PROJECT TITLE: Establishing a Grower-Operated Fire Blight Detection Lab

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PROJECT SUMMARY

Fire blight has become a severe disease for Utah apple growers over the last several years. Losses in some Utah County orchards reached up to 20% in 2008. The disease spreads quickly, especially in high density orchards and in susceptible varieties such as 'Fuji', 'Gala', or 'Honeycrisp', which are commonly grown due to high market demand. Utah growers are limited in treatment options due to the buildup of resistance to the most effective antibiotic (streptomycin). The alternative antibiotic option is less effective, and must be applied at a precise timing to be effective. As a result, growers need instant notification if their trees are harboring the bacterium (*Erwinia amylovora*) that causes fire blight.

This project used a molecular DNA test called LAMP (loop-mediated isothermal amplification) to detect fire blight bacteria in apple flower clusters. LAMP has been previously shown to detect the bacteria in a few hours using reagents and equipment that are relatively cheap and safe. We set up the first ever "grower lab" at an apple processing plant in Utah County, and compared results from the grower lab to the same test run at the Utah State University cooperator lab. Historically, this apple processing plant has served the growers by detecting fire blight bacteria via flower stigma imprints onto an expensive growth media called CCT. If present, bacteria are detected in 36-48 hours, a timeframe that is too late to implement effective control. When LAMP tests were positive in the grower lab, we notified growers to treat when weather conditions were optimal for infection.

PROJECT APPROACH

The project team, Marion Murray (IPM Extension Associate, USU), and Doug Rowley (Manager, Mountainland Apple Processing Plant) conducted the LAMP testing according to specifications developed by Dr. Ken Johnson and Todd Temple, at Oregon State University. Marion and Doug purchased lab equipment, materials, and reagents needed to operate a grower lab at Mountainland Apples processing plant.

The LAMP tests ran from the start of bloom (May 9) to petal fall (May 27). Six growers in Utah County participated by collecting 100 flowers from selected blocks every day through the testing period and delivering them to Mountainland apples. Doug Rowley processed the flower clusters to 6 wash water samples each day, which were then driven to USU in Logan that same morning. Both labs then used the same wash water to run LAMP tests every day for 17 days, and then the two labs compared results. We also sent material from the first week of blossom collection to the USU plant pathology lab run by Claudia Nischwitz (Plant Pathologist, USU) to implement the CCT testing so that we could have a third comparison.

Upon initial detection of the fire blight pathogen via the grower lab LAMP testing, we notified growers by email and alerted them to use the fire blight risk model, called Cougarblight, to determine whether a treatment was required in their orchard. We inspected orchards at the end of the summer and communicated with participating growers and found that most of the orchards had light infestation (less than 1 infection per tree), but one grower had a small block of infested trees, and had to remove 30 trees.

The LAMP results for detecting the presence or absence of bacteria in the grower lab matched the USU lab 82% of the time. The results of the grower lab matched the USU plant pathology lab CCT tests 70% of the time. This error rate is not good enough to guarantee fire blight bacteria test results to growers. Molecular work takes time to perfect, and the inconsistency could have been due to low concentrations of *Erwinia*, contamination, or human error. The developers of the LAMP test confirmed that when low populations exist on the flowers, results can be inconsistent, perhaps because there is not a step in the LAMP procedure to concentrate the samples to increase cell numbers for template preparation. Spring temperatures in 2010 and 2011 were lower than normal and not conducive to bacterial growth during the first half of apple bloom.

LAMP, however, still appears to be a promising rapid detection test when conducted from a grower-sustained operation that is located in close proximity to apple growers. Results from LAMP testing, when compared to weather data and the Cougarblight fire blight forecast model, are useful to determine the need to spray with an antibiotic. We plan to continue to operate the LAMP testing for the 2012 growing season, and will charge growers $9/test, which will cover the cost of supplies. The Oregon State University lab has agreed to verify our LAMP results with their own testing. If we can get to a point of consistent results, this lab will be the first of its kind conducting this type of testing.

Results of this project will be shared with the apple growers at the winter 2012 annual Utah State Horticultural Association growers meeting. Because of the inconsistencies between the two labs, a paper was not written for the industry magazine, Good Fruit Grower, but after a season of self-sustaining operation in spring 2012, we plan to submit a paper in the summer of 2012.

GOALS AND OUTCOMES ACHIEVED

1. *Reduced (optimized) antibiotic sprays*. LAMP technology will enable growers to apply antibiotic sprays only when bacteria are active. Growers who participated in this project **responded to a written email survey**, and the average reduction in their antibiotic sprays was 20% as compared to 2010, and 25% as compared to 2009, a savings of $50/acre per treatment. Growers who participated in the survey reported a savings, on average, of $10,000 per 200 acre block in 2011, and $20,000 per 200 acre block in 2010.

This savings allowed growers to offer affordable fruit to local markets with little to no antibiotic residue.

We expected a 25% reduction for both years, but weather conditions in the spring of 2010 resulted in a reduced need to spray anyway, so that sprays in 2011 were already greater, even without the LAMP results. Reduced antibiotic sprays leads to other benefits: **reduced** farm worker and **consumer exposure** to agricultural antibiotics, reduced production costs, and lowered risk of antibiotic resistance in bacterial populations in orchards. A survey of bacterial populations' resistance to streptomycin was conducted in a separate USU project, the first since 2007. Results were similar to the prior survey, which means that resistance has not increased.

2. *Multi-state collaboration.* Utah State University and Mountainland Apples are collaborators of the LAMP work being done by plant pathologists at Oregon State University. Our results have been used by their lab to help to improve the LAMP testing. Collaborating with a large university such as OSU gives USU visibility and access to expertise and resources beyond our capabilities.

3. *Creation of the only lab of its kind in the western U.S.* The Mountainland grower lab is the only lab of its kind in the western U.S., and with the addition of the service fee starting in 2012, it will be self-sustaining, providing quick and effective fire blight detection results, allowing for implementation of control measures only when necessary.

4. *Student training* – an undergraduate student in the Nischwitz plant pathology lab received training in preparation of CCT media, culturing plant tissue, and identifying Erwinia cultures as part of this project. As a result of her work, she now envisions pursuing a Masters Degree in plant pathology.

BENEFICIARIES

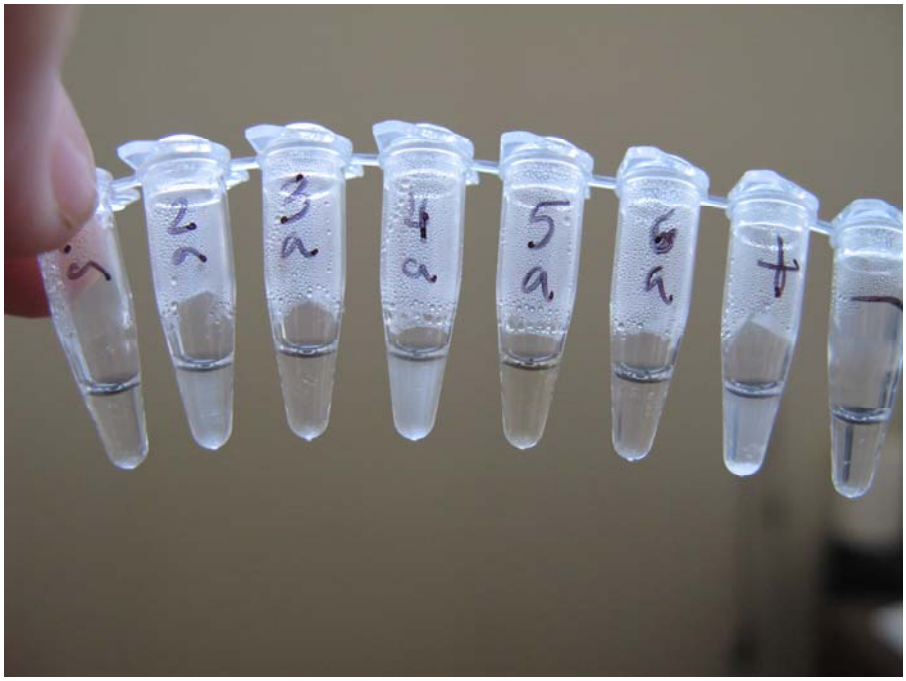
This project directly benefits apple growers in the Utah County region (approximately 1056 acres and 70 operations). Grower organizations in other states can benefit from the success of Utah's grower-operated lab and can utilize its services or learn about setting up their own lab. There are no data for the economic impact of fire blight in Utah, but a severe epidemic, if left unmanaged or mismanaged, could reduce yields by up to 50%, and if a high density planting is destroyed, could cost the grower a $5,000-10,000 loss per acre in a single season. LAMP testing results can help apple growers to be more profitable.

LESSONS LEARNED

1. The LAMP molecular detection of fire blight is a valuable tool for fruit growers, and providing a high speed detection service within driving distance of apple orchards is important, and will provide growers the peace of mind to know whether an antibiotic spray is necessary to prevent serious losses.

2. It is important to compare results of molecular testing with direct culturing of the bacteria onto specialized growth media. The comparisons we conducted showed that the Mountainland grower lab needs further refinement the LAMP protocol. Using the remaining primers and reagents, we will continue to work on the molecular techniques with test runs in winter 2012 to be ready for the spring 2012 season.

ADDITIONAL INFORMATION



In LAMP testing, sample tubes that are positive will have cloudy water while negative samples will have clear water. The sample numbers with cloudy water represent the detection of *Erwinia amylovora*, the bacterial pathogen that causes fire blight, from within healthy-looking flower clusters collected that same day. Using this test, results can be seen within 3-4 hours and growers can be better prepared to implement treatment based on the fire blight risk prediction model.