

Using Genomics to Advance the Management of Pierce's Disease and Grape Breeding

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Introduction:

Many research programs today are using genomics approaches to provide solutions to viticulture problems. Genomics enables unique insights into insect biology, development, and pathology, as well as enables the development of cost saving methods to produce and screen new grape varieties. Researchers now have the tools to examine and/or identify hundreds to thousands of genes at a time, which provides researchers a means to conduct in-depth examinations of pathogen-plant-insect interactions, elucidation of their biology, and enable the creation of new tools and management strategies aimed at reducing disease spread and economic losses from plant diseases, their insect vectors, or other insect pests. As part of the overall national program on building a sustainable viticulture industry, a genomics approach was used to advance efforts to manage leafhopper vectors of Pierce's disease, and to produce tools to aid efforts to breed more disease and insect resistant grapevines.

Importance of Genomics: While the amount of genomic data available for grapes, grape pathogens, and insect pests of grape, has grown the use of genomics is still relatively new. The multinational effort by the international grape genome program, IGGP, has made good progress in advancing grape genomics efforts. As the research on grapevines moves steadily forward, genomic efforts on insects still lags far behind. Before we could conduct comparative genomic studies which provides valuable understanding of the biological processes which are functioning within leafhoppers the lack of genomic data had to be addressed. To address these needs for improved strategies of leafhopper management we produced genomic datasets from three leafhopper species known to be vectors of the plant-infecting bacteria, *Xylella*. [Glassy-winged sharpshooter- *Homalodisca vitripennis*; the Black-winged sharpshooter- *Oncometopia nigricans*; the Blue-green sharpshooters- *Graptocephala atrapunctata*]. The production of these genetic products provides the foundation for conducting many other studies on leafhoppers:

Genetic Products: Six new genomic datasets were produced where none was available before on leafhoppers. These cover three adult species, plus GWSS

5th instar, midgut, and salivary gland. The identification of thousands of gene transcripts and proteins, provides an increased understanding of the enzymes present in leafhoppers. Many of these proteins play a role in basic biology, such as development or reproduction, but others are specific to digestion, and/or salivation events which are linked to, or which aid the acquisition, transmission, or subsequent infection of plant diseases, like *Xylella*, within the plant. Having these genetic products enabled the building of new tools, such as a GWSS-microarray, used for further analyses. The use of these genetic products are being used to:

- Develop *genetic markers* for species identification and biotyping of leafhopper populations;
- Elucidate genetic basis of leafhopper feeding and digestion as relates to disease transmission, host plant selection and use;
- Provides the foundation and tools for *functional genomic* studies which identify important genes regulating and maintaining leafhopper development, diapause, and responses to insecticides.

Leafhopper Pathogens: Previously searching for *viral pathogens* in leafhoppers has been difficult and costly due to problems in sampling and analysis of field caught insects. Using either a sequencing strategy, or microarrays, provides a wide sample which can be screened across known insect viruses and other microorganisms, thus providing a rapid detection and identification of viral species detected in leafhopper samples. **a)** Three new leafhopper viral pathogens, from three different viral groups were discovered using these methods. These pathogens cause increased mortality in the nymphal stages.

Grapevine Breeding: The *identification of genes* linked to increased disease and insect resistance, drought stress, and other growth or quality traits aids efforts of grapevine breeders to produce more vigorous varieties. Use of genetic markers can also reduce the costs and time needed to develop a new variety for release. **a)** Genetic markers are being produced to facilitate marker assisted selection, grapevine breeding programs; **b)** A national North American Grape Genome database was established and provides access of the data to the research community, Florida A&M University. (*These objectives address NGWI initiative #1, #2, and #5*).

Summary:

Genomics approaches continues to provide valuable data needed for the development of new insect management strategies, and production of improved grapevine. All the genetic sequences produced during these projects from leafhoppers and grapevine have been published on the internet for public access. The open and continual production and release of genomic data, continues to advance research on leafhoppers and grapevine.

As is evident from the many other studies presented at this meeting genomics is having a major impact on how researchers are working to solve the viticulture problems of today and tomorrow.

* Today's research environment requires many collaborations. The work herein is no exception, Collaborators are listed in the individual summaries which were handed out. My sincerest thanks and appreciation for all those who have contributed to this effort.

Grapevine Identified Genes: Some of the gene functions identified and shown to be similar to genes in other grapes. Many disease resistance genes, are similar across different plant species. Resistance pathways in rice, soybean, or other plants provide backgrounds for understanding some of the genetic pathways in grapes.

5	Alcohol dehydrogenase [Vitis vinifera]	Stress Response
2	Catalase [Vitis vinifera]	Stress Response
3	Aux/IAA protein [Vitis vinifera]	Hormone Biosynthesis
20	Chalcone synthase [Vitis vinifera]	Color
1	Dihydroflavonol 4-reductase [Vitis vinifera]	Color
4	Leucoanthocyanidin dioxygenase [Vitis labrus...	Color
2	H ⁺ -protonophorase [Vitis vinifera]	Color
1	Malate Dehydrogenase [Vitis vinifera]	
2	NADH glutamate dehydrogenase [Vitis vinifera]	
1	Expansin [Vitis vinifera]	Growth
46	Plasma membrane aquaporin [Vitis v....	Growth
2	Pathogenesis-related protein 10 [Vitis vini...	Disease Resistance
5	Polyphenol oxidase [Vitis vinifera]	Defensive Compound
4	Chitinase [Vitis vinifera]	Disease Resistance
1	Proline rich protein 2 [Vitis vinifera]	
1	PR-4 type protein [Vitis vinifera]	Fruit Ripening
2	GA-like protein 1 [Vitis vinifera]	Fruit Ripening
1	Beta-galactosidase BG1 [Vitis vini...	Fruit Ripening
6	Ripening-related protein [Vitis vi...	Fruit Ripening
1	Pyroline-5-carboxylate synthetase [Vitis	Fruit Ripening
2	Resistance gene analog [Vitis vinifera]	Powdery Mildew Resistance
1	SCUTL2 [Vitis vinifera]	
1	SINA2 protein [imported] - Vitis vinifera -sg14...	
3	Vacuolar invertase, GIN1 [Vitis vinifera-g...	Fruit-Sugar Accumulation

