# Canadian Agri-Science Cluster for Horticulture 3

# Update to Industry

## 2019-2020

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| **Activity title:** Common Scab: Increasing profitability of Canadian potato producers by controlling common scab |
| **Name of Lead Researcher:** Claudia Goyer, Agriculture and Agri-Food Canada (AAFC), Fredericton, NB |
| **Names of Collaborators and Institutions:** Rick Peters (AAFC Charlottetown), Louis-Pierre-Comeau (AAFC, Fredericton), Martin Filion (université de Moncton), Newton Yorinori (Cavendish Farms), Tracy Shinners-Carnelley (Peak of the Market), David MacMullin (Carleton University), Mario Tenuta (University of Manitoba), Khalil Al-Mughrabi (New Brunswick Department of Agriculture, Aquaculture and Fisheries. |
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| **Activity Objectives (as per approved work plan):**<br>The overall objective of the project is to evaluate several methods to control common scab of potato under a range of environmental conditions and soil types across Canada.<br><br>The specific objectives of this project are to:<br>• Sub-activity 1.1 Characterize the genetic diversity of *Streptomyces* spp. causing common scab and develop tools to measure specific genotype,<br>• Sub-activity 1.2. Evaluate methods to control common scab using small plot and field-scale trials in commercial potato fields,<br>• Sub-activity 1.3. Determine the effect of common scab control methods on soil health and quality parameters,<br>• Sub-Activity 1.4. Determine the concentrations of soil isothiocyanates,<br>• Sub-Activity 1.5. Evaluate the effect of common scab control methods on microbial communities. |
| **Research Progress to Date (use plain language):**<br>Common scab results in significant economic losses every year in Canada. Common scab symptoms are characterized by brownish lesions on potato tubers that can result in declassification of tubers in seed production, rejection for the table market, and difficulty in peeling the tubers leading to significant losses in the processing industry. There is currently no chemical registered specifically to control common scab in Canada. The overall objective of the project is to evaluate several methods to control common scab of potato under the different environmental and soil properties of Prince Edward Island (PEI), and Manitoba (MB).<br><br>**Sub-activity 1.1 Characterize the genetic diversity of *Streptomyces* spp. causing common scab and develop tools to measure specific genotype.** In previous years, a collection of 221 pathogenic *Streptomyces* spp. causing common scab were obtained from tubers collected in PEI, NB, and MB. The pathogenicity of the isolates was confirmed by verifying the presence of the *txtA* gene involved in the production of the plant toxins thaxtomins. The pathogenic *Streptomyces* spp. are currently being classified using a technique of DNA profiling named rep-PCR to determine the number of genetically different isolates present in PEI, NB, and MB. So far, at least 24 different genetic groups were identified. |

Quantitative PCR (qPCR) assays specifically targeting some of the most abundant genetic groups identified so far are being developed as field detection/quantification tools. This work is presently ongoing and will lead to the characterization of the diversity of pathogenic *Streptomyces* spp. strains found in NB, PEI, and MB, as well as the determination of their distribution among these provinces.

**Sub-activity 1.2. Evaluate methods to control common scab using small plot and field-scale trials in commercial potato fields.**

**1.2.1 Effect of diverse control approaches on reducing common scab of potato (Tracy Shinners-Carnelley, Peak of the Market, Newton Yorinori, Cavendish Farms, vote 10)**.

**Studies in PEI** (**Newton Yorinori, Cavendish Farms**)

**Study 1:** Several potato varieties are resistant to common scab, however several of the varieties used by the industry are susceptible to common scab but perform very well under growth conditions in PEI and are well adapted to crop systems and processes used by the growers and the industry. Somaclonal variation (SV), which uses the natural ability of the plant to change under stress conditions including tissue culture, can be used to increase resistance to disease in potato. Using SV, the agronomical traits of an existing variety can be kept while increasing disease resistance. Common scab resistance of somaclonal variants of two varieties including Riverdale Russet and Prospect were evaluated in fields infested with common scab in PEI. Around 100 variants of each variety were obtained and screened for common scab resistance in the field since 2017, with the top 25 being selected for each variety. In Summer 2018, the 25 most resistant variants of Riverdale Russet and Prospect were evaluated in three sites across PEI with two replications. The three most promising variants of Riverdale Russet, and four variants of Prospect were selected for further evaluation based on the assessment of disease severity. In summer 2019, three trials were planted at three different sites in PEI. Each trial had three replications. Four variants of Prospect, four variants of Shepody, and three variants of Riverdale russet were used plus the respective original (parental) lines and Russet Burbank as a resistant variety for reference. Trials were planted in June and harvested in October. The scab incidence and severity were evaluated to determine if there was a significant improvement of common scab resistance of the variants compared with the original line. The variants had a slight decrease in incidence and disease severity compared to the original lines however, the difference was not sufficient enough to warrant changing the existing variety to the variant. Other approaches will need to be evaluated to reduce losses due to common scab including agricultural practices and control products.

**Study 2:** The effect of ammonium lignosulfonate (ALS) products, a wood bark extract, and sulfur-based fertilizers on common scab incidence and severity were also assessed at the Cavendish Research site. A previous study done in laboratory conditions showed some potential to use ALS to control common scab. A field trial with the application of liquid and powder formulation of ALS was established using the common scab susceptible variety Prospect. Forty-five days after planting, liquid, and powder ALS products were applied as a side- dressing i.e. at the base of the plants then covered by soil (final hilling) on the same day. Results showed no difference in common scab incidence or severity between the treated and untreated plots. Another field trial evaluated the potential of wood bark extract to control common scab. Wood bark extract was applied as a side-dressing at the base of Prospect plants. Results indicated no difference in common scab incidence and severity between treated and untreated plots. The failure to control common scab control by ALS products and the wood bark extract might be due to the dry weather during July and August (the trials were not irrigated), which probably did not allow the product to be re-distributed in the soil to reach the tuber zone.

Another field experiment was established to verify the efficacy of sulfur-based fertilizers to control common scab. Elemental sulfur and ammonium sulfate were applied in two susceptible varieties to common scab (Shepody and Prospect) at two different timings: in-furrow at planting and as a side dressing at hilling and covered with soil on the same day. None of the treatments resulted in effective control of common scab when compared with control using standard fertilizer. In addition to scab severity, the total and marketable yields were analyzed in this trial. The results indicated that ammonium sulfate applied as side-dressing has the potential to increase yields, however, ammonium sulfate applied in-furrow at the rate of 420 lbs/ac caused phytotoxicity in the plants, reducing the emergence rate of the plants.

**Studies in MB** (**Tracy Shinners-Carnelley, Peak of the Market):**

In Manitoba, a nine treatment replicated small plot field trial was established at the Peak of the Market Research Site in Winkler. AC Peregrine Red was planted for this trial and the treatments evaluated included 1) Serenade

Soil in-furrow, 2) oat as a nurse crop, 3) barley as a nurse crop, 4) oat nurse crop plus Serenade Soil in-furrow, 5) barley nurse crop plus Serenade Soil in-furrow, 6) composted beef manure, 7) composted beef manure plus Serenade Soil in-furrow , 8) mustard meal, 9) control (untreated). The dry spring conditions made it difficult to establish the nurse crops. Delayed and uneven germination of both the oats and barley resulted in a poor herbicide burn-off, creating emergence issues for the potatoes in the nurse crop treatments.
At harvest, there were no significant differences amongst the treatments with respect to any of the data collected from this trial including yield, tuber size profile; and incidence of common scab and russeting. Overall the scab incidence was low, ranging from 1.5-9.0%. Russetting is commonly found on the skin of smooth-skinned varieties and is sometimes referred to as "russet scab". For this reason, it was included as part of the visual tuber assessments for this trial, with the incidence ranging from 7.5- 26.5% across the treatments. To validate these visual assessments, representative tubers with typical russeting and common scab lesions were sent to Claudia Goyer for *Streptomyces* spp. testing. *Streptomyces* was detected from the "typical" common scab lesions, but not from russet lesions.

**1.2.2 Impact of tillage and rotational crop species on common scab severity (Rick Peters, AAFC, vote 1)**
Two studies to control common scab were performed in PEI. Study 1 evaluated the effect of rotation crop system on common scab severity in commercial potato fields. In 2018, six potato fields were identified that were split in the previous year with a variety of rotational crops including brown mustard, sorghum-sudan grass, red clover, buckwheat, ryegrass, pearl millet or a variety mixture. Soil samples were taken from six replicated strips in each treatment area at tuber initiation and analyzed for soil properties. At harvest, six replicated ten-foot strips were harvested from each treatment area and soil samples were also taken from the harvested zones. Soils were analyzed for various properties and harvested tubers were assessed for incidence and severity of common scab. In summer 2019, four field sites in central PEI were identified with different previous rotational crop splits including:
1. (DM) double mustard vs (SSPM) sorghum/sudan + pearl millet + tillage radish
2. (PM) pearl millet vs (SS) sorghum/sudan
3. (BP) Black peas (no mushroom compost) vs (W) wheat (mushroom compost)
4. (SM) single mustard vs (SSPM) sorghum/sudan + pearl millet
The potato crop was established at each field site. Six plots consisting of 10 plants were randomly assigned in each of the previous rotational crop splits (total of 12 plots from each field). Soil from each plot was sampled at tuber initiation and harvest. Soil cores were collected at 30 cm depth using a Dutch agar and pooled.

Soil samples were analyzed for soil physical and chemical properties. All plots were hand-harvested and the harvested tubers were graded for yield and quality. Currently, disease assessments on a subset of 25 marketable tubers per plot are being conducted. Upon completion of the dataset, all data will be analyzed to assess the effects of prior rotational crops on soil properties, tuber yield, and quality, and disease.

In study 2, the effect of fertilizer-based products on the severity of common scab was evaluated. The treatments included 1)Tropico ($CaNO_3$), 2) ammonium sulfate, 3) elemental sulfur, and 4) conventional ammonium nitrate applied in-furrow to small plots in Harrington, PEI. Soil samples were collected at defined intervals during the growing season and stored at 4 degrees C for subsequent analysis of soil carbon and other chemical/physical properties or stored at -80 degrees C for subsequent DNA extraction and detection/quantification of the common scab pathogen. Plots were harvested and graded for yield and quality. Currently, disease assessments on a subset of 25 marketable tubers per plot are being conducted. Upon completion of the dataset, all data will be analyzed to assess the effects of fertility inputs on soil properties, tuber yield, and quality and disease severity, in addition to the impacts on pathogen population levels in the soil.

**Sub-activity 1.3. Determine the effect of common scab control methods on soil health and quality parameters (Claudia Goyer, and Louis-Pierre Comeau, AAFC, vote 1)**
**Quantification of pathogenic *Streptomyces* from soils**
The abundance of the common scab pathogen at the Peak of the Market research site was measured from soil samples collected in May using quantitative PCR (qPCR). No detection or very low abundance of the common scab pathogen was measured. This result raised some questions as to the efficacy of the quantification tool to quantify common scab pathogen from soils of Manitoba. Soils from the surface of tubers or the hills were

collected in four commercial potato fields and from Peak of the Market research site. The results showed that common scab was detected in all samples except one sample from the Peak of the Market research site. The abundance of the common scab pathogen was the greatest in a field where the most severe common scab was observed showing a logical trend in the data. Overall, the results indicated that the quantification tool was adequate to measure the common scab pathogen in the fields of Manitoba but that the abundance must be high enough to be detectable.

In Manitoba, tubers frequently present symptoms that are not typical of common scab or russeting, complicating the disease evaluation. For this reason, the abundance of pathogenic *Streptomyces* was quantified from tubers with typical symptoms, atypical symptoms, and russeting. Three tubers were taken randomly in each category i.e. suspected scab, common scab, and russeting. Lesions from each of the tubers were taken separately. Three PCR reactions were used for each tuber tested (9 reactions per category). Controls consisted of 1) DNA from the common scab pathogen, 2) a tuber with typical common scab symptoms from NB, and 3) a healthy potato tuber. The positive and negative controls were as expected demonstrating that the PCR reaction was good and that there was no contamination (Table 1). The tubers from MB and NB with typical symptoms of common scab were all positive with low Ct around 16-25 which indicates abundant pathogen in the lesions. The russeting and atypical common scab symptoms were all negative or below detection (Table 1). Negative results are difficult to interpret as it might mean that the abundance of the pathogenic *Streptomyces* spp. was too low to be detected or that the atypical symptoms and russeting are not caused by common scab. Further investigation will be necessary to determine the exact cause of the atypical symptoms and russeting.

**Table 1. Detection of pathogenic *Streptomyces* spp. in tubers with typical, suspected common scab and russeting lesions**

| Sample | Average Ct* of 3 replicates | Presence of common scab |
|---|---|---|
| MB Suspected Scab Tuber #1 | Not detected | undetermined |
| MB Suspected Scab Tuber #2 | Not detected | undetermined |
| MB Suspected Scab Tuber #3 | Not detected | undetermined |
| MB Scab Tuber #1 | 18 | positive |
| MB Scab Tuber #2 | 26 | positive |
| MB Scab Tuber #3 | 19 | positive |
| MB Russetting Tuber 1 | Not detected | undetermined |
| MB Russetting Tuber 2 | Not detected | undetermined |
| MB Russetting Tuber 3 | Not detected | undetermined |
| Tuber with typical scab (NB) | 16 | positive |
| "Clean" tuber i.e no symptoms | 33 | negative |
| Control: std 10-3 txtC gene | 22 | not applicable |
| Control: std 10-4 txtC gene | 25 | not applicable |
| Control: std 10-4 txtC gene | 28 | not applicable |
| Control - water | Not detected | not applicable |
| blank | Not detected | not applicable |

*Cycle threshold: the lower values indicate more abundant pathogenic *Streptomyces* compared to greater value

Soils from trials done to evaluate the effect of fertilizers on the abundance of pathogenic *Streptomyces* spp using qPCR were received in late March and will be processed in 2020-2021. Soil physico-chemical properties of fields in PEI will be analyzed to evaluate the effect of common scab control methods on soil quality. Soil samples were received late in the fall of 2019 and are currently being processed and analyzed.

**Sub-Activity 1.4. Determining the concentrations of soil isothiocyanates (David McMullin, University of Carleton, vote 10).**

Isothiocyanates (ITCs) are volatile compounds produced through the degradation of mustard meal and mustard residues in soils. ITCs are known to reduce the populations of soil-borne-disease. It is useful to evaluate the amount of ITCs produced in soils as it varies with plant material, growth stage, crop management, and environmental conditions which might affect the efficacy of control of common scab. Methods to quantify ITCs

were identified and optimized. ITCs can be quantified using quantitative gas chromatography-mass spectrometry (GC-MS). GC-MS methods were developed for four of the major soil ITCs (2-propenyl-ITC, methyl-ITC, 2-phenethyl-ITC, and benzyl-ITC) using analytical standards. The limit of detection of each method was determined and was shown to be in the low nanogram range. The lower and upper limits of quantification of the ITCs were evaluated and spanned several orders of magnitude. An Honours food science student was successfully hired to perform this research.

**Sub-Activity 1.5. Evaluate the effect of common scab control methods on microbial communities.**
Possible relationships between the diversity and richness of microbial communities and the abundance of the pathogenic *Streptomyces* spp. causing common scab or disease severity will be evaluated on trials where common scab was successfully controlled.

**Extension Activities (presentations to growers, articles, poster presentations, etc.):**

- Shinners-Carnelley, Tracy. Presented field trials and preliminary results to control common scab to growers. Peak of the Market Field Day, Winkler, August 15th, 2019.
- Yorinori, Newton. 2019. Presented field trials and preliminary results to control common scab to growers. Cavendish Field Day, Annan, PEI. September 27th.
- Goyer, Claudia. 2019. Visited growers and Peak of the Market research field site and facility to discuss issues related to common scab of potato. Manitoba, September 9-12th

**Early Outcomes (if any) or Challenges:**

- A collection of 221 pathogenic *Streptomyces* spp. causing common scab was obtained from tubers collected in PEI, NB, and MB. The pathogenic *Streptomyces* spp. are currently being classified using a DNA profiling technique named rep-PCR and so far, at least 24 different genetic groups were identified.
- Variants of Riverdale Russet and Prospect that were obtained using somaclonal variation, a technique that can increase resistance to common scab, showed a modest reduction in the severity of the disease compared to the parental line. The reduction in disease severity might not be sufficient to warrant changing the existing varieties.
- Several approaches to control common scab were evaluated in Manitoba and Prince-Edward-Island including nurse crops, biopesticide, compost, mustard meal, wood bark extract, ammonium lignosulfonate products and sulfur-based fertilizers however none of these approaches resulted in a decrease in common scab severity. Overall disease pressure at the MB site was low, and therefore did not provide a good assessment of the treatments evaluated.
- Tubers from Manitoba with typical common scab symptoms, atypical symptoms, and russeting were tested for the presence of pathogenic *Streptomyces* spp. Only tubers with typical symptoms tested positive for the presence of pathogenic *Streptomyces* spp. The result will improve the evaluation of common scab in Manitoba.
- A method to quantify isothiocyanates, the volatile compounds produced via the degradation of mustard meal and mustard residues, was developed and optimized.

**Key Message(s):**
This project will allow a better understanding of the diversity of the common scab pathogen in NB, PEI, and MB to improve the detection of specific species or strains of the common scab pathogen. The tools to evaluate the effect of the agricultural practices on disease severity including the molecular tool to quantify common scab pathogen and the measurement of isothiocyanates are ready to be used.

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