



Canadian Agri-Science Cluster for Horticulture 2

Progress Report October 2015

Activity 12, Apple 11

New Biological Control Agents for Postharvest Diseases of Pome Fruit

Lead Researcher

Louise Nelson, PhD, University of British Columbia

Collaborators

Deena Errampalli, PhD, Agriculture and Agri-Food Canada

Jennifer DeEil, PhD, Ontario Ministry of Agriculture, Food and Rural Affairs

Danielle Hirkala, PhD, British Columbia Tree Fruits Cooperative

Bill Wolk, PhD, British Columbia Tree Fruits Cooperative

Peter Toivonen, PhD, Agriculture and Agri-Food Canada

Activity Objectives

We have identified several bacterial antagonists from Canadian soils which show efficacy against postharvest pathogens of pome fruit under controlled atmosphere and air storage. Our objectives are to test these antagonists in storage trials in British Columbia and Ontario in order to assess their potential for commercial development. This will be accomplished by determining:

1. the efficacy of the bacterial antagonists under varying storage conditions and with different fruit varieties,
2. optimal concentrations and timing of application of the antagonists,
3. the effect of the antagonists on fruit quality,
4. the performance of the antagonists alone or in combination with other chemical control methods
5. the suitability of the antagonists for commercial development

The long-term objective of this research is to enhance the quality and safety of Canadian fruit and increase Canada's competitive ability in global markets.

Research Progress to Date

British Columbia: For 2015-16, the British Columbia postharvest disease management studies were conducted using four apple cultivars, Gala, McIntosh, Spartan and Ambrosia, obtained from the BC Tree Fruit Cooperative, Winfield, BC. Commercial storage trials of all four cultivars began between September 19 and 27, 2015. We have initiated three experiments related to objectives 2-4. The first experiment investigates the optimal concentration of the antagonists (obj. 2). We are testing the antagonist, *P. fluorescens* 4-6, at 4 different concentrations, 10^6 , 10^7 , 10^8 and 10^9 CFU/ml, on two apple varieties, McIntosh and Ambrosia, against blue mold (*P. expansum*), gray mold (*B. cinerea*), and mucor rot (*M. piriformis*). Following inoculation the apples will be incubated for 15 weeks at

4 °C and evaluated for disease incidence and lesion diameter every 5 weeks. Our second experiment addresses objective 3. Three antagonists are being tested for biological control activity against blue mold on all four apple cultivars. Positive and negative controls, the chemical fungicide, Scholar, and the registered biocontrol, BioSave, will be compared with three strains of *P. fluorescens*, 4-6, 1-112 and 2-28. Following inoculation, the apples will be incubated at 4 °C and evaluated for disease incidence and lesion diameter every 5 weeks for 15 weeks. After the 15 week incubation period the apples will be assayed for firmness, starch, sugar and malic acid content, in order to assess the effect of the antagonists on fruit quality. Our third experiment addresses objective 4. We will test the efficacy of the antagonist, *P. fluorescens* 4-6 in combination with the GRAS compounds sodium bicarbonate, salicylic acid or calcium chloride against the three fungal pathogens on McIntosh and Ambrosia apples. The antagonist in combination with each GRAS compound and each pathogen was inoculated onto wounded apples. Positive and negative controls, the chemical fungicide, Scholar and the biocontrol, BioSave, were also included. The apples will be incubated at 4 °C and evaluated for disease incidence and lesion diameter every 5 weeks for 15 weeks. To further our understanding of the mechanism of action of the antagonists scanning electron microscopy studies of the antagonist and each pathogen were conducted and showed that *P. fluorescens* 4-6 strongly adheres to and degrades the hyphae of the fungal pathogens on culture plates. Further studies of the pathogen/antagonist interaction on wounded apples are planned.

Ontario: For 2015-16, in Ontario postharvest disease management studies were conducted using apples from three different cultivars, Gala, McIntosh and Empire obtained from Agriculture and Agri-Food Canada (AAFC) Farm in Jordan Station, Ontario from trees maintained according to standard orchard practices. Commercial storage trials were commenced between September 17 and October 28, 2015. We have initiated 2 major experiments for objective 4 - to determine the performance of the antagonists alone or in combination with other chemical control methods for control of blue mold (*P. expansum*) and gray mold (*B. cinerea*) of apples. In the first experiment, 4a, we are testing the combination of two select antagonists, *P. fluorescens* 4-6 and *P. fluorescens* 1-112 alone or in combination, against blue mold and gray mold on McIntosh and Empire apples. Positive and negative controls, as well as the chemical fungicide, Scholar are being compared against *P. fluorescens*. Following the inoculations, the apples will be incubated at 4 °C and evaluated for disease incidence and lesion diameter every 4 weeks for a total of 24 weeks. After the 24-week incubation period the remaining apples will be placed in a Conviron growth cabinet at 20 °C and no light for 1 week and then evaluated for disease incidence. To determine the quality of apples, physiological parameters will be tested on 10 apples of all the three cultivars each month by DeEll's laboratory. These apples will be stored at 4 °C. For the second experiment, 4b, we are assessing the application of *P. fluorescens* 4-6 in combination with GRAS compounds, sodium bicarbonate, salicylic acid, and calcium chloride for the management of blue mold and gray mold on Gala and McIntosh apples. The combination of the antagonist *P. fluorescens* 4-6 and one of the GRAS compounds were mixed with pathogen and applied to the wounded apples by drenching. Positive and negative controls, as well as two chemical fungicides, Scholar and Mertect, were also compared against blue mold and gray mold. Following the treatments, the apples will be incubated at 4 °C and evaluated for disease incidence and lesion diameter every 4 weeks for a total of 24 weeks. After the 24-week incubation period the remaining apples will be placed in a Conviron growth cabinet at 20 °C and no light for 1 week and then evaluated for disease incidence.

Early Outcomes (if any) or Challenges

Studies in British Columbia and Ontario show similar commercial storage trial results for the three *Pseudomonas fluorescens* strains. The level of control provided by the antagonists varied with pathogen, apple variety and storage environment and was less than that observed for the fungicide Scholar.

Key Message(s)

Our findings suggest *P. fluorescens* provides control comparable to BioSave, but was less effective than the synthetic fungicide Scholar. In order to achieve levels of control comparable to fungicides, *P. fluorescens* will need to be applied in combination with food additives (currently being tested) or lower doses of fungicides.

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