PROGRESS REPORT TO THE MAINE POTATO BOARD
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Project Title

Building Varietal Resistance to Disease using Marker-assisted Selection

Investigators

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Executive Summary

A key strategy for managing crop disease is improving varietal resistance. This strategy is particularly useful for PVY, late blight, and golden nematode (GN) in potato, as these resistance genes are dominant and stackable, and when present provide immunity (PVY) and resistance (late blight and GN). Marker-assisted selection efficiently identifies the presence or absence of some of these genes in potato, expediting development of new varieties with these resistances. This project continues to facilitate the development of PVY, late blight, and GN resistant potato clones through a marker-assisted selection program.

Late blight: Protocols for markers associated with late blight genes *R1*, *R2*, *R3a*, *R3b*, *R8*, *R10*, *RB/Rpi-blb1*, *Rpi-blb2*, *Rpi-blb3*, *Rpi-phu1*, *Rpi-abpt1*, and *Rpi-mcd1* are being applied to known resistant parental material, based on pedigree. The potential genetic interactions of these genes (i.e. if there is suppression or additive benefits where multiple genes are present) are being studied through phenotypic responses by these clones and varieties to different late blight races.

PVY: Two PVY markers (RYSC3 for Ry_{adg}, and YES3 for Ry_{sto}) are currently being used to quickly identify PVY-resistant clones. A third marker (Ry186, a marker for Ry_{chc}) has been added for future use in our marker-assisted breeding efforts.

Golden nematode: The H1 marker is used to identify golden nematode resistance.

The information found in this program is used for clone selection in the immediately succeeding assessment cycle. It is also used to select parents for use in our crosses and this result in more rapid progress in developing disease resistant varieties. In the long-term, we can identify potential varieties with multiple resistances useful to the Maine potato industry.

The Maine Potato Board has provided funding since 2012 to incorporate this new technology into the UMaine Potato Breeding Program for the purpose of speeding selection of resistant varieties within the program. This project builds on that foundation, further solidifying and developing the marker-assisted approach for Maine's needs: increasing the likelihood of keeping resistant material through the selection process and improving the chances of releasing varieties with late blight, PVY, and GN resistance.

Duration of Project

We requested funding for April 1, 2017 to March 31, 2018 to continue marker-assisted resistance breeding as an important part of the potato breeding pipeline at Aroostook Research Farm. Our goal was to continue the implementation of simple and effective DNA-based protocols for genotyping established resistant varieties, new clone selections, and their derivatives for three disease resistances.

Objectives

To reduce the impact of late blight through the development of new varieties and breeding lines with effective resistance to diverse populations of the late blight pathogen

To reduce the impact of PVY and golden nematode by identifying sources of resistance within the program

To use DNA marker-assisted selection strategies for combining known resistances genes for one disease (e.g., late blight resistance in *R1*, *R3a*, etc.) with others (e.g., PVY resistance in Ry_{adg}, or Ry_{sto}) in common genetic backgrounds that are economically important to the potato industry in Maine and the eastern United States

Grant Received

$6,000 ($6,000 was requested)

Progress

This past year was the first that the marker-assisted selection program delivered PVY and H1 results to the UMaine potato breeder ahead of schedule. While the total number of samples to be tested was slightly reduced from previous years, this advancement can be attributed to experience of the laboratory technicians and the tweaking of the protocols to improve efficiency and consistency. In addition to these three genes, and in recognition of the difficulty the problem PVY causes to the industry, we tested two recently identified markers for a PVY resistance gene derived from *Solanum chacoense* (Ry_{chc}). Both markers were tested and the protocol was confirmed; one marker, Ry186, was selected because its length may lend itself well to potential multiplexing, or combining multiple markers at once per sample. While not fully implemented in 2017, this marker protocol is ready to be used for future years since Ry_{chc} genes have now been introduced to Maine clones via crosses with Sakai 35, a source of this potentially valuable PVY resistance gene.

In 2017, 214 samples were tested with markers RYSC3, YES3, and H1. Twenty-two clones tested positive for RYSC3. Thirteen clones tested positive for for YES3. Twenty-five clones tested positive for H1. Six clones were positive for multiple genes. Two clones tested positive for RYSC3 and YES3. Two clones tested positive for RYSC3

and H1. Two clones tested positive for YES3 and H1. No clones were positive for all three markers.

Late blight resistant gene protocols continued to be tested against known resistant potato varieties and clones. Just as protocol tweaking took place to improve the efficiency and consistency for the PVY markers, 2017 was a year of tweaking for these markers, albeit with less success. The results for *RB/Rpi-blb1* are completed, and join *R1*, *R3a*, and *R3b* as markers ready to use in the marker-assisted selection protocol. *R8* has consistently run negative for all samples. The marker for *Rpi-phu1* has also run negative for every sample, however a recent accession of germplasm will serve as a check to confirm the accuracy of this test (this test should be confirmed by February 13). The protocol in the literature for *R10* was ambiguous, and initial runs ran negative. Markers are run in pairs, a forward primer and reverse primer applied to the DNA sample; in this case, I used the incorrect the reverse primer. I have what I believe are now the correct primer pair, and am hopeful for the next run. Markers for *Rpi-blb2* are also in hand and are slated to run by the end of February. The protocol for the markers of *Rpi-blb3*, like *R10*, is ambiguous and will be run last in the program.

The phenotyping greenhouse experiment was set back this past summer by an unexpected plague of cockroaches, which ate approximately 25% of the research material. It was necessary to wait for harvest to obtain healthy tubers to plant. This experiment will be carried out in February and March, tested as a whole plant assay against late blight race US24.