2011 SRSFC report

Title: Understanding blueberry mosaic disease

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Research Proposal

PI: Ioannis E. Tzanetakis, Dept. of Plant Pathology, Division of Agriculture, University of Arkansas, Fayetteville, AR 72701, Tel: 479 575 3180, Fax 479 575 7601, itzaneta@uark.edu
co-PI: M. Elena Garcia, Dept. of Horticulture, Division of Agriculture, University of Arkansas, Fayetteville AR 72701, 479-575-2789, Fax: 479-575-8619, megarcia@uark.edu

Objectives

Our efforts will focus on **A.** understanding the causal agent of the disorder, including its identification and characterization, **B.** develop detection protocols that will be used to survey fields and determine the areas affected by the agent.

Justification

Blueberry has expanded dramatically in the last twenty years and the fruit value reached historic highs primarily because of the publicity of the beneficial effects of the antioxidants carried in the fruit. Arkansas has the environment to grow blueberries and the potential to become a major growing state in the Midsouth, alas the industry is lingering due to cultivation problems that seems to be a combination of cultural, abiotic and biotic stresses, caused primarily by pathogens. The industry may be facing another problem in Mosaic or Variegation. The disease has been reported in major blueberry producing areas (West coast, Michigan, New Jersey) and the affected area is expanding (Converse, 1987). During a blueberry virus survey in Arkansas in 2009, we observed several plants showing typical mosaic disease symptoms. In 2010, the disease was observed in additional plants indicating movement of the causal agent. Mosaic was never reported in Arkansas before and its discovery in the state adds to an already stressed industry

Methodologies

Total nucleic acids were extracted from Arkansas symptomatic material and used in DOP-RT-PCR as described (Tzanetakis and Martin, 2008) to obtain a cDNA library. The DNA was used for deep sequencing using the Illumina platform. Sequence analysis was done as described (Laney et al., 2011). Several virus-like sequences were found in the particular plant used in the analysis. Those sequences were used to develop RT-PCR protocols and determine the role of the new viruses in blueberry mosaic disease.

Results
The RT-PCR protocols developed were used in 25 mosaic samples collected from AR, MI, OR and NJ. One of the new viruses identified using deep sequencing was found in all symptomatic material as well as two symptomless samples from AR that were growing in close proximity to symptomatic plants.

Conclusions
We identified a new virus closely associated with blueberry mosaic disease. This is the first step in understanding mosaic and study the disease in further detail. This will determine the disease impact in a deteriorating Arkansas industry. The finding on two asymptomatic plants in our survey is of concern. In the past, rogueing has been the approach used to minimize movement but if the virus is latent under particular environmental conditions or in particular cultivars it may be that eradication is a very inefficient control method.

Impact Statement
Diseases are difficult to predict given the interaction between the environment, the host and the pathogen. This is particularly important in the case of the mosaic virus and the fact that it has also been found in latent infections. There is no knowledge on the virus vectors, their vectoring efficiency and the interaction of the virus with the cultivars grown in Arkansas and elsewhere. Reports from other states indicate poor yield and overall weakened bushes. Given the situation of the Arkansas blueberry industry, where most farms have significant problems with plant vigor, an additional widespread problem could lead to further problems that the industry cannot afford. This project will help identify potential vectors and though the detection protocols minimize infected material movement given the latent infections. This will have a major effect not only in the farm level but also in nurseries, as non-meristemed clonal propagation of blueberry could lead to dissemination of the agent to thousands of plants

Citations:

Converse RH, ed. 1987. Virus diseases of small fruits. US Dept of Agric. Handbook 631.

Laney, A.G., Keller, K.E., Martin, R.R. and Tzanetakis I.E. 2011. A discovery 70 years in the making: Characterization of the Rose rosette virus. Journal of General Virology 92: 1727–1732.

Tzanetakis, I.E. and Martin, R.R. 2008. A new method for extraction of double stranded RNA from plants. Journal of Virological Methods 149:167-170.