

END-POINT DETECTION OF PIERCE'S DISEASE IN WINE GRAPES USING RAPID TEST KITS

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INTRODUCTION

Pierce's disease (PD), caused by the bacterium *Xylella fastidiosa*, is a major challenge to sustainable wine grape production in Georgia. Pierce's disease is vectored by sharpshooter insects which can rapidly spread the bacteria throughout a vineyard. Cold winters help to moderate the effects of PD, however, mild winter temperatures in recent years have led to increased spread of PD in north Georgia. When PD is detected in a vine, the recommended method of treatment is immediate vine removal, which helps to prohibit the spread of inoculum to surrounding vines. Currently, established methods of disease identification are diagnosis of visual symptoms and expensive multi-day laboratory testing for confirmation. Recent advances in technology have allowed for rapid detection. Agdia's AmplifyRP XRT rapid detection kits were purchased for a pilot program and were housed in the Lumpkin and White County Extension offices. Grape producers and Extension agents were encouraged to send in suspected PD-positive samples for same-day diagnosis free-of-charge.

METHODS

Test samples were collected from 14 different grape cultivars. Fifty-five test samples were collected from 16 vineyards throughout 11 counties in north Georgia. Petioles were collected analyzed at the Lumpkin and White County Extension offices. 300 mg of petiole tissue was inserted into a mesh extraction bag. 3 mL of extraction buffer was added and the petiole tissue was ground in a mesh bag with the end of a pen. 1 mL of pellet diluent fluid was added to a 1.5 mL micro-centrifuge tube, and 5 µL of sample extract was mixed with the pellet diluent. 25 µL of sample extract was transferred from the micro-centrifuge tube to a reaction pellet tube, mixed well, and placed into a portable heat block for 4 minutes. Pellet tubes were then mixed again and reinserted into the heat block for 16 minutes of further incubation. Pellet tubes were then removed and inserted into the Amplicon detection chamber, where it mixed with a solution and reacted with a lateral flow strip. Results were interpreted after a 20-minute wait period.



Images 1,2,3: Visual symptoms of Pierce's disease in wine grapes

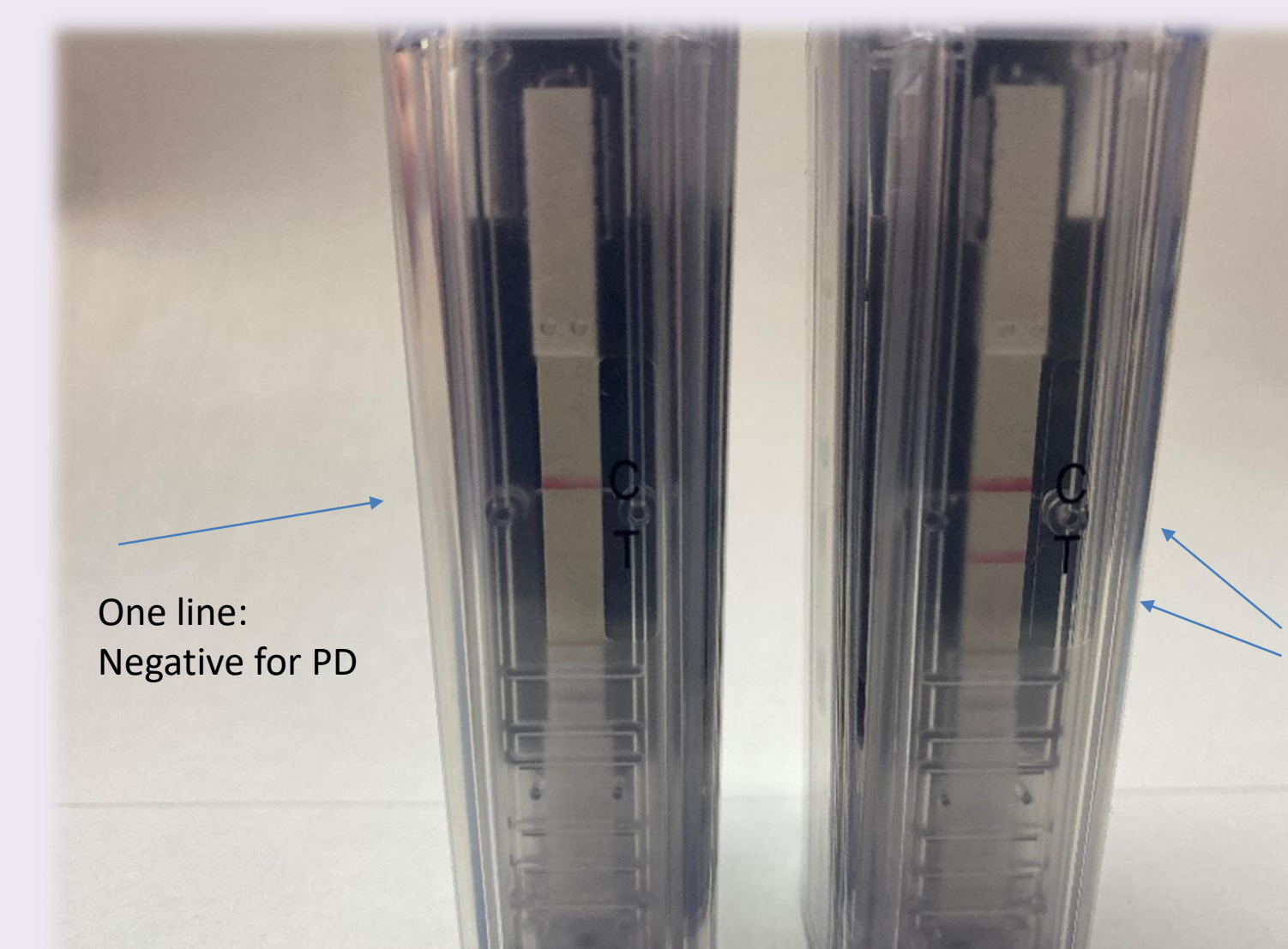
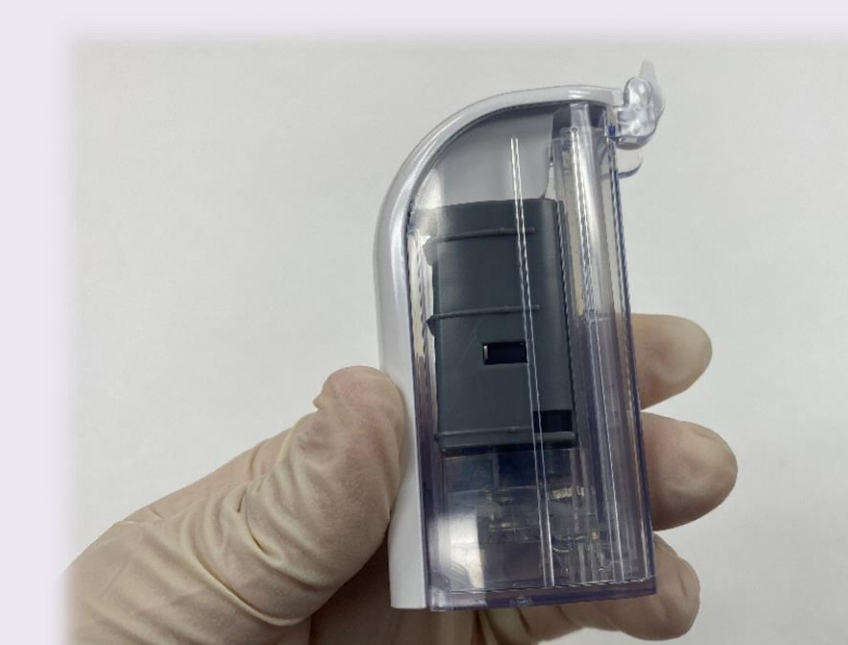
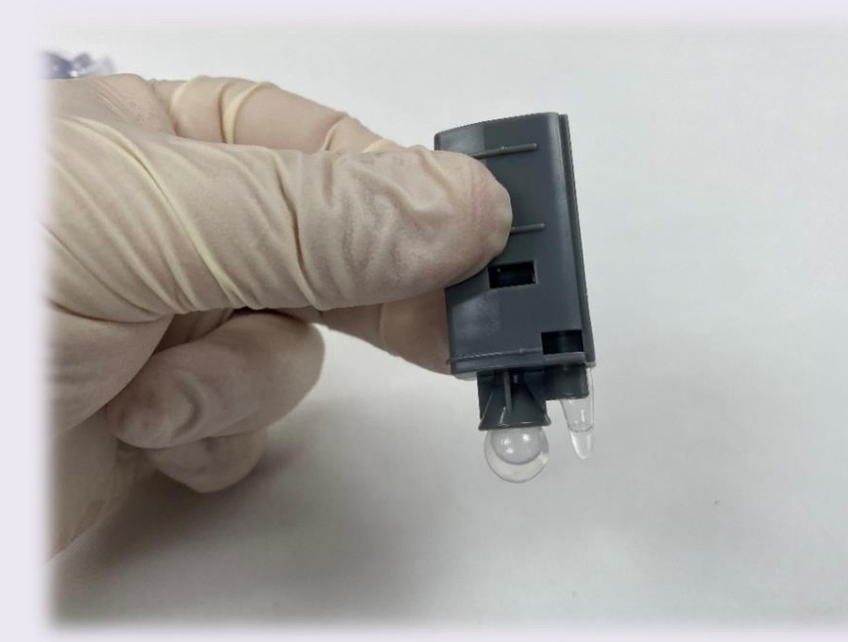
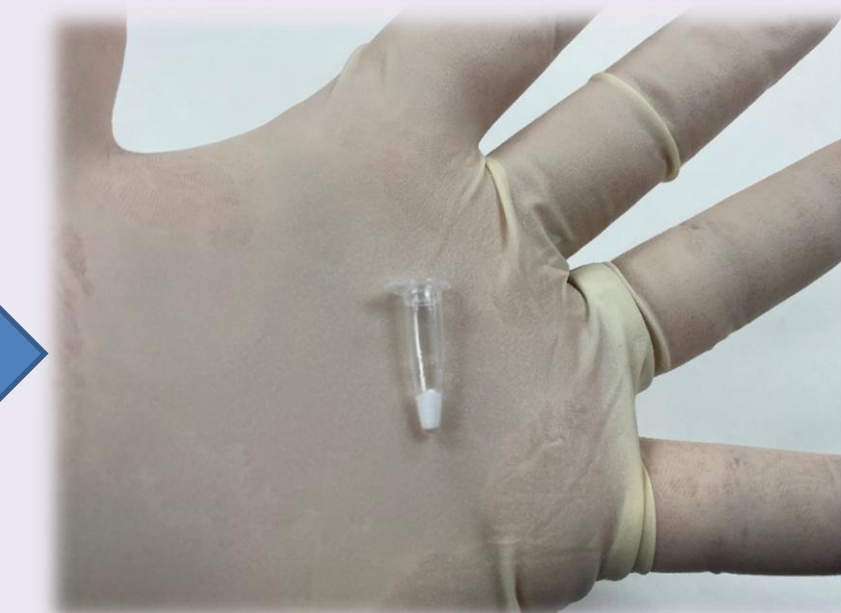
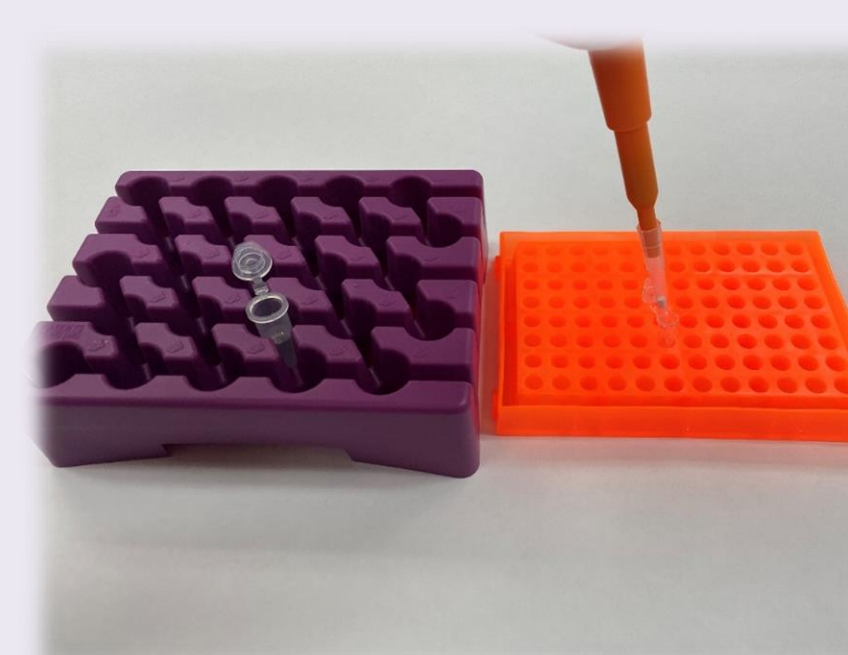
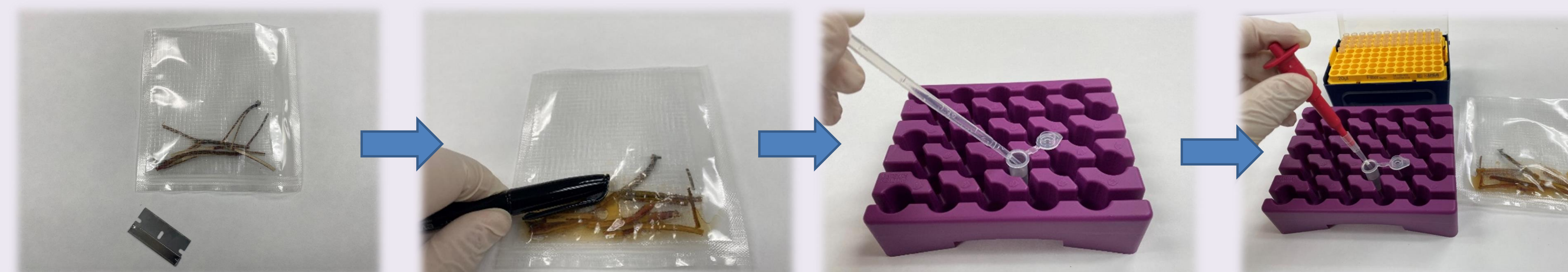


Image 2



Image 3

AmplifyRP XRT test sampling procedure



One line:
Negative for PD

Two lines:
Positive for PD

RESULTS

- ❖ Tissue samples were collected from vineyards in Barrow, Cherokee, Elbert, Fannin, Gilmer, Habersham, Lumpkin, Rabun, Towns, Union, and White Counties.
- ❖ 38% of the 55 samples tested were confirmed positive for Pierce's disease infection. *Not all samples tested were suspected positive for PD. Some were tested to see if known PD-resistant varieties, like Norton, carried PD inoculum without showing symptoms*
- ❖ Typical sample completion time was under 1 hour, compared to a 2 – 3 week turnaround time for samples shipped to a lab for diagnosis.
- ❖ One-on-one agent evaluations revealed that grape producers surveyed were very pleased with the rapid response time from the AgDia AmplifyRP XRT kits.
- ❖ Many samples suspected to be PD-positive through visual assessments were found to be PD-negative, which shows that visual assessment methods alone are not always accurate for diagnosing Pierce's disease.

CONCLUSIONS

- ❖ AgDia's AmplifyRP XRT rapid detection kits are a quick, efficient alternative to lengthy laboratory analysis of Pierce's disease infections in grapevines.
- ❖ Rapid detection kits reduced results turnaround time by up to 95% compared to laboratory testing.
- ❖ At \$2500 for a package of kits which tests 48 samples, cost might be prohibitive for small and medium-sized vineyards.
- ❖ However, with relatively minimal grant funding, or by charging a small fee, kits could be stocked in multiple UGA Extension county offices to serve the PD diagnostic needs of grape producers throughout the state.
- ❖ Rapid Pierce's disease testing could result in an annual savings of \$675,900 for vineyards in the participating counties through removal of infected vines and prohibiting PD from spreading.