Virginia Wine Board
Project #15-1662-01
Semi-annual Report - December 2015

Identifying genomic markers for fungicide resistance in grape powdery and downy mildew

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Objectives

1. Develop genomic markers associated with quinoxyfen (Quintec) resistance (QR) of grape powdery mildew.
2. Identify genetic targets of quinoxyfen as well as genes associated with resistance in grape powdery mildew.
3. Develop genomic markers to identify species and phosphite resistant isolates in grape downy mildew.

Powdery Mildew

Quinoxyfen: To date we have generated 340 million reads from two pools of DNA (10 resistant, 10 susceptible isolates). From these reads we have isolated 1000 highly differentiated markers as determined by Fisher's exact test. These markers can now be screened to identify a refined set that would be diagnostic for identifying resistant isolates. In addition, we have a panel of markers that can be screened for utility in identifying emergent resistance isolates. We now have sufficient fungal material to begin our experimental transcriptomics analysis, which aims to determine the genetic mechanism of resistance and co-locate a specific resistance marker within our genomic data.

Strobilurin: We have generated two pools of high quality DNA (10 resistant, 10 susceptible isolates) that are currently being sequenced. From these data and coupled genomic resources (annotated genome) we will identify a similar set of markers that can be screened to identify a refined set diagnostic for resistant isolates.

Genomic resources: In the process of generating the data for identifying markers unique to resistant isolates we are generating a substantial set of genomic resources that can be leveraged to generate a high quality genome assembly. We are taking advantage of new sequencing platforms and leveraging our existing data to generate an annotated high-quality draft genome sequence for our Virginia powdery mildew isolates. To date current

genome assemblies are limited to 14,000 scaffolds which is a major limitation to rapidly identifying markers and genes associated with resistance. We anticipate substantially improving this genome assembly and, along with our other data, releasing these data for public use.

Downy Mildew

We have collected a substantial number of isolates for genetic screening and are in the process of collecting sufficient DNA for sequencing. Reducing the genetic complexity in the samples by growing individual single-spored isolates has taken a substantial amount of time; however, at this point we have completed 70% of this collection. We are again taking advantage of new sequencing technologies to generate a large number of markers for our identified isolates. From these data we will identify several thousand markers, which will be refined to identify population-specific markers. We will also identify structural variants (large physical changes in the genome) which have been recently identified as important determinants in fungicide resistance among crop oomycetes.