate than field comparisons. All research information supports the use of this greenhouse assay as an appropriate method for the screening of potato germplasm for powdery scab susceptibility.

The effects of *S.s.s.* sporeball contamination and a possible source:

For much of Phase 1 and Phase 2, the un-inoculated control pots contained *S.s.s.* sporeball contamination which needs to be addressed. The contamination was present in every experiment and varied between different crops. When contamination was present in un-inoculated soils, powdery scab % incidence levels were typically more comparable with results from the inoculated soils than severity index levels for the same potato cultivar. In pots which

were intentionally inoculated, a substantial increase in disease level was not observed in trials with highly contaminated un-inoculated pots (based on root gall severity and % incidence) versus un-inoculated pots with low levels of contamination matching results found in Phase 1.

In Phase 1 (trial 2, crop 2, soil type 1), no powdery scab lesions were observed in the un-inoculated control. However, tubers from the inoculated soil had a severity index that was not significantly different from tubers in crop 1, which had contamination in the un-inoculated control. The level of contamination present in the trials did not significantly affect disease results.

There are a few possible sources for the observed soil contamination in the un-inoculated controls. In Phase 1 (trial 1), inoculated and un-inoculated pots were randomized and placed on the same benches as each other. Possible contamination could have occurred with splash during fertilizer events which may have moved sporeballs from one pot to another. During cooling events, wind caused by the exaust fan could have moved spores from inoculated to un-inoculated soils or contaminated field soil could have been sucked in from the outside. In the cultivars susceptible to powdery scab tuber lesion development, seed could have had unobserved small lesions or sporeballs which would have resulted in contamination. However, this explanation does not necessarily apply to contamination found in the russet potatoes. Also, when soil was mixed and placed in pots, contamination could have occurred at this time. However, several precautions were employed to limit any possible contamination (e.g. mixing and planting seed in the un-inoculated soil before any inoculum was ever used).

The most likely source of contamination was from the sand used for the soil. The sand was not sterilized prior to use and was obtained from an outside source, but was the same as used in the breeding program located at the SLV Research Center. This would also explain why, in Phase 1 (trial 2, crop 2, soil type 1) there was no contamination detected, but in crop 1, contamination was present (Figs. 6-8). Crop 2 was planted and grown nearly one year after crop 1 was planted. The sand used for this second crop was not the same sand that was used for crop

1; therefore, the level of contamination from the crop 2 sand could have been much lower than in the sand used in crop 1. Sand containing *S.s.s.* sporeballs, one of the previously mentioned contamination sources, or a combination of several sources is likely to have resulted in the observed contamination throughout phases 1 and 2.

Evaluation of Cultivars:

Potato cultivars with high susceptibility to powdery scab tuber lesion development: (DT6063-1R, Rio Colorado, VC0967-2R/Y, Mountain Rose)

Each of the four red-skinned potato cultivars used in this phase were chosen because of their high susceptibility to powdery scab root gall and tuber lesion development. VC0967-2R/Y tubers have yellow flesh and Mountain Rose tubers have red flesh. The cultivars, DT6063-1R and Rio Colorado, produce tubers with white flesh and were also used throughout Phase 1.

Results:

When evaluating the cultivar DT6063-1R, root gall severity readings were not significantly different between the two greenhouse crops or between the three years of field data (Fig. 13). When comparing the three years of field data to the two greenhouse crops, a significant difference was observed only between the 2005 field data and greenhouse crop 2. For % tuber incidence, the two greenhouse crops were not significantly different from each other and field data was significantly different across all three years (Fig. 13). When comparing the two greenhouse crops to the three years of field data, a significant difference was detected between greenhouse crop 1 and the 2005 and 2006 field data. For severity index (SI), there was no significant difference between the two greenhouse crops or between greenhouse crop 2 and the three years of field data (Fig. 13). There was a significant difference between greenhouse crop 1 and the 2005 or 2006 field data. For the three years of field data, 2005 and 2006 (avg. SI = 100) were significantly different from 2003 (SI = 325). High variability existed between the three years of field data for SI. However, there was no significant difference between crop 2 and the three years of field data, despite the high variability in the field.

The cultivar Rio Colorado resulted in no significant differences between the two greenhouse crops or between the greenhouse crops and the three years of field data for root gall severity (Fig. 14). The three years of field results were significantly different from each other. For % tuber incidence, there were no significant differences between either greenhouse crop or the three years of field data (Fig. 14). For severity index, there were no significant differences between greenhouse crops or between greenhouse crop 2 and the three years of field data (Fig. 14). There was a significant difference between greenhouse crop 1 and the 2006 year of field data. The three years of field data were significantly different from each other for severity index.

For the cultivar VC0967-2R/Y, there were no significant differences between either of the two greenhouse crops or the three years of field data for root gall severity (Fig. 15). For % tuber incidence, there was a significant difference between the two greenhouse crops but there was no significant difference between either greenhouse crop compared to the 2005 field data. The 2005 field data was significantly different from the 2004 and 2006 field data for % tuber incidence. The average % tuber incidence for the 2004 and 2006 field data (25%) was much lower than the two greenhouse crops and the 2005 field data (avg. for all three = 80%) (Fig. 15). For severity index, there were no significant differences between the two greenhouse crops or the 2005 field data (Fig. 15). The 2005 field data (SI = 240) was significantly different than the 2004 and 2006 years (avg. SI = 55). A large degree of variability existed between different years of field data for the cultivar VC0967-2R/Y.

In the cultivar Mountain Rose, there were no significant differences between the two greenhouse crops or when comparing the two greenhouse crops with the 2005 and 2006 years of field data for root gall severity (Fig. 16). The 2004 field data was significantly lower than the other two years of field data. For % tuber incidence and severity index, there were no significant differences between the two greenhouse crops (Fig. 16). Greenhouse crop 1 was significantly higher than the 2004 and 2006 field data, and crop 2 was significantly higher than the 2004 field

data field for % tuber incidence and severity index. The three years of field data were significantly different from each other when looking at % tuber incidence and severity index.

Discussion:

When looking at disease readings for the potato cultivars of DT6063-1R and Rio Colorado, there were no significant differences between the two greenhouse crops. For the three years of field data, there were significant differences for four disease readings (only root gall severity in DT6063-1R and % incidence in Rio Colorado were not significantly different) out of a total of six disease readings for both cultivars. This shows that the results from trials conducted in a greenhouse are more consistent than results from field trials collected over several years. This is likely due to more consistent environmental conditions in a greenhouse which are more conducive to powdery scab development when compared to environmental conditions in the field.

For the potato cultivars VC0967-2R/Y and Mountain Rose, the only disease reading resulting in no significant differences between years of field data was root gall severity in the cultivar VC0967-2R/Y. When comparing both greenhouse crops with the year of field results that had the highest powdery scab reading for VC0967-2R/Y and Mountain Rose, no disease levels were significantly different from each other except % tuber incidence in VC0967-2R/Y and severity index in Mountain Rose. This suggests that these two cultivars, when grown in a greenhouse environment that is conducive to powdery scab development, result in more consistent expression of disease symptoms when compared to the same cultivars grown in field trials for several years.

It is important to note that field variation can give the impression that a cultivar has powdery scab resistance when indeed it does not. For instance, cultivars VC0967-2R/Y and DT6063-1R showed what appeared to be levels of resistance for tuber symptoms in two out of three years of field testing. If those two years were used to determine resistance, a false impression would be obtained suggesting resistance was real. Using the greenhouse results,

however, dispels this by demonstrating that both cultivars have much higher tuber susceptibilities. The greenhouse system gives a more realistic scenario for resistance screening.

Potato cultivars with moderate susceptibility to powdery scab tuber lesion development: (Purple Majesty, VC1002-3W/Y, Atlantic, Superior)

Each of these four potato cultivars was used because they had a relatively moderate susceptibility to powdery scab tuber lesion development. Purple Majesty produces tubers with purple skin and purple flesh. VC1002-3W/Y produces tubers with white skin and yellow flesh. Atlantic and Superior produce tubers with white skin and white flesh. The cultivars VC1002-3W/Y and Superior had a high susceptibility to root gall development in the field. Purple Majesty and Atlantic had a relatively moderate susceptibility to root gall development.

Results:

For the cultivar Purple Majesty, there were no significant differences between the two crops grown in the greenhouse or between the two greenhouse crops and the 2005 field data for root gall severity, % tuber incidence, and severity index (Fig. 17). Also, the 2005 field data was significantly higher than the 2004 and 2006 field data for each of the disease readings.

In the case of the cultivar VC1002-3W/Y, there were no significant differences between the two greenhouse crops or between the greenhouse crops and the three years of field data for root gall severity (Fig. 18). For greenhouse crop 1, % tuber incidence and severity index readings were significantly higher than any of the years of field data (Fig. 18). Greenhouse crop 1 was not significantly different from crop 2 or the 2005 field data for % tuber incidence. For severity index, there were no significant differences between greenhouse crop 2 and the 2005 field data. However, the % tuber incidence and severity index results from the 2005 field year were significantly higher than the field results from 2004 and 2006.

The cultivar Atlantic resulted in no significant differences between the two greenhouse crops or between both greenhouse crops and the three years of field data for root gall severity (Fig. 19). Also, for % tuber incidence and severity index, no significant differences were

observed between the two greenhouse crops. Greenhouse crop 1 was not significantly different from the 2005 field data and crop 2 was significantly different from all three years of field data for the % tuber incidence and severity index readings (Fig. 19). Also, field results varied between years for % tuber incidence and severity index.

For the cultivar Superior, there were no significant differences between the two greenhouse crops for root gall severity, % incidence, and severity index (Fig. 20). For the disease readings of root gall severity and % tuber incidence, results were significantly different across all three years of field data but no differences were observed for severity index between the different years. Both greenhouse crops were not significantly different from the 2003 and 2005 field data for root gall severity, and there was no significant difference between both greenhouse crops and the 2003 field results for % tuber incidence. For severity index, all three years of field data were significantly less than the two greenhouse crops.

Discussion:

In the cultivar Purple Majesty, the results from both greenhouse crops were not significantly different from each other. However, when looking at field results, there were high levels of variability between the different years. This again indicates that powdery scab disease results from the field are much more variable across different years than between crops grown in a greenhouse with environmental conditions favorable for powdery scab development. Greenhouse results across the two greenhouse crops were more variable in the cultivar VC1002-3W/Y than in Purple Majesty. However, the greenhouse results were more consistent than field results when examining both cultivars. While they were similar in root galling, the potential for mistakenly thinking that both have resistance to infection and symptom development based on field results is real. The greenhouse trials show that % tuber incidence and severity index are quite similar under the controlled environmental conditions.

For the potato cultivar VC1002-3W/Y, the three years of field data, in addition to the two greenhouse crops, were variable in severity index. This may be due to differences between this

cultivar and the other cultivars evaluated in Phase 2, rather than inconsistencies between the environmental parameters in each of the greenhouse crops. An unknown environmental parameter(s) not evaluated in this project may have varied between the two greenhouse crops and might have no effect on powdery scab development in other cultivars. This parameter(s), in combination with the specific properties of this cultivar, could result in differences in severity index between the two greenhouse crops.

For the potato cultivars of Atlantic and Superior, no significant differences were detected between the two greenhouse crops for all three disease readings. The only disease reading in which field data was the same across all three years was for the severity index readings in the cultivar Superior. In the cultivar Atlantic, variability existed between the two greenhouse crops and field data which may be due to the low levels of tuber lesion severity in this cultivar. Tuber symptom expression for the cultivars of Atlantic and Superior were higher in the greenhouse than in the field. This may be due to greater inoculum uniformity in the greenhouse soil and small pot size restricting tuber number and size which are not present under field conditions.

When comparing the cultivars Atlantic and Superior, the results demonstrate a difference between the greenhouse and the field with the greenhouse being significantly higher in powdery scab severity for roots and tubers. Yet, the results between these two cultivars clearly indicate a difference between root gall severity, % tuber incidence and severity index. In the case of Atlantic, the root gall severity average between crops is 3.1, % tuber incidence is 74%, and severity index is 133.5. Comparing this to Superior shows that Superior is more susceptible to powdery scab. The average for Superior is 3.7 for root gall severity, 96% for % tuber incidence, and 250 for severity index. This indicates that the greenhouse can be utilized to screen germplasm resistance differences.

Potato cultivars with low susceptibility to powdery scab tuber lesion development: (Ranger Russet, Freedom Russet, Rio Grande Russet, Canela Russet, CO94035-15RU)

Each of these six potato cultivars was used because they had a relatively low susceptibility to root gall and tuber lesion development. Each of these cultivars produce tubers which have russet skin and white flesh. Potato tubers with a russet skin tend to be resistant to tuber lesion development. Russet Burbank is the only cultivar in this group that has a relatively moderate root gall susceptibility based on several years of field results.

Results:

For the potato cultivar Ranger Russet, there were no significant differences between the two greenhouse crops for root gall severity (Fig. 21). All three years of field data were significantly different from each other and the greenhouse crop 1 was significantly different from the 2005 and 2006 field years. Greenhouse crop 2 results were significantly different from the 2003 and 2005 field results. For the cultivar Freedom Russet, no significant differences were detected between the two greenhouse crops or between the two greenhouse crops and the 2006 field data for root gall severity (Fig. 22).

The cultivars Russet Burbank and Rio Grande Russet showed no significant differences between the two greenhouse crops for each cultivar when evaluating root gall severity (Fig. 23). For Russet Burbank, both greenhouse crops were not significantly different from the 2005 field year and were significantly lower than the 2003 and 2006 field years. For Rio Grande Russet, there were no significant differences between the three years of field data for root gall severity (Fig. 24). Also, no significant differences were present when comparing field data from 2006 with greenhouse crop 2 data; greenhouse crop 1 had a significantly higher root gall severity than the three years of field data.

When evaluating the cultivar Canela Russet, no significant differences were detected between the two greenhouse crops or between the greenhouse crops and the 2005 and 2006 field

years for root gall severity (Fig. 25). The 2003 field data (0.5) was significantly lower than the other two years of field data and the two greenhouse crops (avg. of all four was 2.5).

For the cultivar CO94035-15RU, there were no significant differences between the two greenhouse crops or between the greenhouse crops and the 2004 and 2006 field data (Fig. 26). The 2005 field data was significantly less than the two greenhouse crops and the 2004 and 2005 field data.

Discussion:

For the cultivars, Ranger Russet and Freedom Russet, a wide range of variability existed in root gall severity between the three years of field data for each cultivar and no significant variability existed between the two greenhouse crops for root gall severity. Since russet skinned potatoes are more resistant to symptom development, the amount of tuber lesion development was low. However, a low level was detected in the field and greenhouse but no significant differences between field years or greenhouse crops were present (Figs. 21 and 22). Only % tuber incidence is being reported because powdery scab severity had a value of 1 for the cultivars of Ranger Russet and Freedom Russet.

For nearly every cultivar evaluated in Phase 2, the crops grown in the greenhouse tended to equal or have higher levels of disease than the field data from the year with the highest level of disease; however, the opposite was true in the case of Russet Burbank grown during Phase 2. Root gall data from Phase 1, trial 1 (Fig. 3.2) for the same soil type and inoculum level showed similar values as the years with the highest levels of root galling (2003 and 2006). A difference in climate or soil type, which effected the reaction of *S.s.s.* to Russet Burbank, must have been present during both crops of Phase 2 which were not present during Phase 1, trial 1. This resulted in lower root gall severity in the greenhouse.

For Rio Grande Russet, there was no variability between the two greenhouse crops or between the three years of field data for root gall severity. For Canela Russet, the high level of variability between the 2003 field results and the 2005/2006 field results illustrate that a

greenhouse assay is necessary for consistent powdery scab development for the evaluation of potato germplasm resistance to root galling.

The cultivar CO94035-15RU showed that root gall severity levels were lower in greenhouse and field trials than in any other cultivar evaluated in Phase 2. Based on these results, this cultivar could potentially be used in a breeding program for the development of a potato cultivar which is fully resistant to powdery scab root galling. Results from Phase 2 also indicate that a cultivar such as Rio Grande Russet could be used for powdery scab resistance development. Low root galling was present in the field and after adjusting for the differences for field and greenhouse environments, Rio Grande Russet still had a relatively low root gall rating. While Rio Grande Russet is a good candidate, results indicate that CO94035-15RU might be a better candidate.

Summary of Cultivar Evaluations:

Overall, the crops grown in the greenhouse, using the environmental and soil conditions which were determined during Phase 1, produced more consistent powdery scab expression than three years of data collected from SLV field trials. Due to fluctuations in field environments throughout different growing seasons, evaluating powdery scab susceptibility should be conducted in a greenhouse setting in order to obtain consistent results for potato cultivars with varying levels of susceptibility to this disease. One major advantage of using the greenhouse over the field is being able to manipulate the environment and soil conditions in order to achieve maximum powdery scab development for every crop.

In addition to consistency of disease symptom expression, conducting powdery scab research in a greenhouse has several advantages over field research. The number of crops is not limited to one per year in a greenhouse as it is in the field. Finding a potato grower that is willing to cooperate with researchers and that has a field with soil which is uniformly inoculated with *S.s.s.* sporeballs and subjected to conditions which are favorable for powdery scab symptom

expression consistently over several years can also be difficult. Additionally, conducting this research in the greenhouse is much more cost efficient than field research.

For nearly every powdery scab symptom which was evaluated for the cultivars in Phase 2, there were no significant differences between the two greenhouse crops. Also, the two greenhouse crops produced similar disease results with at least one or more years of field data. The highest level of powdery scab found throughout the three years of field data typically matched greenhouse results for both greenhouse crops. A greenhouse can be used for evaluating powdery scab expression for potato cultivars of varying susceptibility to powdery scab and will result in more consistent disease expression than in trials conducted in field environments. The greenhouse results tend to match the worst case field scenario for powdery scab expression for each potato cultivar evaluated.

Relative Ranking of Field and Greenhouse:

Throughout Phase 2 of this project, powdery scab symptom expression was higher in the greenhouse than in the field for nearly every cultivar evaluated. Since disease expression was more consistent in the greenhouse than in the field, it makes sense to use this greenhouse assay rather than field trials to evaluate potato cultivars for powdery scab resistance. However, most potato cultivar evaluations on powdery scab resistance have been conducted using field trials and the greenhouse tends to produce higher disease levels than the field. One possible explanation for these differences in disease levels may be the presence of less variable environmental conditions in the greenhouse, resulting in more time that the plants are exposed to conditions favorable for powdery scab symptom expression. Another possible explanation might be a more uniform distribution of *S.s.s.* sporeballs in the soil used in the greenhouse than in naturally infested fields. The dynamics of root structure may also play a role in powdery scab severity levels in the field and greenhouse when looking at root gall severity. In a greenhouse, when potato plants are grown in a 15x15 cm pot, the root mass is confined within a pot. In the field, the roots of a potato

plant are capable of reaching much farther away from the seed piece and possibly beyond the presence of sporeballs, resulting in a fewer number of infections for the entire root mass. For researchers to use the results from this greenhouse assay and compare them with known field results, a relative ranking of cultivar susceptibility needs to be established to obtain an accurate comparison of greenhouse and field results.

To simplify this relative ranking, three levels of powdery scab susceptibility were established (low, medium, and high – see Table 2). All fourteen cultivars evaluated in Phase 2 of this project have been included in a relative ranking table (Table 1). This relative ranking has been developed for root gall severity and tuber severity index. The greenhouse disease values listed in the table are the means of both Phase 2 greenhouse crops and the field disease values are the means from several years of field trials. Powdery scab levels in the greenhouse tend to be higher than field trial data. Due to the high levels of disease in the greenhouse, a scale was developed to appropriately recognize relative ranking of field results and place them at the appropriate level in the greenhouse. For example, Rio Grande Russet was among the lowest ranking for root gall severity in the field over three years (1.2). To compensate for the higher greenhouse results (2.4), the scale was adjusted from 0 to 1.5 for the field up to 0 to 2.5 for the greenhouse. This adjustment still keeps Rio Grande Russet in the low root gall severity category while acknowledging the relatively high root gall severity obtained in the greenhouse.

When examining root gall severity, the relative ranking of the greenhouse to the field tends to give rankings which are mostly consistent for the low, medium, and high levels. For example, the five cultivars which have low root galling in the field also have low root gall readings in the greenhouse. However, there are a few differences when comparing field and greenhouse results. The cultivars of Russet Burbank and DT6063-1R were ranked as low in the greenhouse but were ranked as medium (Russet Burbank) and high (DT6063-1R) in the field. Also, the cultivars of Purple Majesty and Atlantic were ranked high in the greenhouse but were ranked low in the field for root gall severity.

Severity index rankings between field and greenhouse were very similar for all the cultivars evaluated. All six of the cultivars with russet skin were ranked low in the greenhouse and low in the field. Also, four of the five cultivars that had a high ranking in the field also had a high ranking in the greenhouse. Only the cultivars of Purple Majesty, VC1002-3W/Y, and Superior had rankings that differed between the field and greenhouse for severity index.

When examining the fourteen cultivars for root gall severity and severity index, in general the relative rankings remained similar between the field and greenhouse. At most, a cultivar only differed by one ranking level with the exception of root gall severity in the cultivar DT6063-1R. A relative ranking of low was present in the greenhouse but was high in the field for this cultivar. Root mass in DT6063-1R was relatively low when compared with the other cultivars in the greenhouse when roots were evaluated for root gall severity. This has not been observed in the field and may have been a factor of seed quality or other differences in this cultivar when grown in the greenhouse. A relative ranking of low in the greenhouse may have been a result of low root mass, which would have resulted in a fewer number of root galls, rather than a low susceptibility to powdery scab root galling.

The goal of conducting cultivar evaluation trials is primarily to discover potato germplasm that is resistant to powdery scab development. Based on the relative ranking system, all cultivars which were relatively low in the field were also low in the greenhouse for root gall severity and severity index. Using this relative ranking system is necessary for comparing field to greenhouse results when evaluating potato cultivars for resistance to powdery scab.

Phase 2 Conclusions:

Phase 2 of this project took the most appropriate greenhouse conditions determined in Phase 1 and tested those conditions using fourteen potato cultivars with varying susceptibility to powdery scab. The results collected from the greenhouse were then compared with known

powdery scab results from SLV field trials over several years. For the cultivars evaluated, results from two greenhouse crops resulted in more consistent powdery scab levels than field trial disease levels. Also, powdery scab levels from the greenhouse typically equaled disease levels from the field trial year that had the highest severity when evaluating field trials from three different years. The disease potential for each potato cultivar evaluated in Phase 2 was obtained in the greenhouse. Since field trials often result in lower disease levels than the disease potential for each cultivar, the greenhouse is an important tool for determining the highest level of powdery scab a potato cultivar could develop when grown under favorable conditions.

In order to compare known powdery scab levels from the field with powdery scab levels from the greenhouse, a relative ranking needed to be developed since disease levels in the greenhouse were typically higher than field levels. A relative ranking for powdery scab susceptibility of low, medium, or high was assigned to each cultivar for root gall severity and severity index. Overall, the relative ranking assignments given to each cultivar matched field and greenhouse disease levels with only a few exceptions which were discussed earlier. Results from Phase 2 indicate that under the appropriate conditions and using an appropriate ranking system, a greenhouse can be used to accurately determine potato germplasm which are resistant to powdery scab development.

CHAPTER 6 - CONCLUSIONS

Powdery scab is a disease that can damage the roots and tubers of potato plants and has very specific requirements for infection and symptom expression. For determining the level of resistance to this disease in potato germplasm, field trials have previously been utilized for gathering this information. Trials conducted in the field have limitations including variable environmental conditions and the number of trials which can be performed each year due to seasonal restrictions. Powdery scab resistance in potato germplasm can be perceived at varying levels in the field depending on the environmental conditions within a given growing season. The purpose of this project was to determine an appropriate greenhouse environment for consistent powdery scab expression when evaluating potato germplasm for resistance.

In Phase 1, the factors of soil type, inoculum level, inoculum source, soil moisture, and potato cultivar were evaluated based on the levels of powdery scab root gall severity, % tuber incidence and severity index. Consistency in disease severity, relative compatibility with known SLV field results, and the disease potential within each potato cultivar were used as criteria for determining which combination of factors was the most appropriate for powdery scab development in a greenhouse. The factors determined to be the best combination for powdery scab expression in a greenhouse were: a soil with 50% sand, an inoculum level of 1 sb/g mixed uniformly throughout the soil, and a relatively high (0-10 cbars) soil moisture level during tuber set.

In Phase 2, the best combination of factors from Phase 1 was tested using fourteen potato cultivars with varying susceptibility to powdery scab and these results were compared with known SLV field results. Phase 2 results indicated that crops grown in the greenhouse provided more consistent symptom development than when compared with three years of SLV field results. Also, greenhouse results consistently matched results from the field year with the highest level of powdery scab severity. This provides evidence that the powdery scab severity level for a

potato cultivar grown in a greenhouse matches the disease potential for that cultivar, which may not be the case when using averaged multiple year field trials for resistance screening. Phase 2 results confirmed the use of the combination of factors determined in Phase 1 for evaluating potato germplasm for powdery scab resistance in a greenhouse.

When comparing the overall mean of field results to greenhouse results for powdery scab severity, the greenhouse tends to produce higher disease levels. A relative ranking system was developed in order to compare field and greenhouse results. When using the relative ranking system for the fourteen cultivars evaluated in Phase 2, it was found that the cultivars with a ranking of low, medium, or high in the field matched the relative ranking from cultivars in the greenhouse in most cases. When using a greenhouse for the evaluation of potato cultivars for powdery scab resistance, accurate comparisons can be made with known field results if this relative ranking system is used. Using the greenhouse assay developed in this project is a suitable method for evaluating potato germplasm for resistance to powdery scab symptom development.

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APPENDIX: TABLES AND FIGURES

Table 1. Potato cultivars with corresponding root gall and powdery scab lesion severity index (average of several years of SLV field results and average of both greenhouse crops – phase 2).

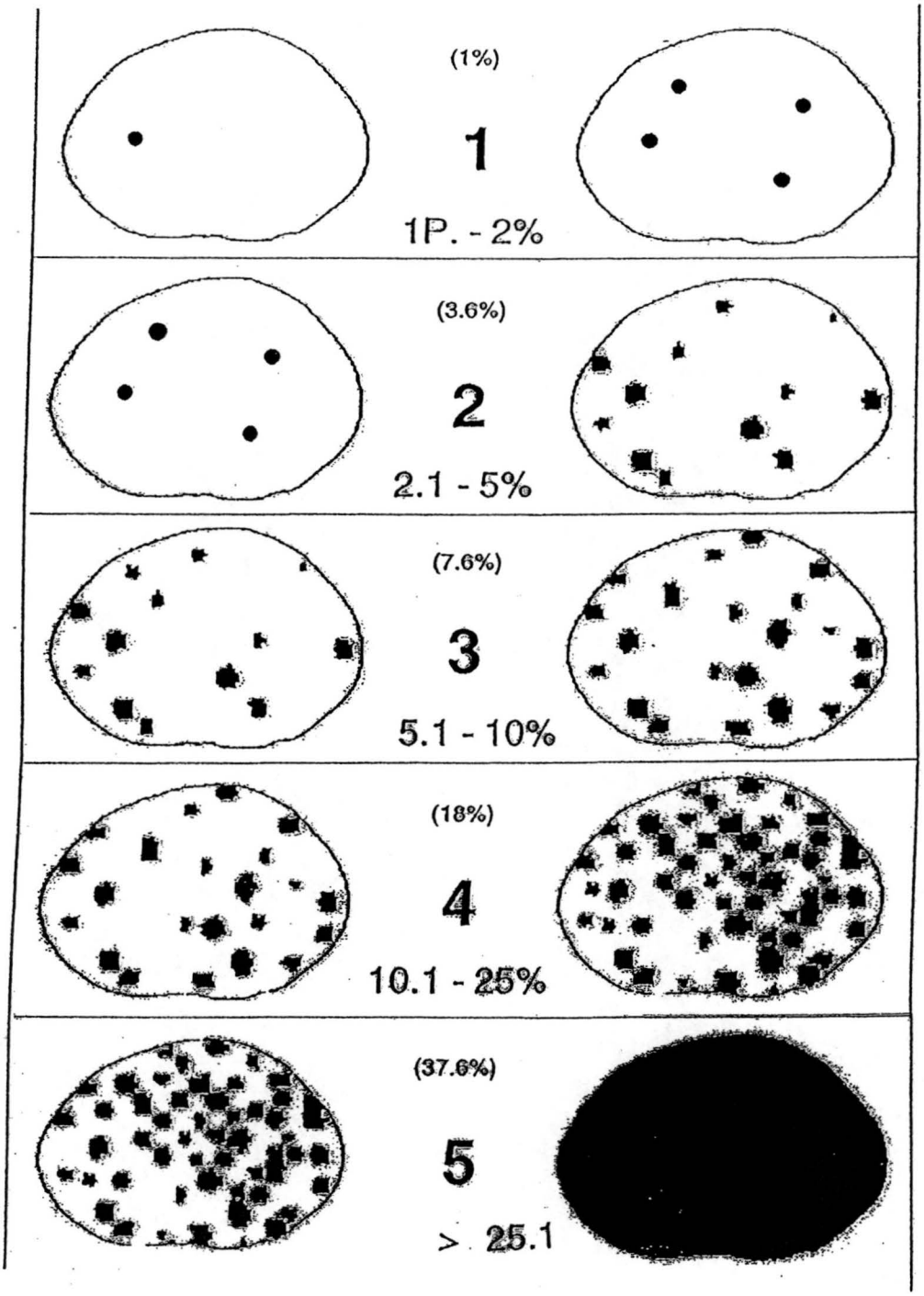
Potato Cultivar	Root Gall Severity (0-4)		Tuber Severity Index: % incidence x severity rating (1-5)		Years of Field Results (i.e., 2002 = 02, etc.)
	Field	Greenhouse	Field	Greenhouse	
1. VC0967-2R/Y	3.8 H (1)[a]	3.8 H (1)	112.9 H (3)	220.8 H (5)	Three (04,05,06)
2. DT6063-1R (Cherry Red)	2.8 H (4)	2.3 L (10)	100.8 H (4)	275.0 H (3)	Four (02,04,05,06)
3. Mountain Rose (CO94183-1R/R)	2.3 M (8)	3.0 M (6)	199.0 H (1)	284.0 H (2)	Three (04,05,06)
4. Rio Colorado (NDC5281-2R)	2.5 M (5)	2.6 M (7)	193.7 H (2)	303.0 H (1)	Four (02,03,05,06)
5. Purple Majesty (CO94165-3P/P)	2.3 M (7)	3.4 H (4)	79.1 H (5)	157.0 M (7)	Three (04-06)
6. VC1002-3W/Y	3.7 H (2)	3.7 H (2)	54.7 M (7)	200.0 H (6)	Three (04,05,06)
7. Superior	2.8 H (3)	3.7 H (3)	67.6 M (6)	250.0 H (4)	Five (02-06)
8. Atlantic	2.4 M (6)	3.1 H (5)	24.1 M (8)	133.5 M (8)	Five (02-06)
9. Russet Burbank	2.3 M (9)	1.3 L (13)	0.0 L (12)	0.0 L (12)	Five (02-06)
10. Ranger Russet	1.4 L (11)	2.0 L (12)	3.7 L (9)	13.2 L (10)	Five (02-06)
11. Canela Russet (AC92009-4RU)	1.4 L (10)	2.5 L (8)	0.0 L (11)	1.5 L (11)	Four (02,03,05,06)
12. Rio Grande Russet (AC89536-5RU)	1.2 L (12)	2.4 L (9)	0.0 L (13)	0.0 L (13)	Three (02,03,06)
13. Freedom Russet	0.9 L (13)	2.1 L (11)	2.5 L (10)	16.0 L (9)	Four (03-06)
14. CO94035-15RU	0.7 L (14)	1.2 L (14)	0.0 L (14)	0.0 L (14)	Three (04,05,06)

[a]( ) = relative numerical rank of powdery scab severity.

Table 2. Powdery scab ranking of tuber and root gall severity for field and greenhouse trials.

Relative Ranking for Root Gall Severity (maximum rating = 4)			Relative Ranking for Tuber Severity (maximum rating = 500)	
Disease level	Field	Greenhouse	Field	Greenhouse
Low (L)	0-1.5	0-2.5	0-20	0-50
Medium (M)	1.6-2.5	2.6-3.0	21-75	51-175
High (H)	2.6-4.0	3.1-4.0	76 - 500	176-500

Figure 1. Powdery Scab Scoring Table



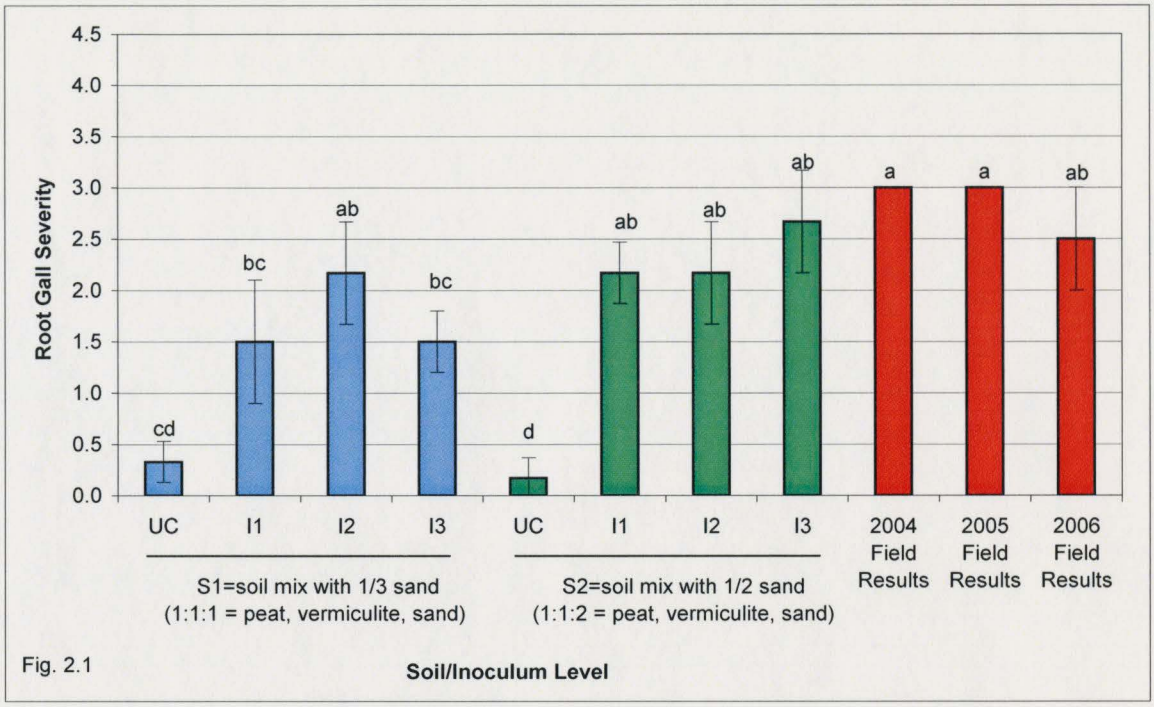


Fig. 2.1

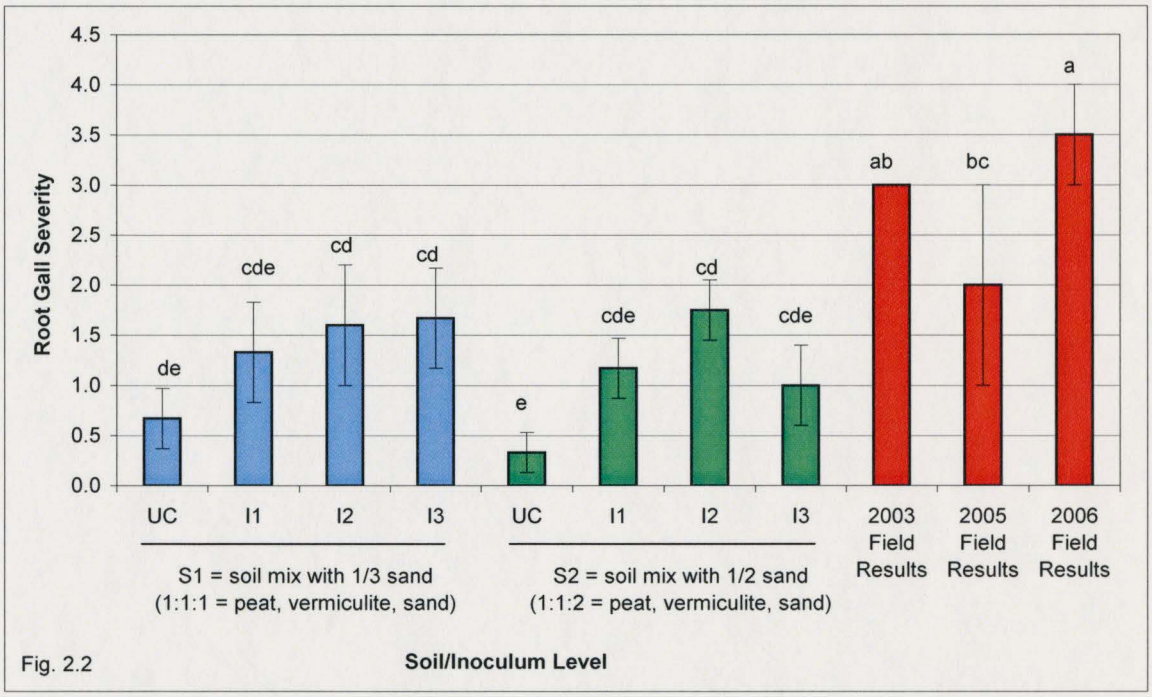


Fig. 2.2

Fugures 2.1 - 2.2. Evaluation of Different Soil Types and Inoculum Levels in a Greenhouse Environment Compared with Three Years of Field Data for Susceptibility to Powdery Scab Root Gall Formation.

Potato cutivar: Figure 2.1 = DT6063-1R (Cherry Red), Figure 2.2 = Rio Colorado (NDC5281-2R). Soil Inoculum levels: UC = uninoculated control, I1 = 1 spore ball (sb)/g of soil, I2 = 5 sb/g of soil, I3 = 10 sb/g of soil. Roots were given a symptom rating of 0 - 4 based on root gall presence and severity (0 = no root galls observed; 1 = one to three small or one large gall; 2 = 4 to 10 small or 2 to 3 large galls; 3 = 11 to 20 small or 4 to 8 large galls; 4 = > 30 root galls observed). The data was analyzed using a LSD separation and all means were compared against each other within each graph. The data are expressed as means +/- standard error. Means followed by the same letters are not significant at P=0.05.

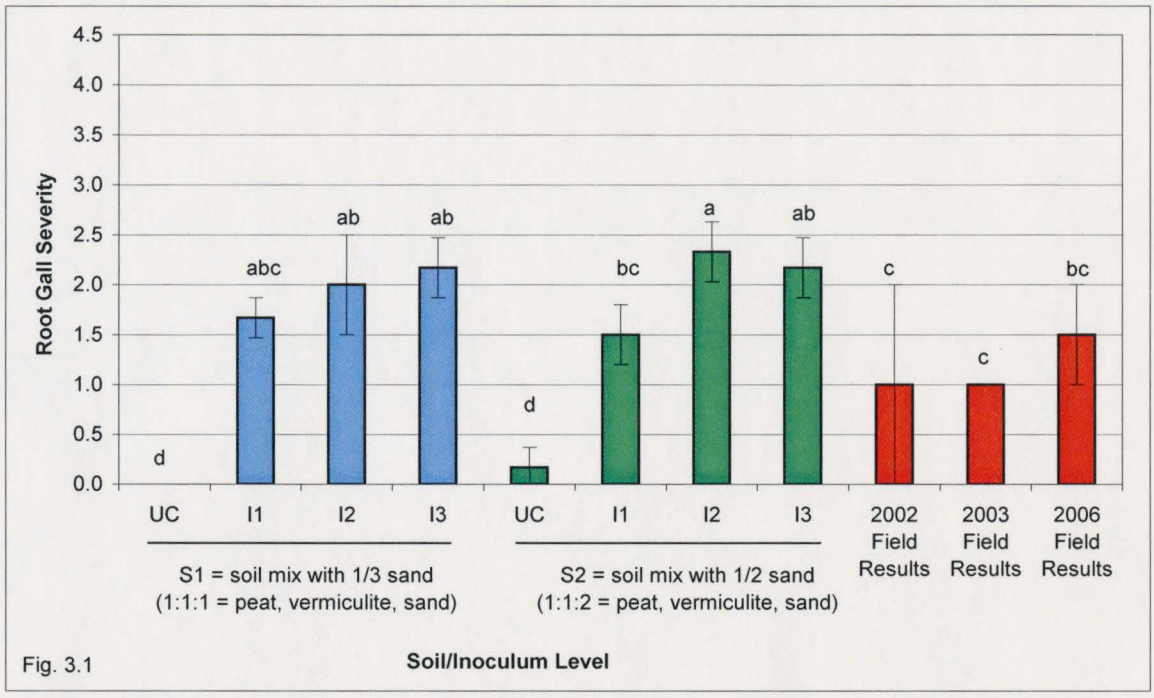


Fig. 3.1

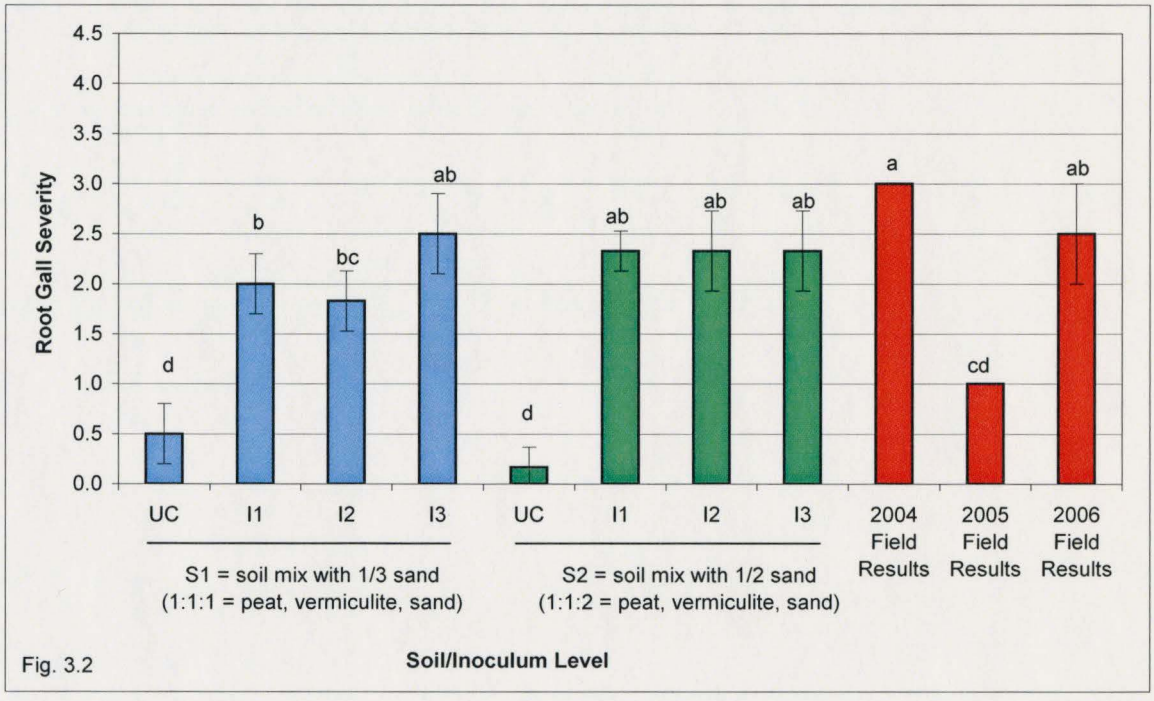


Fig. 3.2

Figures 3.1 - 3.2. Evaluation of Different Soil Types and Inoculum Levels in a Greenhouse Environment Compared with Three Years of Field Data for Susceptibility to Powdery Scab Root Gall Formation.

Potato cutivar: Figure 3.1 = Rio Grande Russet, Figure 3.2 = Russet Burbank. Soil Inoculum levels: UC = uninoculated control, I1 = 1 spore ball (sb)/g of soil, I2 = 5 sb/g of soil, I3 = 10 sb/g of soil. Roots were given a symptom rating of 0 - 4 based on root gall presence and severity (0 = no root galls observed; 1 = one to three small or one large gall; 2 = 4 to 10 small or 2 to 3 large galls; 3 = 11 to 20 small or 4 to 8 large galls; 4 = > 30 root galls observed). The data was analyzed using a LSD separation and all means were compared against each other within each graph. The data are expressed as means +/- standard error. Means followed by the same letters are not significant at P=0.05.

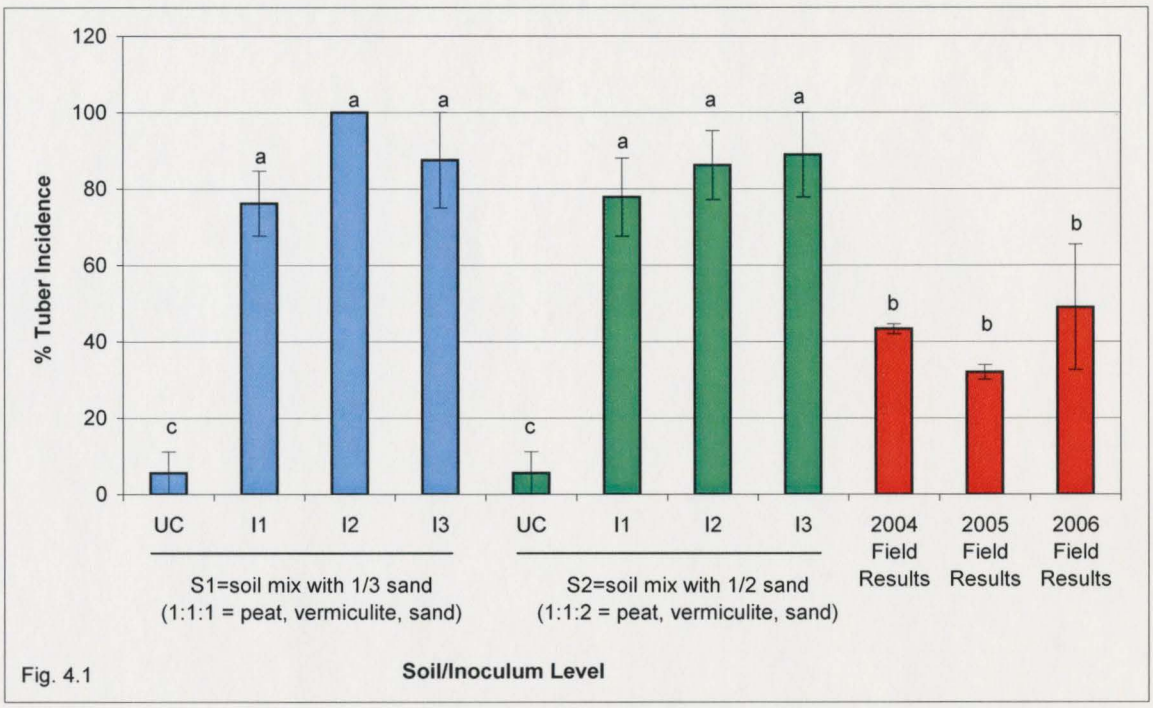


Fig. 4.1

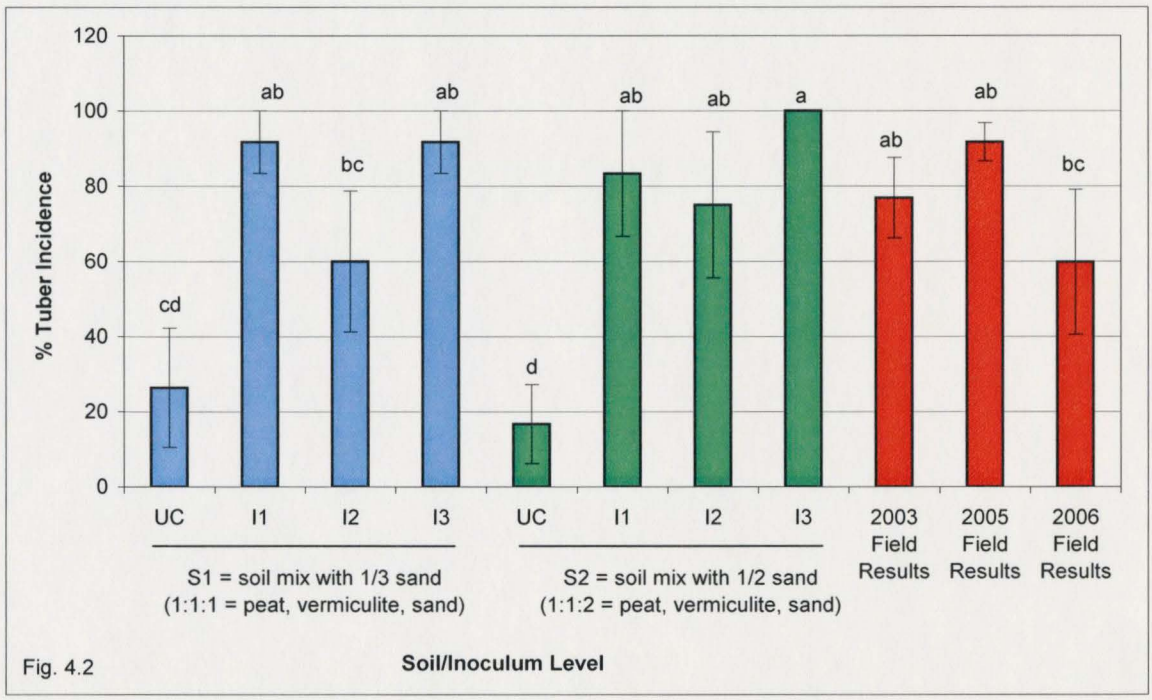


Fig. 4.2

Figures 4.1 - 4.2. Evaluation of Different Soil Types and Inoculum Levels in a Greenhouse Environment Compared with Three Years of Field Data for Susceptibility to Powdery Scab Incidence on Tubers.

Potato cutivar: Figure 4.1 = DT6063-1R (Cherry Red), Figure 4.2 = Rio Colorado (NDC5281-2R). Soil Inoculum levels: UC = uninoculated control, I1 = 1 spore ball (sb)/g of soil, I2 = 5 sb/g of soil, I3 = 10 sb/g of soil. % Incidence = total percentage of tubers with powdery scab lesions at harvest. The data was analyzed using a LSD separation and all means were compared against each other within each graph. The data are expressed as means +/- standard error. Means followed by the same letters are not significant at P=0.05.

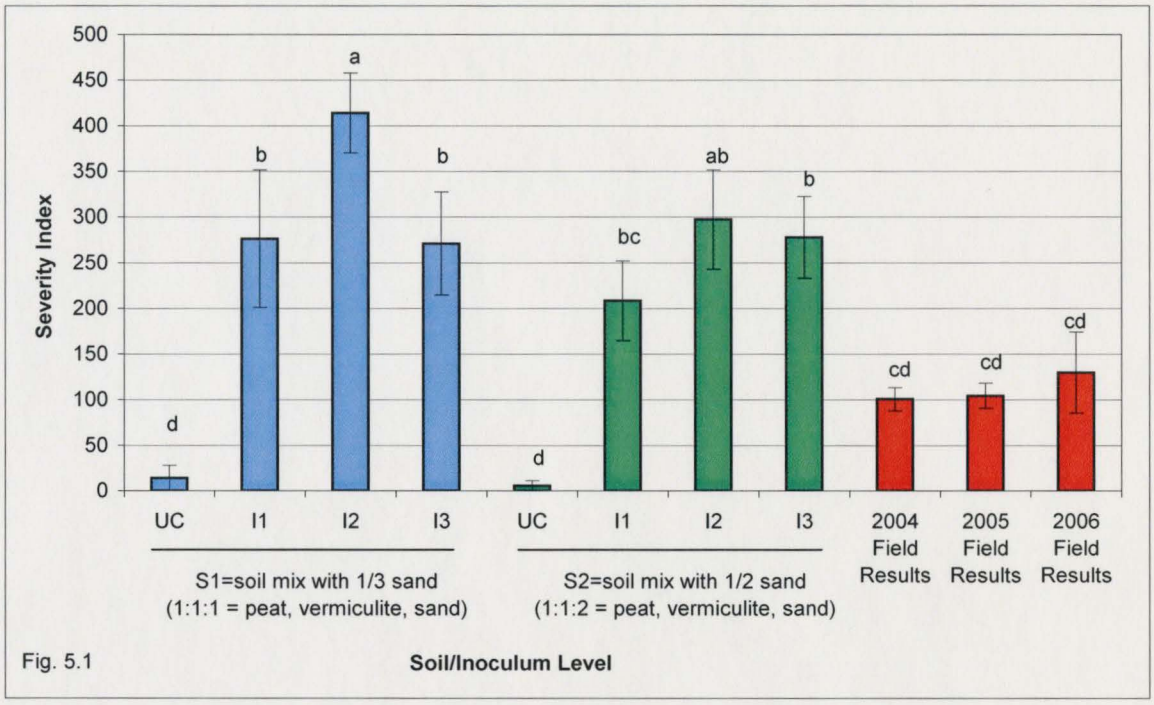


Fig. 5.1

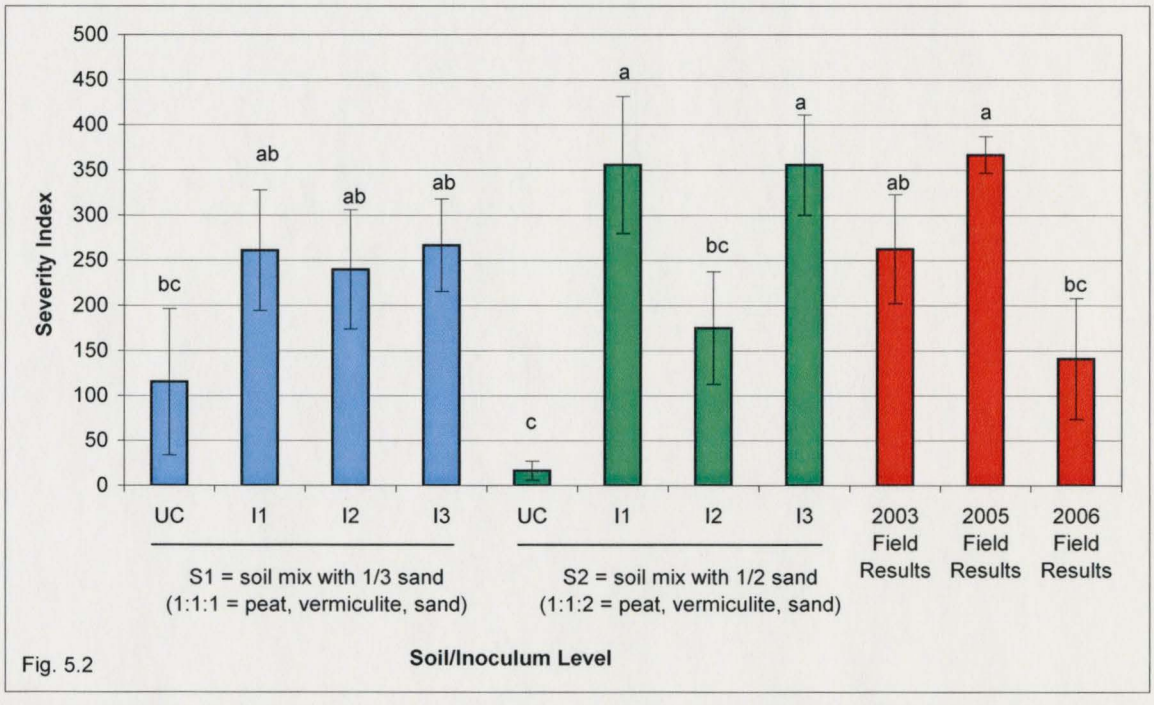


Fig. 5.2

Figures 5.1 - 5.2. Evaluation of Different Soil Types and Inoculum Levels in a Greenhouse Environment Compared with Three Years of Field Data for Susceptibility to Powdery Scab Tuber Lesion Severity.

Potato cutivar: Firgure 5.1 = DT6063-1R (Cherry Red), Figure 5.2 = Rio Colorado (NDC5281-2R). Soil Inoculum levels: UC = uninoculated control, I1 = 1 spore ball (sb)/g of soil, I2 = 5 sb/g of soil, I3 = 10 sb/g of soil. Severity Index = total percentage of tubers with powdery scab lesions at harvest multiplied by the severity of lesions (severity was rated on a scale of 1 to 5 (1=1 lesion to 2% coverage, 2=2.1 to 5%, 3=5.1 to 10%, 4=10.1 to 25%, 5=<25%)). The data was analyzed using a LSD separation and all means were compared against each other within each graph. The data are expressed as means +/- standard error. Means followed by the same letters are not significant at P=0.05.

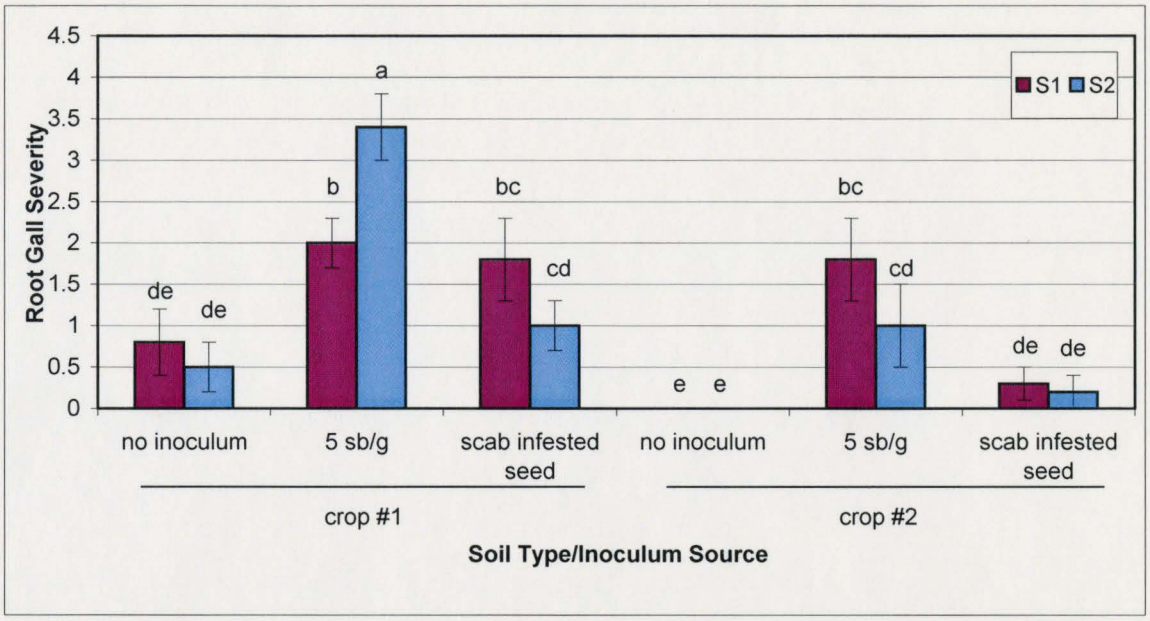


Figure 6. Evaluating two inoculum sources for root gall severity in a greenhouse for the potato cultivar Rio Colorado (NDC5281-2R). Inoculum Source: two sources were evaluated, 5 sb/g=soil was inoculated with 5 sporeballs/g of soil, scab infested seed=seed with has powdery scab lesions. Soil Type: S1=1/3 of soil is sand, S2=1/2 of soil is sand. Data analyzed using a LSD separation, bars with same letters are not significant at P=0.05. Data are expressed as means +/- standard error.

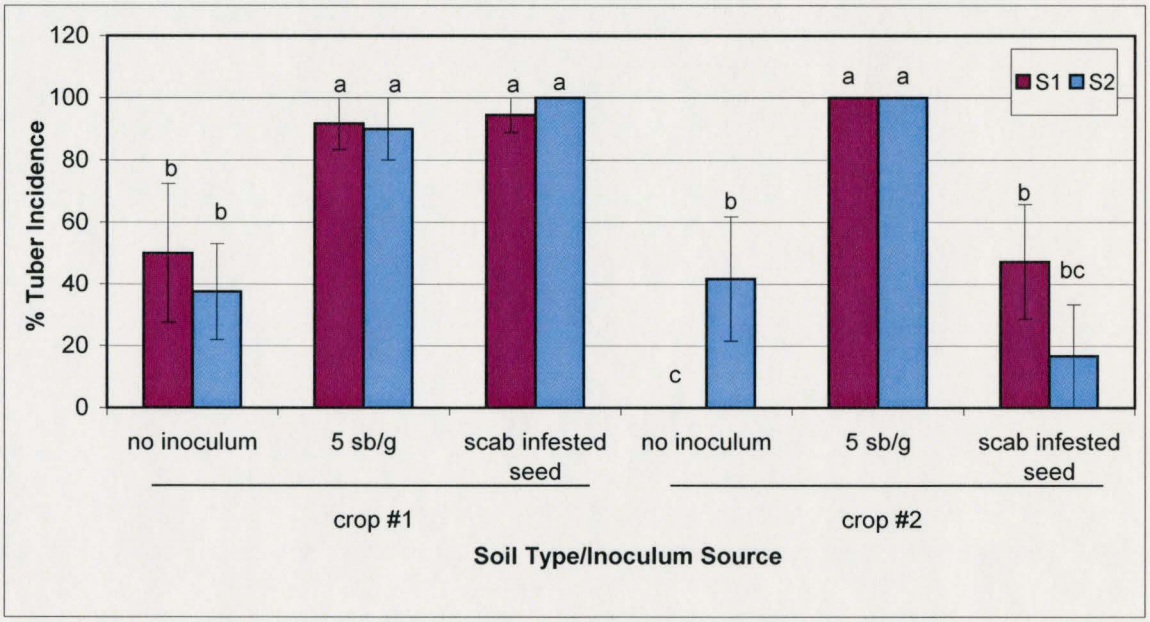


Figure 7. Evaluating two inoculum sources for powdery scab incidence in a greenhouse for the potato cultivar Rio Colorado (NDC5281-2R). Inoculum Source: two sources were evaluated, 5 sb/g=soil was inoculated with 5 sporeballs/g of soil, scab infested seed=seed with powdery scab lesions. Soil Type: S1=1/3 of soil is sand, S2=1/2 of soil is sand. Data analyzed using a LSD separation, bars with same letters are not significant at P=0.05. Data are expressed as means +/- standard error.

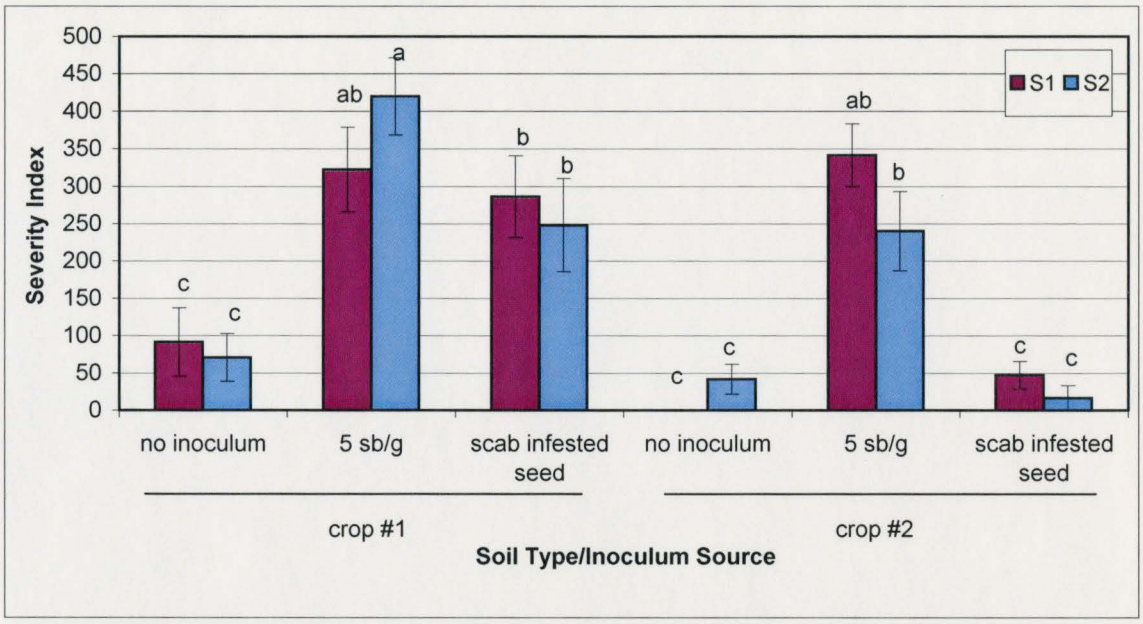


Figure 8. Evaluating two inoculum sources for powdery scab severity index in a greenhouse for the potato cultivar Rio Colorado (NDC5281-2R). Inoculum Source: two sources were evaluated, 5 sb/g=soil was inoculated with 5 sporeballs/g of soil, scab infested seed=seed with powdery scab lesions. Soil Type: S1=1/3 of soil is sand, S2=1/2 of soil is sand. Data analyzed using a LSD separation, bars with same letters are not significant at P=0.05. Data are expressed as means +/- standard error.

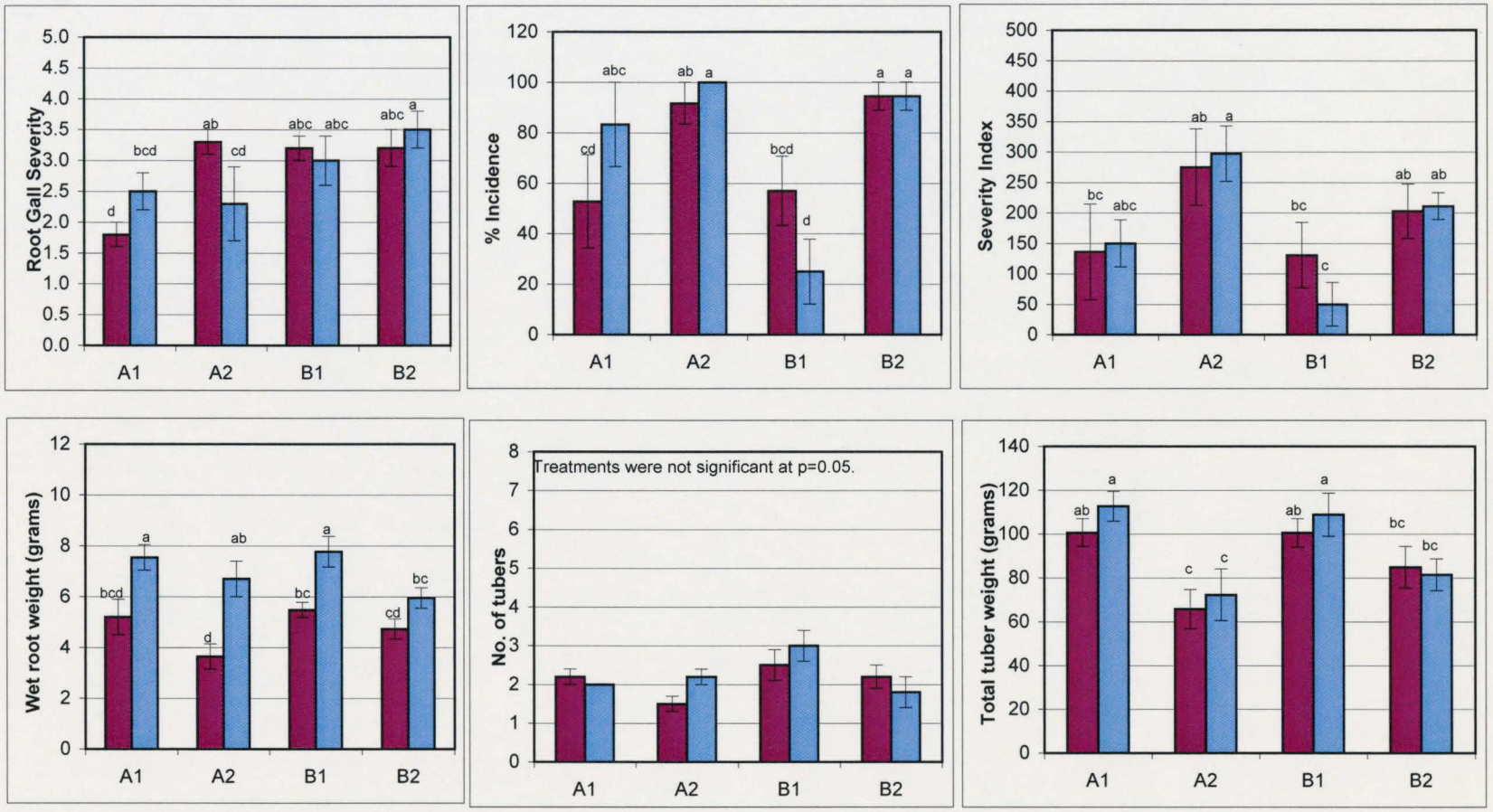


Figure 9. Evaluating powdery scab severity and plant health using two irrigation regimes in a greenhouse for the potato clone DT6063-1R (Cherry Red). X axis for all charts= Soil Type/Irrigation Regime. Trt. A1 = UC-STD, A2 = I-STD, B1 = UC-ALT, B2 = I-ALT. = crop #1 = crop #2

Soil Type: UC=soil is not inoculated, I=soil is inoculated with 1sporeball/g of soil.

Irrigation Regime: STD = Standard Irrigation regime required for powdery scab development, ALT = 50% less irrigation water during the time of tuber set.

Example of x axis treatment abbreviation: (UC-STD = Untreated control soil, standard irrigation)

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

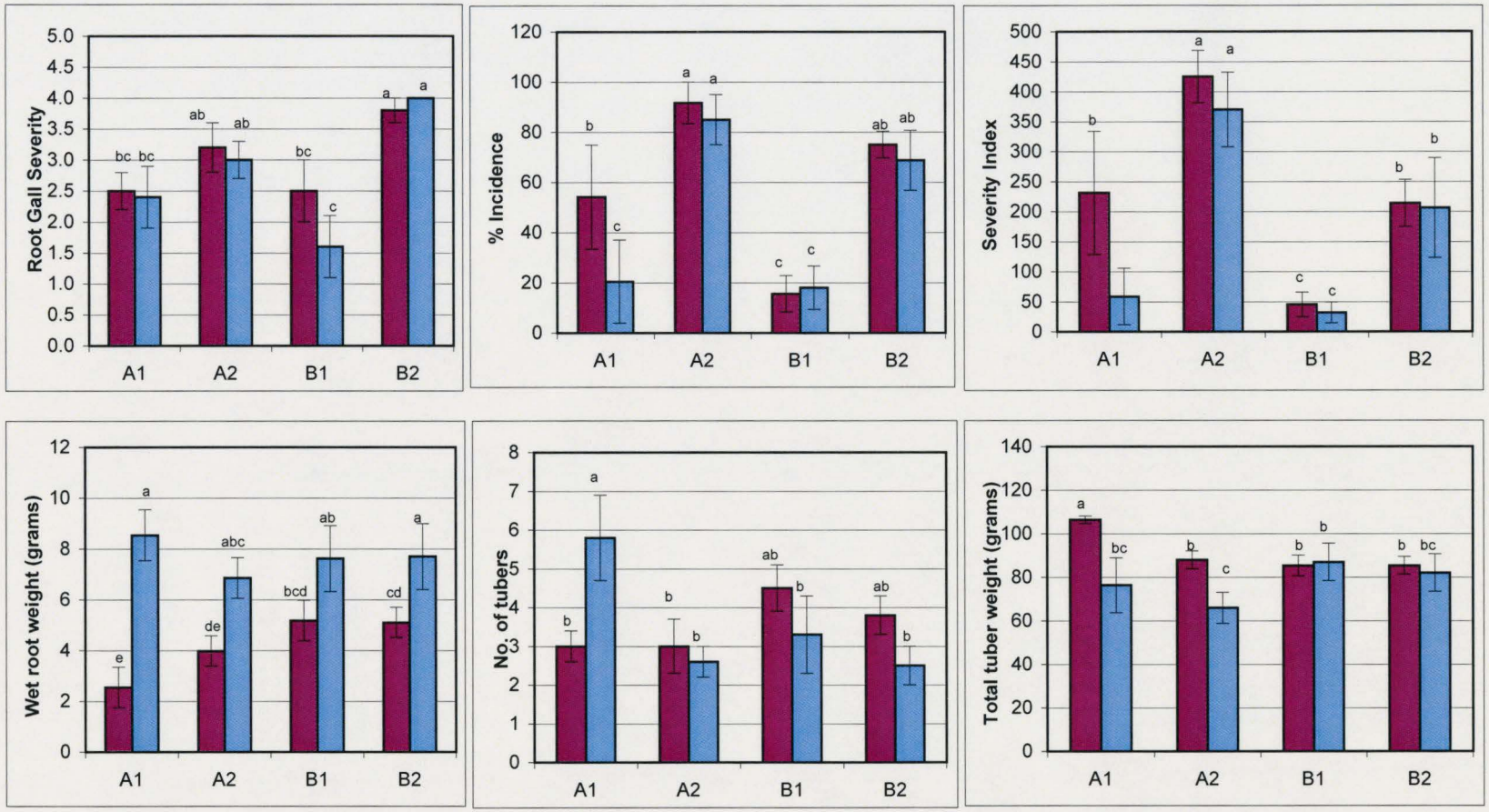


Figure 10. Evaluating powdery scab severity and plant health using two irrigation regimes in a greenhouse for the potato cultivar Rio Colorado (NDC5281-2R). X axis for all charts= Soil Type/Irrigation Regime. Trt. A1 = UC-STD, A2 = I-STD, B1 = UC-ALT, B2 = I-ALT. = crop #1 = crop #2

Soil Type: UC=soil is not inoculated, I=soil is inoculated with 1sporeball/g of soil.

Irrigation Regime: STD = Standard Irrigation regime required for powdery scab development, ALT = 50% less irrigation water during the time of tuber set.

Example of x axis treatment abbreviation: (UC-STD = Untreated control soil, standard irrigation)

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

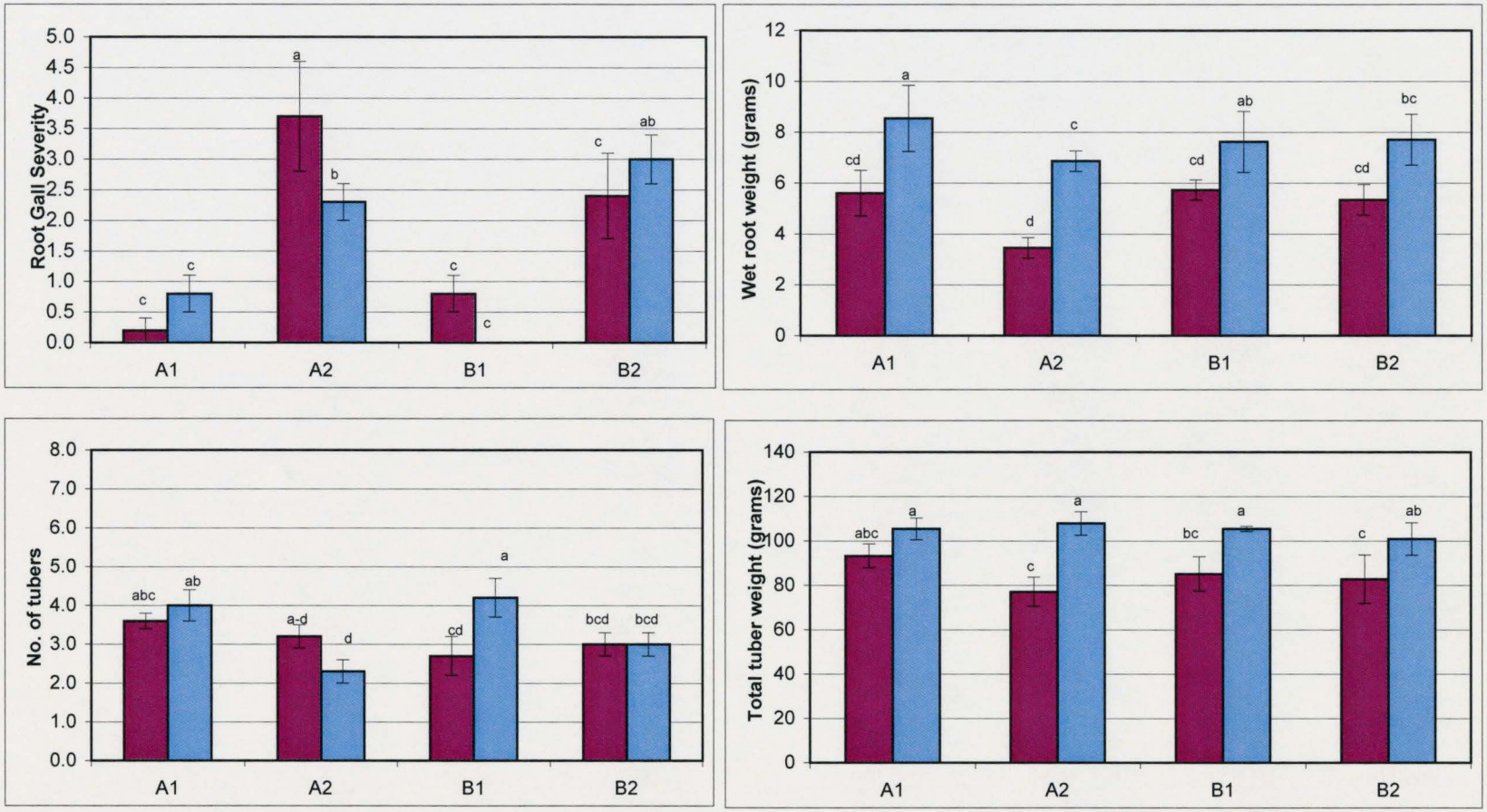


Figure 11. Evaluating powdery scab severity and plant health using two irrigation regimes in a greenhouse for the potato cultivar Rio Grande Russet. X axis for all charts= Soil Type/Irrigation Regime. Trt. A1 = UC-STD, A2 = I-STD, B1 = UC-ALT, B2 = I-ALT. = crop #1 = crop #2

Soil Type: UC=soil is not inoculated, I=soil is inoculated with 1sporeball/g of soil.

Irrigation Regime: STD = Standard Irrigation regime required for powdery scab development, ALT = 50% less irrigation water during the time of tuber set.

Example of x axis treatment abbreviation: (UC-STD = Untreated control soil, standard irrigation)

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

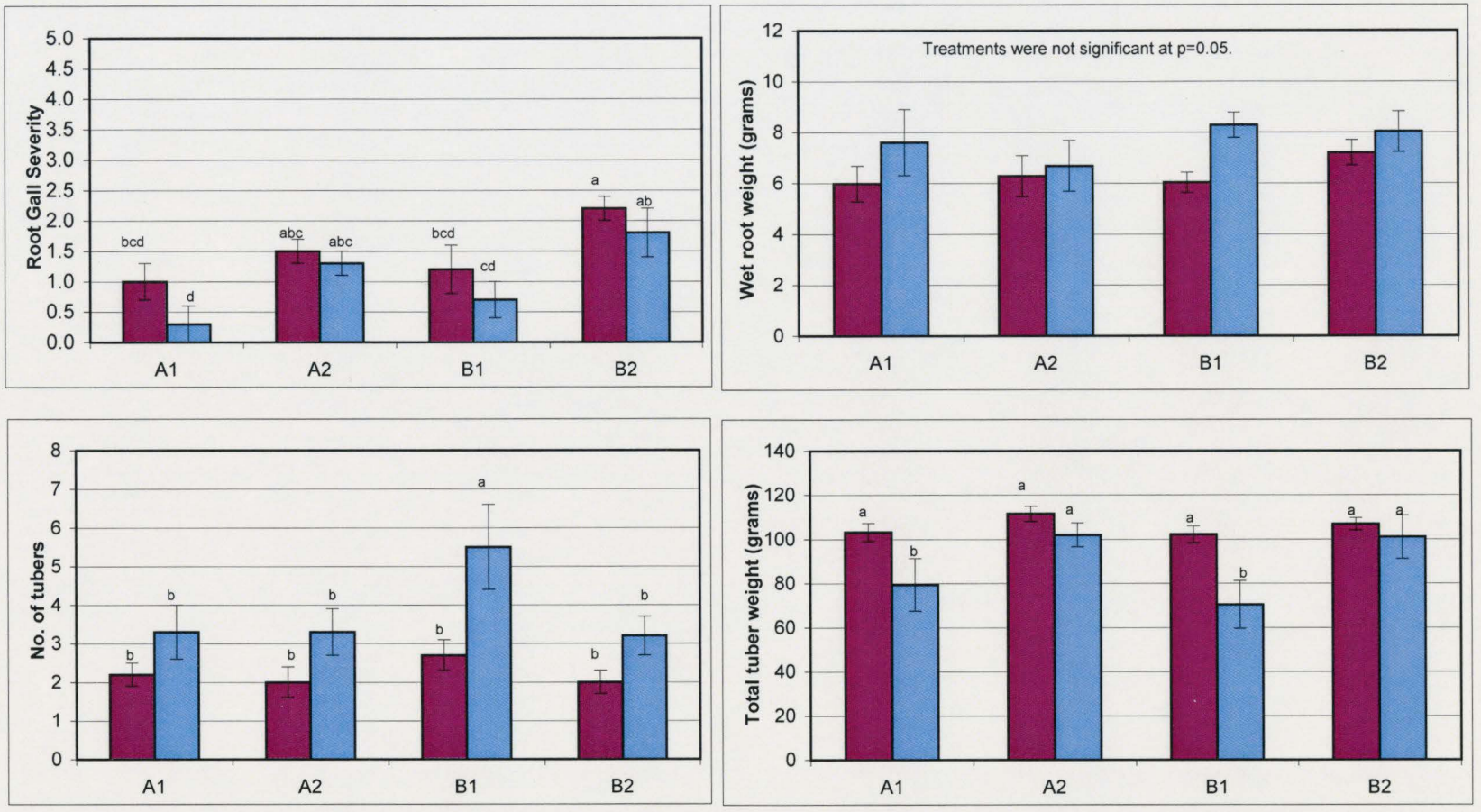


Figure 12. Evaluating powdery scab severity and plant health using two irrigation regimes in a greenhouse for the potato cultivar Russet Burbank. X axis for all charts= Soil Type/Irrigation Regime. Trt. A1 = UC-STD, A2 = I-STD, B1 = UC-ALT, B2 = I-ALT. = crop #1 = crop #2

Soil Type: UC=soil is not inoculated, I=soil is inoculated with 1sporeball/g of soil.

Irrigation Regime: STD = Standard Irrigation regime required for powdery scab development, ALT = 50% less irrigation water during the time of tuber set.

Example of x axis treatment abbreviation: (UC-STD = Untreated control soil, standard irrigation)

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

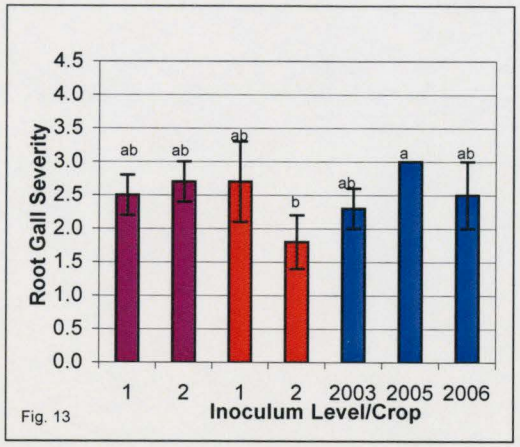
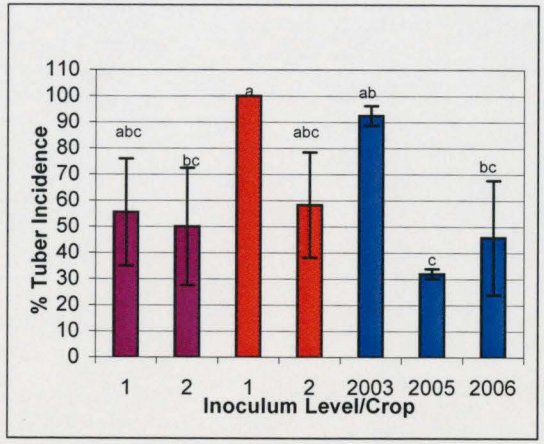
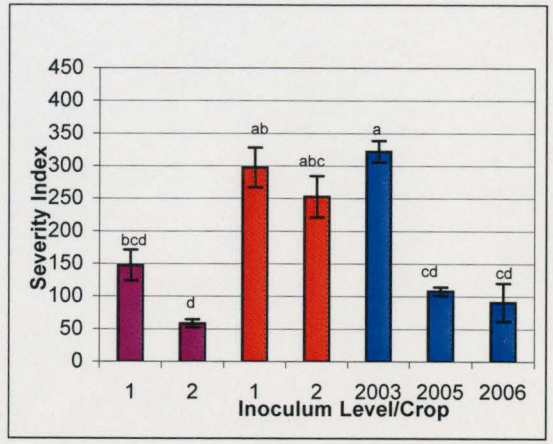


Fig. 13





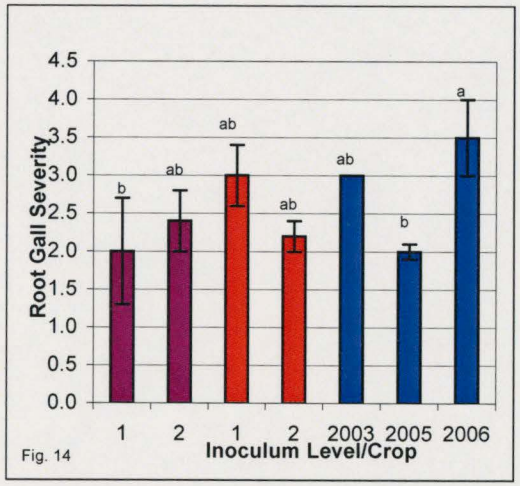
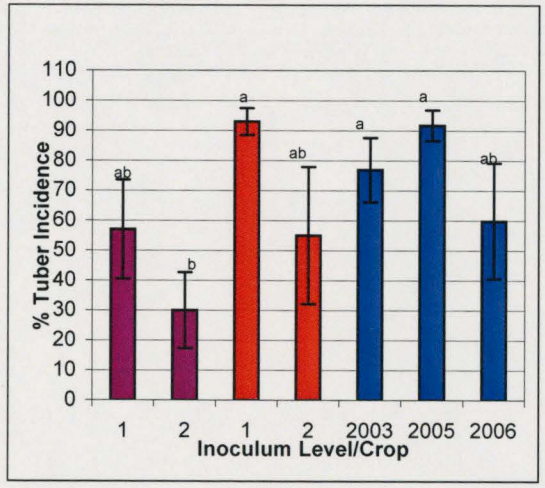
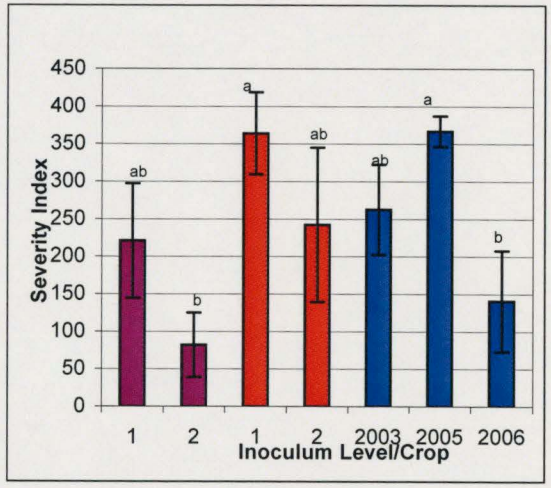


Fig. 14



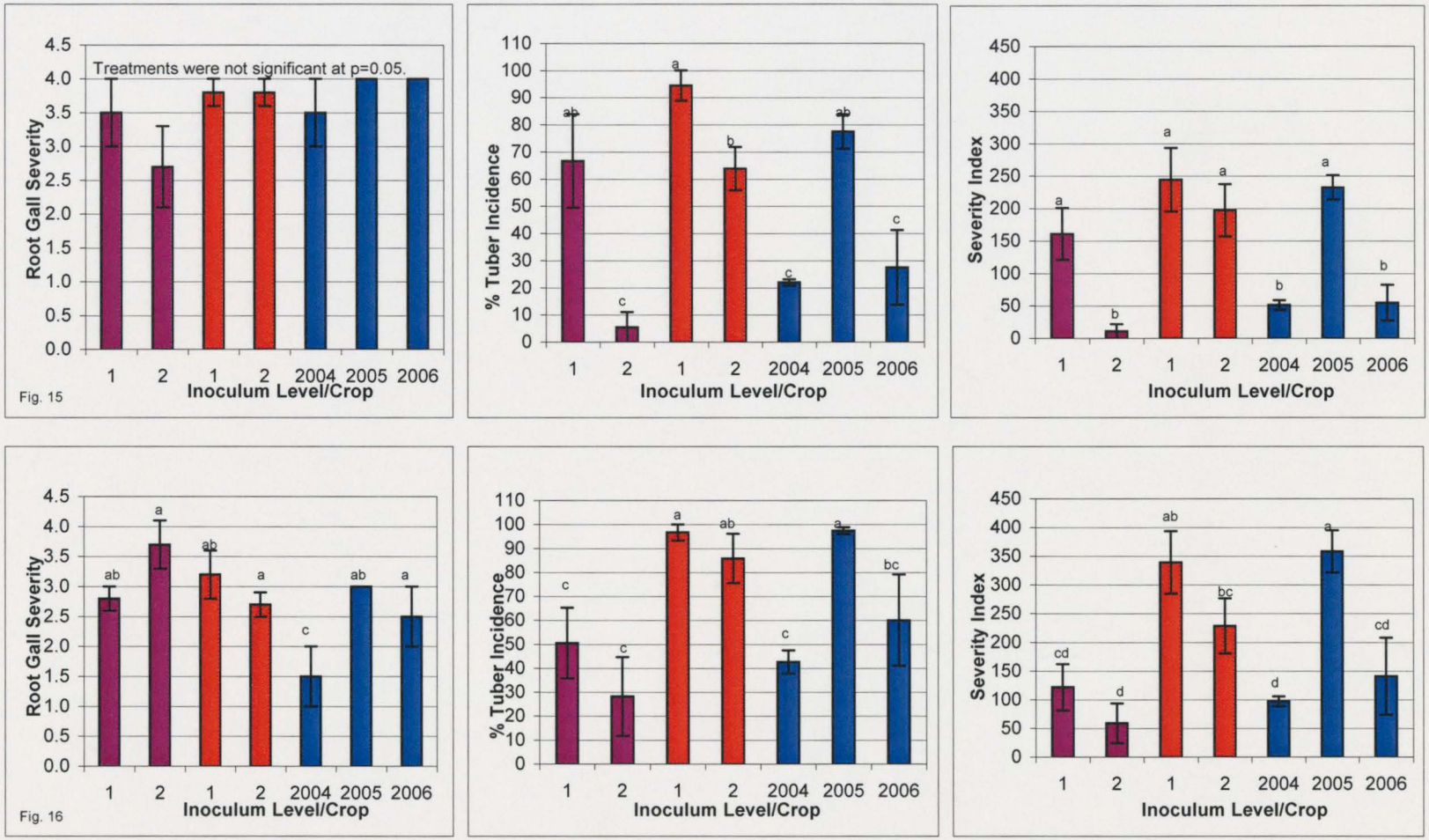


Figures 13 & 14. Comparison of three years of field data to two greenhouse crops evaluating powdery scab severity.

x-axis: 1 = greenhouse crop #1; 2 = greenhouse crop #2; 2003, 2005, 2006 = years of field data. Potato cultivar: Fig. 13 = DT6063-1R, Fig. 14 = Rio Colorado.

= uninoculated greenhouse soil, = greenhouse soil inoculated with 1 sporeball/gram of soil, = Field data from years 2003, 2005, & 2006.

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

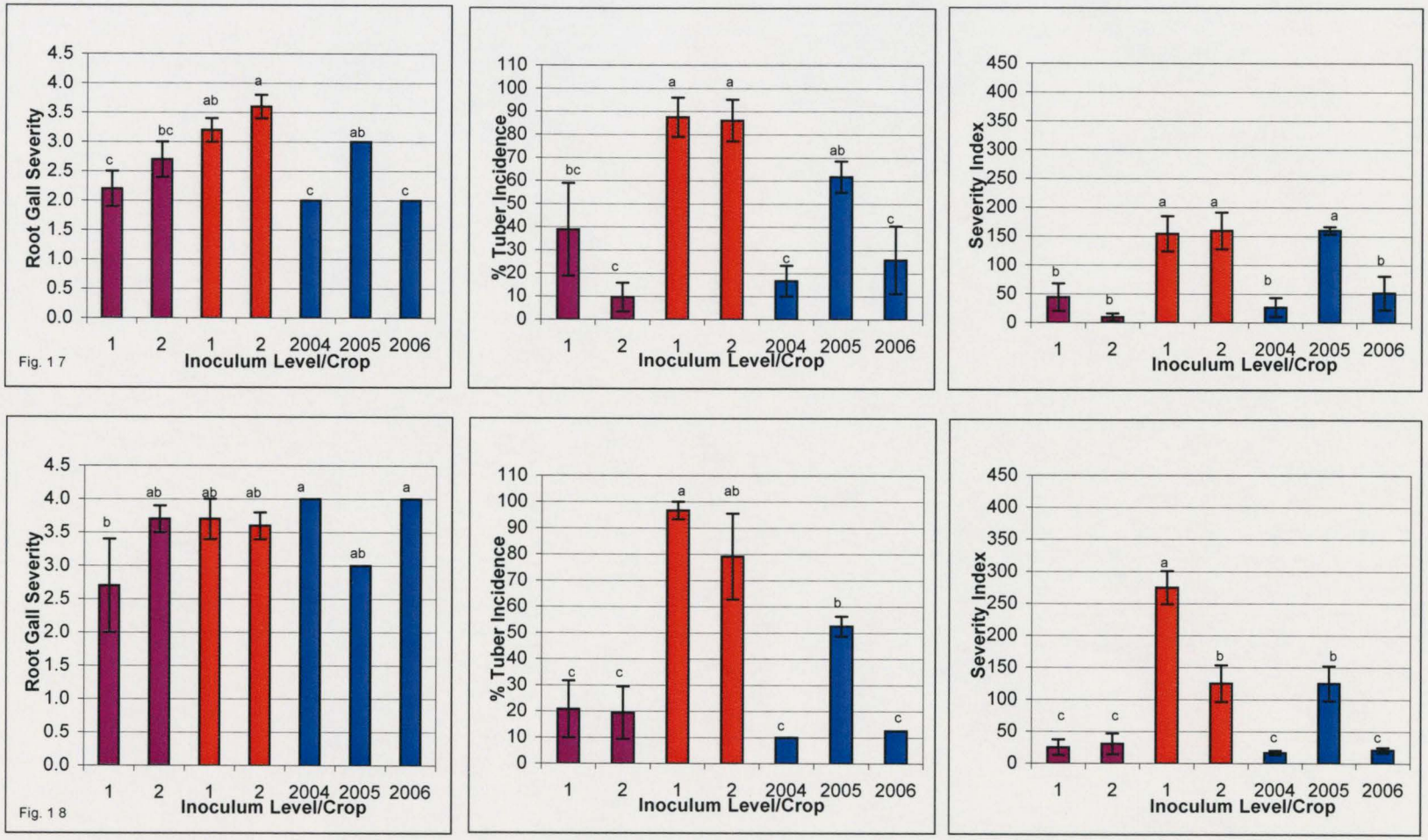


Figures 15 & 16. Comparison of three years of field data to two greenhouse crops evaluating powdery scab severity.

x-axis: 1 = greenhouse crop #1; 2 = greenhouse crop #2; 2004, 2005, 2006 = years of field data. Potato cultivar: Fig. 15 = VC0967-2R/Y, Fig. 16 = Mountain Rose.

= uninoculated greenhouse soil, = greenhouse soil inoculated with 1 sporeball/gram of soil, = Field data from years 2004, 2005, & 2006.

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.



Figures 17 & 18. Comparison of three years of field data to two greenhouse crops evaluating powdery scab severity.

x-axis: 1 = greenhouse crop #1; 2 = greenhouse crop #2; 2004, 2005, 2006 = years of field data. Potato cultivar: Fig. 17 = Purple Majesty, Fig. 18 = VC1002-3W/Y.

= uninoculated greenhouse soil, = greenhouse soil inoculated with 1 sporeball/gram of soil, = Field data from years 2004, 2005, & 2006.

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

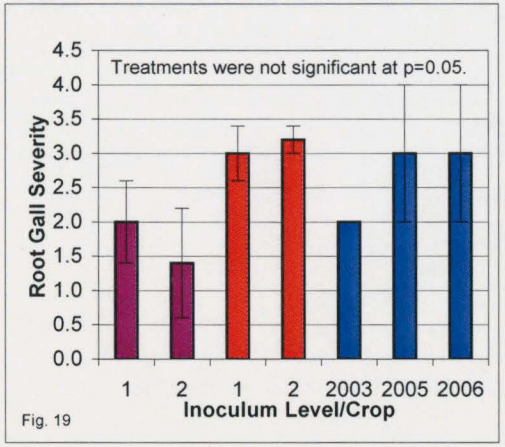
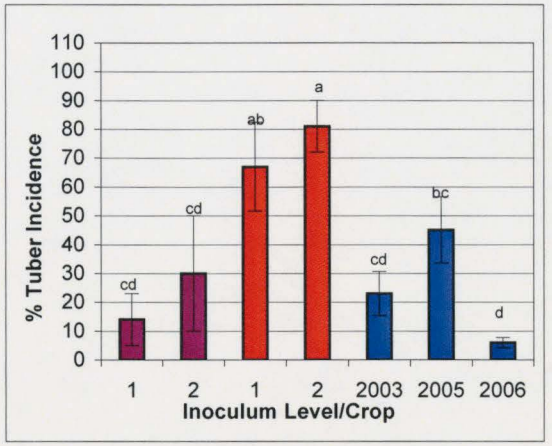
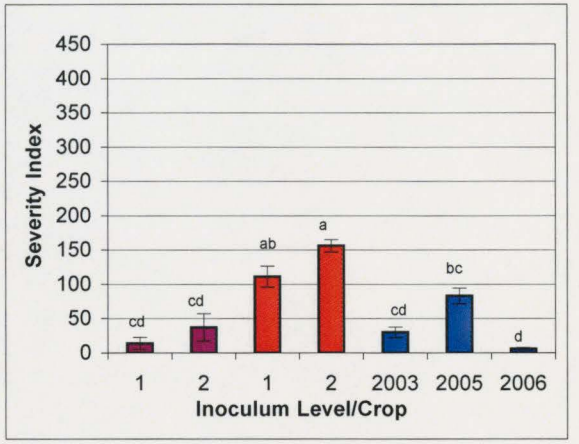


Fig. 19





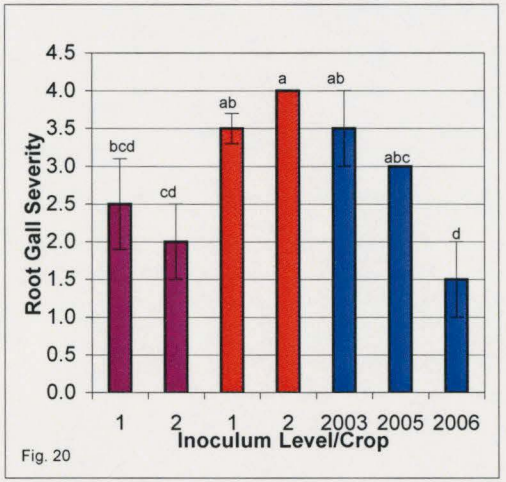
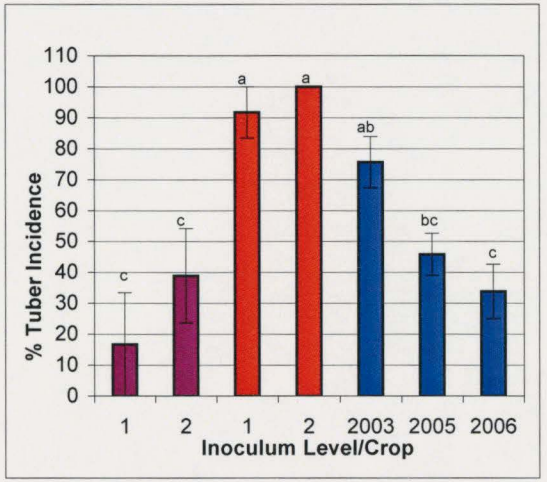
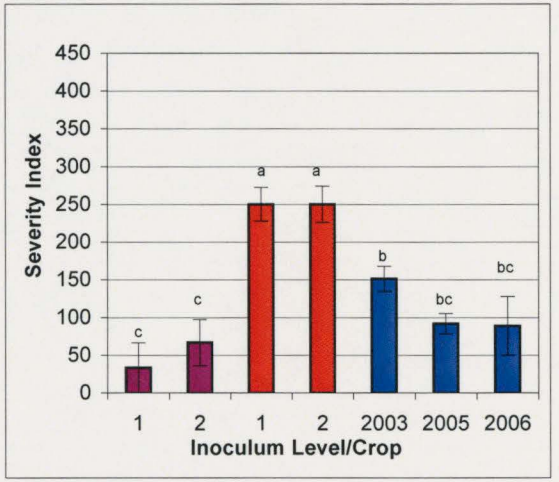


Fig. 20





Figures 19 & 20. Comparison of three years of field data to two greenhouse crops evaluating powdery scab severity.

x-axis: 1 = greenhouse crop #1; 2 = greenhouse crop #2; 2003, 2005, 2006 = years of field data. Potato cultivar: Fig. 19 = Atlantic, Fig. 20 = Superior.

= uninoculated greenhouse soil, = greenhouse soil inoculated with 1 sporeball/gram of soil, = Field data from years 2003, 2005, & 2006.

Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

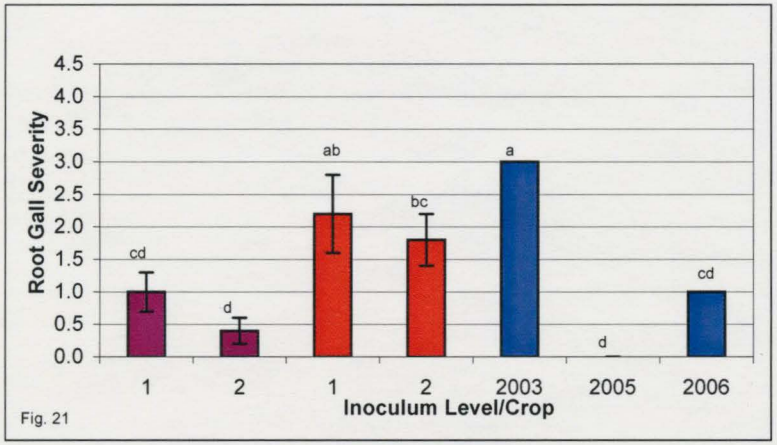
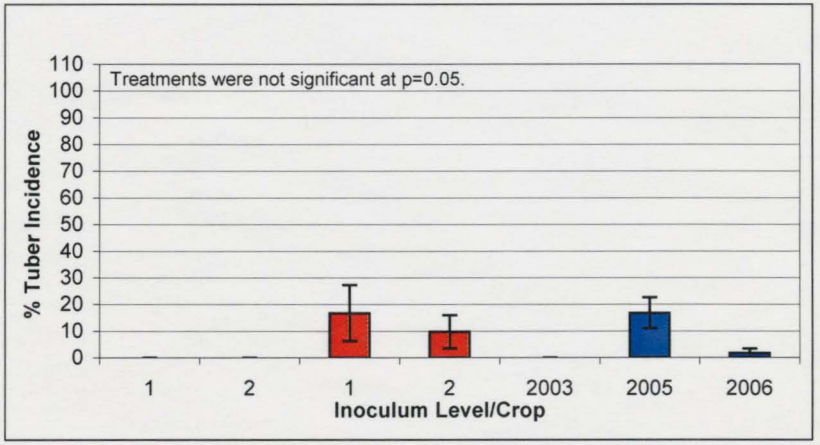


Fig. 21



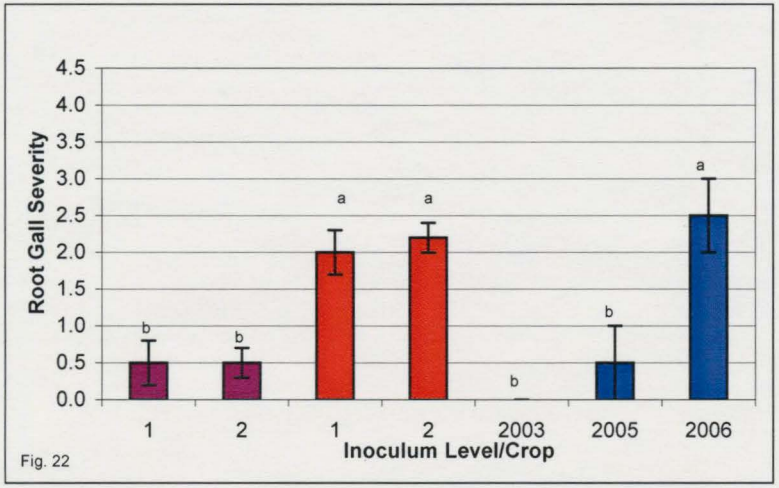
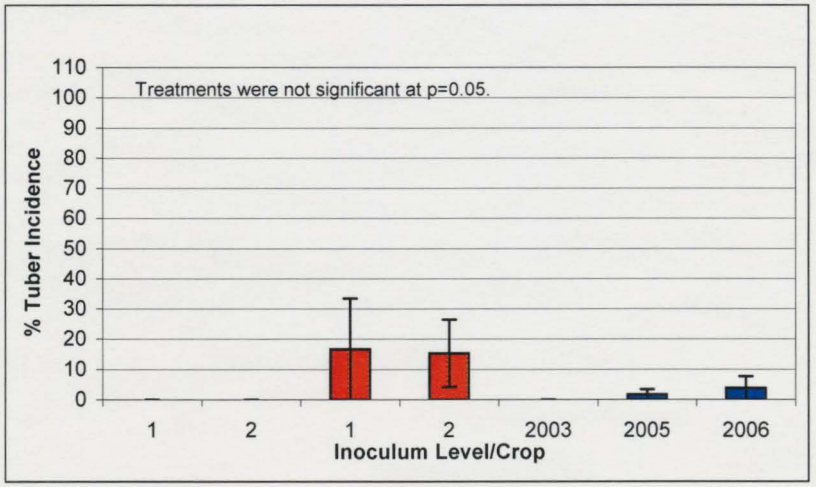


Fig. 22



Figures 21 & 22. Comparison of three years of field data to two greenhouse crops evaluating powdery scab severity.

x-axis: 1 = greenhouse crop #1; 2 = greenhouse crop #2; 2003, 2005, 2006 = years of field data. Potato cultivar: Fig. 21 = Ranger Russet, Fig. 22 = Freedom Russet. = uninoculated greenhouse soil, = greenhouse soil inoculated with 1 sporeball/gram of soil, = Field data from years 2003, 2005, & 2006. Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

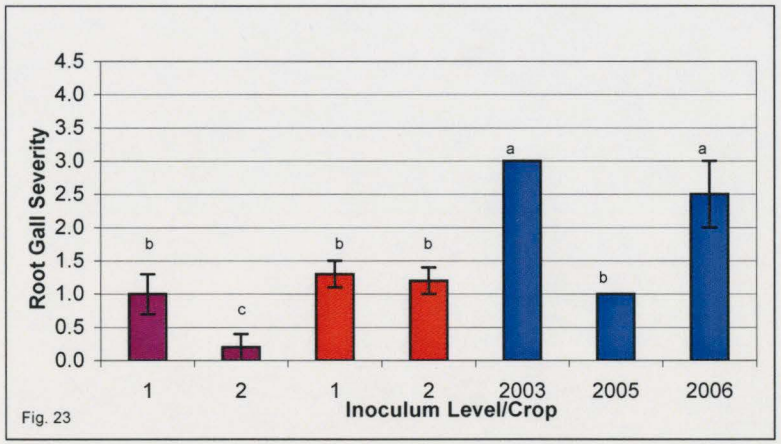


Fig. 23

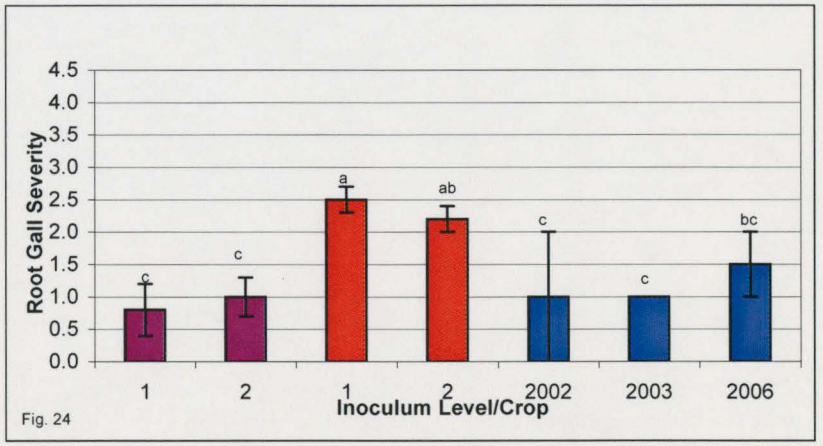


Fig. 24

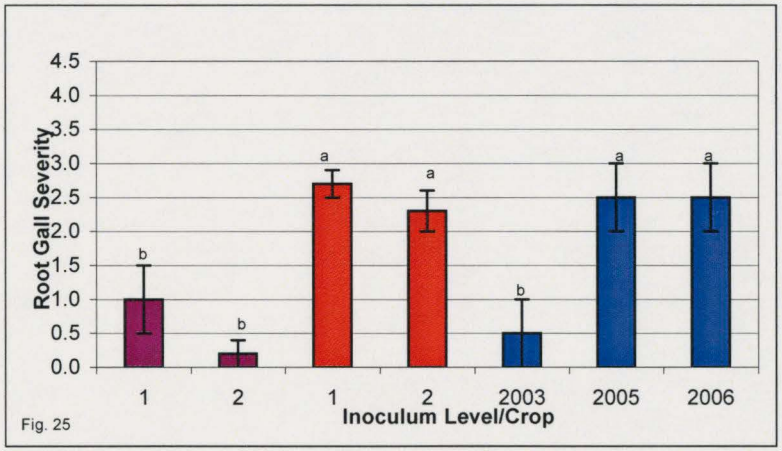


Fig. 25

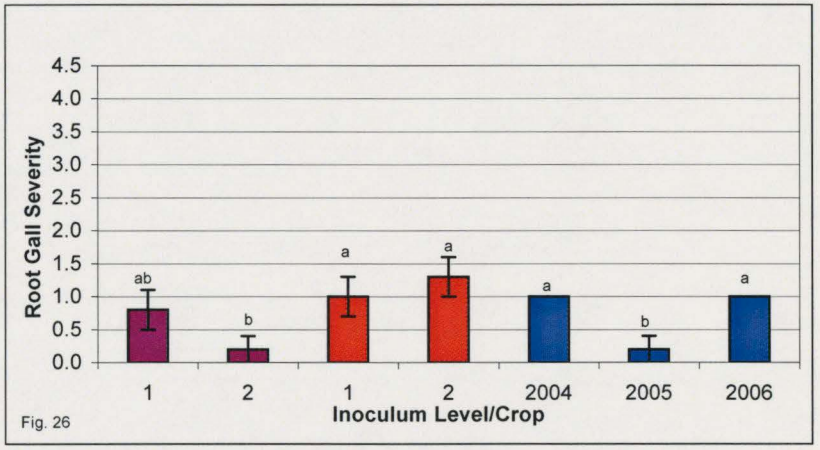


Fig. 26

Figures 23-26. Comparison of three years of field data to two greenhouse crops evaluating powdery scab severity.

x-axis: 1 = greenhouse crop #1; 2 = greenhouse crop #2; 2002, 2003, 2004, 2005, 2006 = years of field data. Potato cultivar: Fig. 23 = Russet Burbank, Fig. 24 = Rio Grande Russet, Fig. 25 = Canela Russet, Fig. 26 = CO94035-15RU.

= uninoculated greenhouse soil, = greenhouse soil inoculated with 1 sporeball/gram of soil, = Field data from years 2002, 2003, 2004, 2005, or 2006. Data analyzed using a LSD separation, bars with the same letters are not significant at P=0.05. The data are expressed as means +/- standard error.

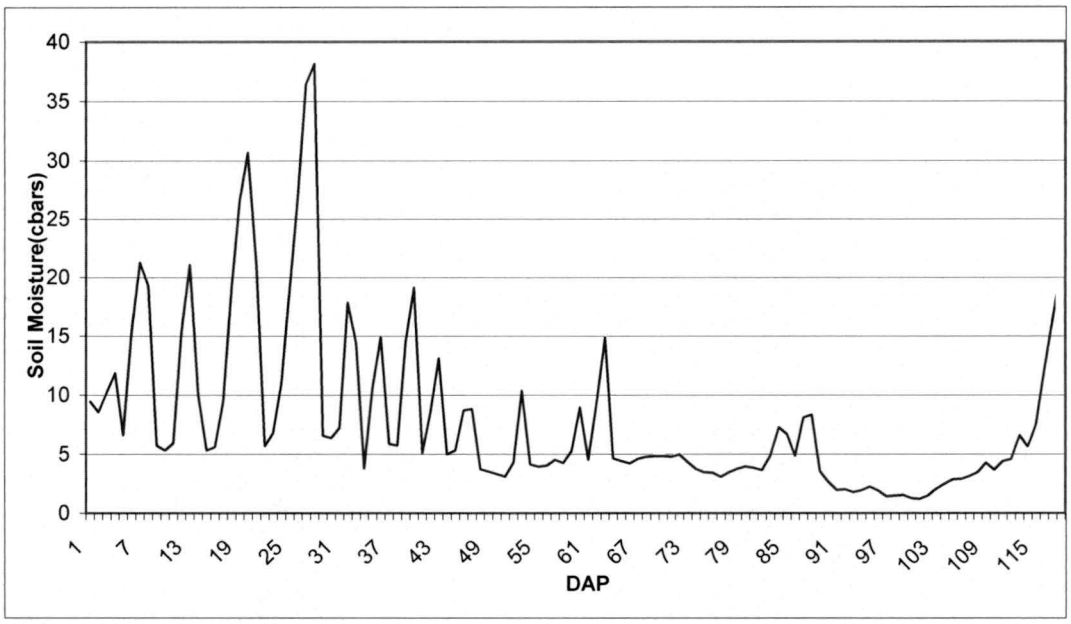


Figure 27. Average Soil Moisture for Trial #1 (soil type x inoculum level x cultivar).
Soil moisture was measured in cbars. In the greenhouse, one weather station which recorded soil temperature was set up monitoring two reps due to possible environmental variation between reps. Weather stations were set up at planting time and removed at harvest time.

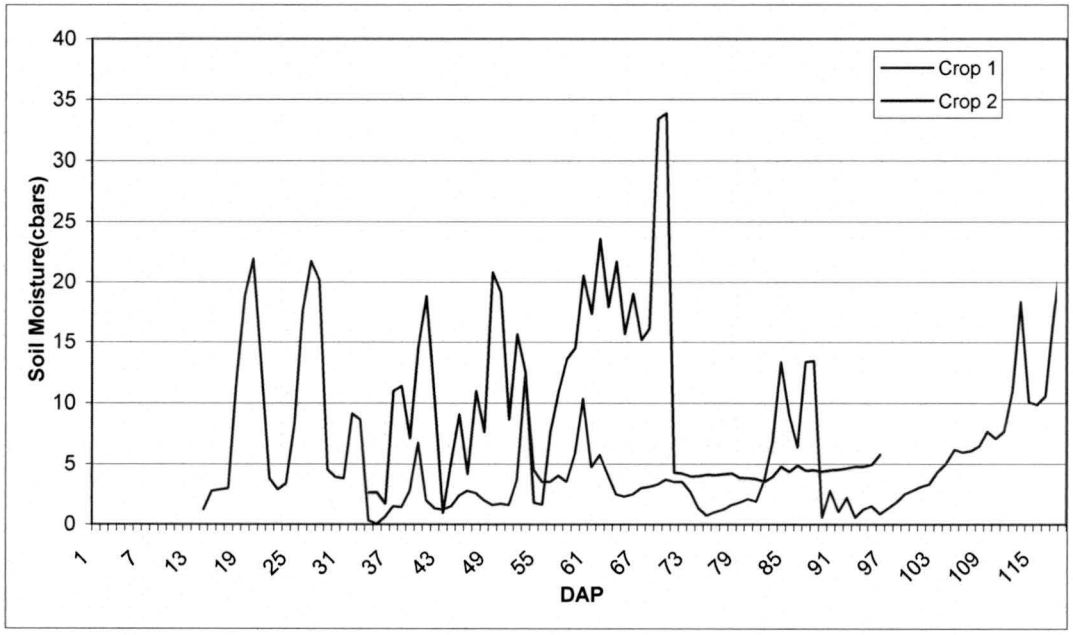


Figure 28. Average Soil Moisture for Trial #2 (inoculum source x soil type).
Soil moisture was measured in cbars. Two crops are represented, which were irrigated differently. Weather stations were set up at planting time and removed at harvest time.

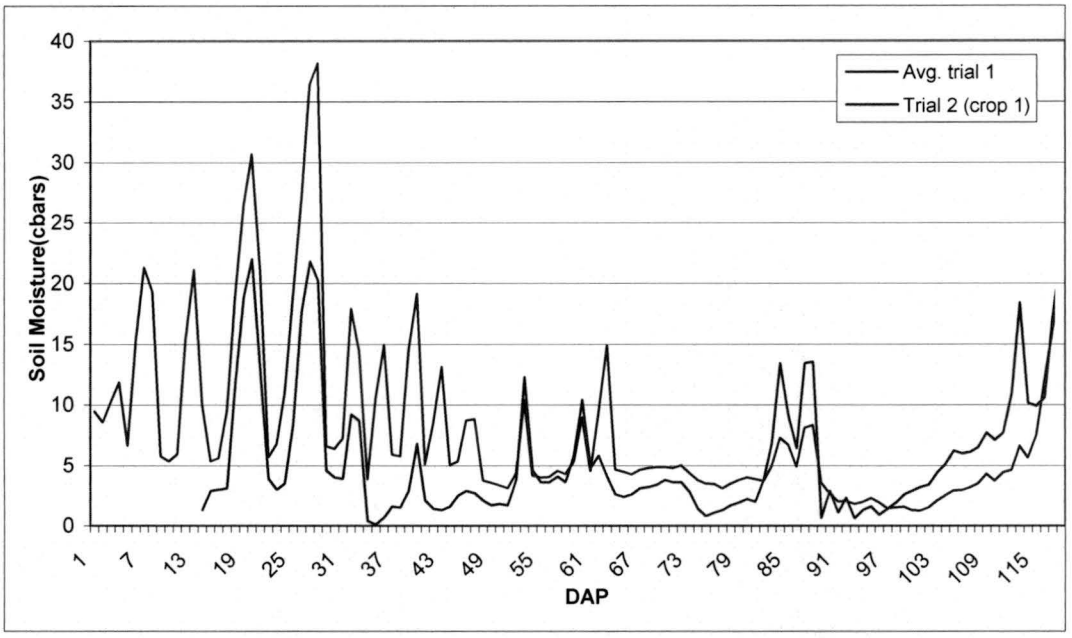


Figure 29. Average Soil Moisture for Trial 1 (soil type x inoculum level x cultivar) vs. trial 2 crop 1 (inoculum source x soil type).

Soil moisture was measured in cbars. Trial 2 (crop 1) was grown in a cooler area of the greenhouse. Two crops are represented, which were irrigated differently. Weather stations were set up at planting time and removed at harvest time.

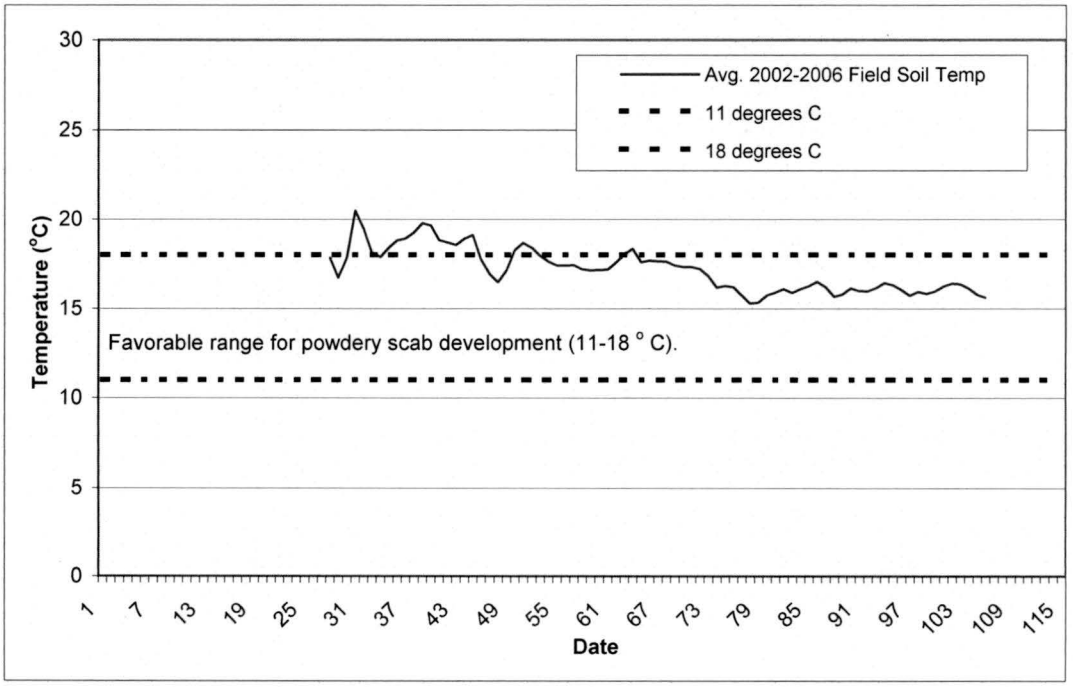


Figure 30. Average Field Soil Temperature in the San Luis Valley for 2002-2006.

Potato crops in the San Luis Valley are typically planted during the first couple of weeks in May and are harvested early to mid September. Soil temperature data was only collected between the first week of June and the end of August due to the timing to powdery scab symptom development.

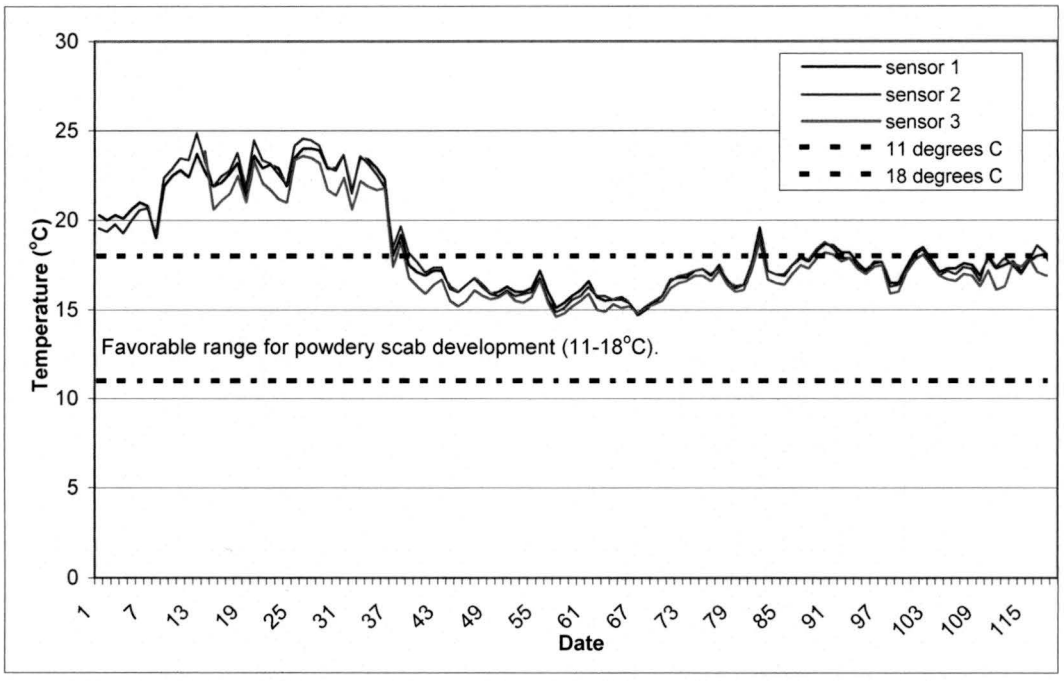


Figure 31. Average Soil Temperature for Trial #1 (soil type x inoculum level x cultivar).
In the greenhouse, one weather station which recorded soil temperature was set up monitoring two reps due to possible temperature variation between reps. Weather stations were set up at planting time and removed at harvest time.

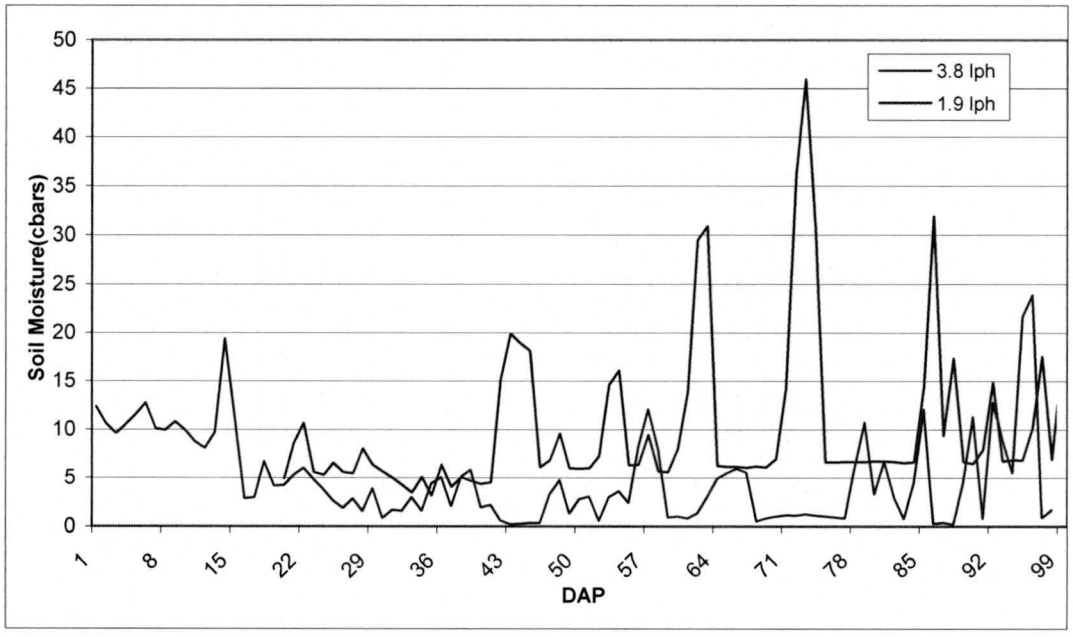


Figure 32. Average Soil Moisture for Trial 3 (soil moisture x inoculum level x cultivar) for high soil moisture (3.8 lph) and low soil moisture (1.9 lph) during tuber set.
Soil moisture was measured in cbars. The soil moisture data represented is from crop 1.

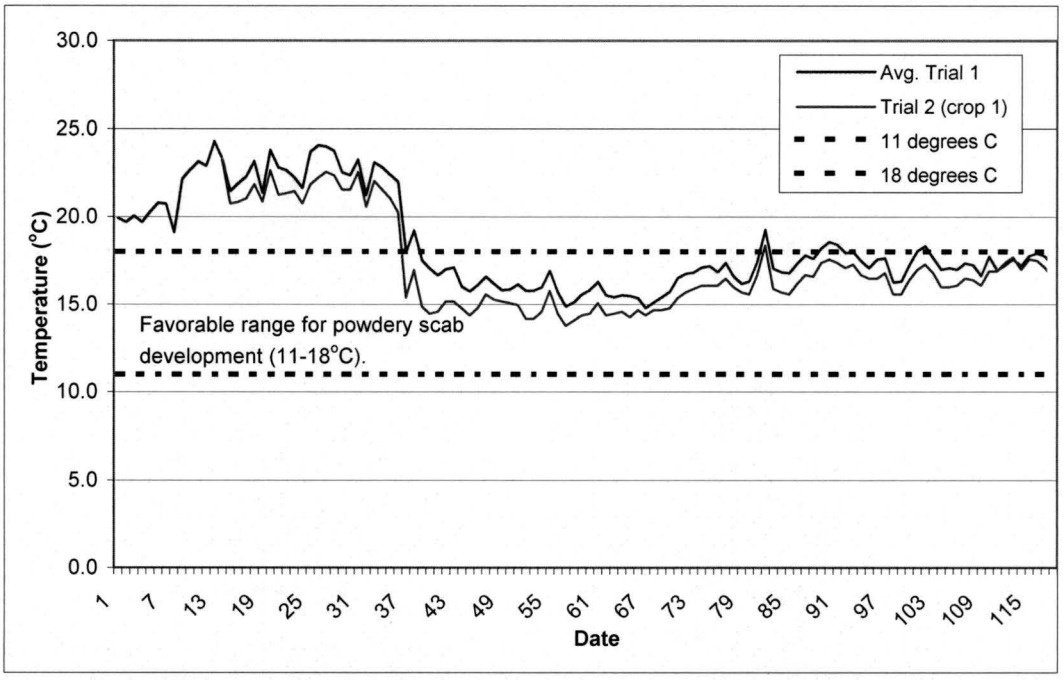


Figure 33. Average soil temperature for trial 1 (soil type x inoculum level x cultivar) vs. soil temperature for trial 2 (inoculum source x soil type).

For trial 1, the average of the soil temperature collected from three weather stations is represented in the above figure. Only one weather station was used to record soil temperature data in trial 2. Weather stations were set up at planting time and removed at harvest time.