



VitisGen Voice

Mapping the way to the next generation of grapes



How Low Can You Go? Understanding the Genetics of Low Temperature Responses in Grapevines

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The grapevine family (*Vitis*) has evolved over a wide range of environmental conditions, from warm and humid conditions in the subtropics to dry, desert-like climates, and extreme cold in the northern US and Canada. Because of its highly-valued fruit quality, commercial production has been historically dominated by a single grape species, *Vitis vinifera*, which evolved, and is traditionally grown in, milder, Mediterranean climates. When these grapes are planted in regions with significantly different climatic conditions, such as extreme temperatures in the winter or

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At a glance...

- One of the biggest challenges faced by growers in colder climates is selecting varieties that will withstand severe winters, mature during short growing seasons, and still produce an acceptable quantity and quality of fruit.
- The ability to resist damage after low temperature events like extreme cold winter temperatures and early spring frosts is dependent upon multiple, complex, physiological adjustments (acclimation) to events such as decreasing day length and the onset of low temperatures.
- *VitisGen* scientists are working to develop a better understanding of the genetic mechanisms involved in acclimation, dormancy and freezing tolerance, with the goal of giving breeders better information to help them develop new varieties that can thrive under climatic circumstances that previous made grape growing a major challenge.



Presspad Podcast #11: Winter Injury

<http://blogs.cornell.edu/presspad/2014/04/18/presspad-podcast-11-winter-injury/>



Dead primary (lower) and secondary (upper) grape buds due to winter cold injury. Different species and varieties of grapes have different abilities to survive particular cold temperatures, depending in part on their genetic makeup.

highly variable temperatures in the early spring, these vines can incur significant damage or even death. As interest and consumer demand has led to an expansion of grape cultivation to non-traditional growing areas with more drastic environmental variation, the need for broader information regarding the genetic and physiological mechanisms that impact survival and productivity has also increased, including an understanding of how grapevines protect themselves from extreme low temperatures or avoid early bud break in the spring.

One of the biggest challenges faced by growers in these kinds of locations is selecting varieties that will withstand severe winters, mature during short growing seasons, and still produce a sufficient quantity and quality of fruit. Winter injury can result in significant losses in both grape and wine production. For example, a single cold event in the Finger Lakes region during the winter of 2004, resulted in

over \$63 million in losses of crop and wine production (Martinson and White, 2004). Cultivars of *Vitis vinifera* can start to experience damage to buds and canes at temperatures below 0°F, while species that evolved in colder climates, like *Vitis labrusca* and *V. riparia*, can tolerate much colder temperatures without suffering significant damage. However, the fruit quality of these more hardy species is generally not valued as highly by consumers as that from *V. vinifera* cultivars. Thus, a greater understanding of the mechanisms underlying low temperature tolerance could allow for the development of new cultivars, as well as improve cultural practices for existing varieties.

Survival at low temperatures is dependent upon multiple, complex, physiological adjustments (acclimation) to events such as decreasing day length and the onset of low temperatures. Exposure to short days and colder temperature through the fall and winter initiates protective biochemical measures within the vines to minimize cellular damage and initiate dormancy. Acclimation and dormancy, while closely interrelated, are distinct phenomena. The dormancy of grape buds is an adaptive strategy for survival with multiple stages, including *paradormancy*, *endodormancy*, and *ecodormancy*. Each of these stages is crucial for bud and vine survival, but in this article we will focus primarily on endodormancy and ecodormancy.

“Survival at low temperatures is dependent upon multiple, complex, physiological adjustments (acclimation) to events such as decreasing day length and the onset of low temperatures.”

time when next season’s buds stop their development. Endodormancy prevents new growth from occurring in these buds when temperatures can vary widely, fluctuating between optimal and non-optimal growth conditions, and ultimately, endodormancy protects the grapevine from damage due to frost and dehydration as they prepare for the winter.

In fall, as day length and temperatures begin to decrease, the vine begins to prepare itself to enter winter by developing periderm (the outer layers of woody stems/roots) along the one-year old shoots and then enters endodormancy, the

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VitisGen Delivers: Fourth Annual Meeting Overview

By Elizabeth Takacs, VitisGen Project Manager



The fourth annual VitisGen meeting of project directors, collaborators, and industry advisory panel members took place January 8–9, 2015 in San Diego, California.

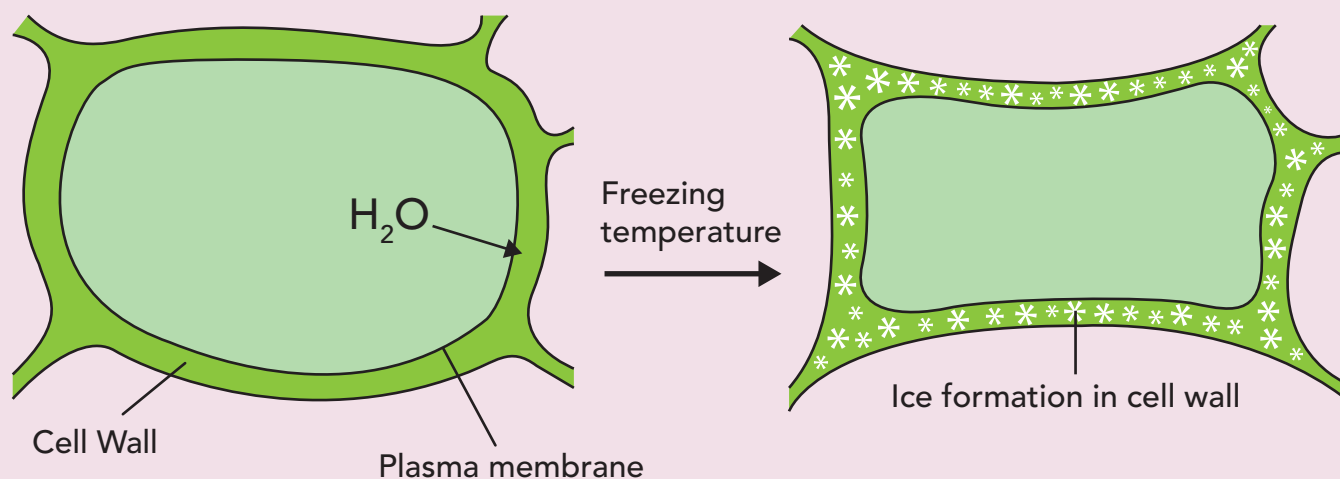
The fourth annual VitisGen meeting of project directors, collaborators, and industry advisory panel members took place January 8–9, 2015 in San Diego, California. Over the span of the two-day meeting, 33 VitisGen participants (including 13 project directors and 9 industry advisory panel members) reflected on the past year's accomplishments; communicated project deliverables; discussed and delineated plans for "VitisGen 2" (a successor project); facilitated international collaborations; and encouraged a continued dialogue and partnership with industry.

On the first day of the meeting, project directors, collaborators, and the industry advisory panel reviewed project accomplishments, discussed future goals, and learned about the endeavors by the international community to provide open data in grapevine breeding as well as French and European projects in grapevine breeding and genetics. Team reports on breeding, genetics, trait evaluation, extension and outreach, and trait economics focused on major accomplishments to-date, underscored impacts on the industry, and proposed immediate future goals. In addition to team reports, guest speaker Dr. Anne-

Françoise Adam-Blondon, Deputy Director of the Plant Biology and Breeding Department of the French National Institute for Agricultural Research (INRA), presented on *Open Data in Grapevine Breeding*.

On the second day of the meeting, breakout sessions were held to discuss VitisGen 2 plans to help delineate putative objectives and to receive feedback from the Industry Advisory Panel. Jean-Mari Peltier (President of the National Wine and Grape Initiative) and Bruce Reisch (VitisGen Project Leader and Professor at Cornell University) concluded the meeting by summarizing the key points from discussions held throughout the two-day meeting. The meeting was immediately followed by a workshop on genotyping-by-sequencing (GBS; a DNA based technology being used by project participants to map grapevine chromosomes and locate genes controlling important traits) data analysis. Twelve meeting attendees participated in the workshop facilitated by the two new VitisGen postdoctoral associates, Jonathan Fresnedo and Shanshan Yang. Further VitisGen advances are explained in the Team Updates section (beginning on page 6). ■

How do grapevines protect their tissues from freezing in the winter?



Credit: Levitt, J. 1980. *Responses of Plants to Environmental Stresses: Chilling, Freezing, and High-Temperature Stresses*, Vol. 1 New York: Academic Press.

To avoid being damaged by temperatures below freezing, buds use the process of 'supercooling' that reduces the amount of water in the cell, increases concentration of solutes and reduces freezing temperatures.

Water inside a cell is like water in a ziplock bag, which is in turn inside a shoebox. Inside the cell (the ziplock bag) are all of the important structures like the nucleus and the chloroplasts. When tempera-

tures go below freezing, the cells push water from inside the cell into the cell wall structure (shoebox) and extracellular space where it can freeze safely, away from those important organelles. With a lower concentration of water inside the cell, the remaining water will freeze at temperatures lower than 0°C, and thus allowing those cells within the bud tissues to survive below freezing temperatures.

Like many other fruit crops, grapes require a certain number of "chilling hours" during the dormant season in order to properly begin growth the following season. Beginning in endodormancy, grape buds begin to "count" the number of chilling hours, those between 0–7°C, they receive. Depending on the type of winter and geographic location of the vineyard, chilling hours may occur quickly (mild winter with lots of days above freezing) or occur slowly (cold winter with lots of days below freezing). Different grape varieties and species have varying chilling requirements (e.g., from 500–2000 hours) that must be met before bud break can successfully occur. This adaptation helps to ensure that new bud growth does not hap-

pen during short temperature fluctuations that may occur throughout the winter. The satisfaction of the chilling requirement causes the plant to enter ecodormancy. As the season progresses, extended periods of extreme temperatures promotes increase freezing tolerance, protecting the vines from environmental extremes—in this case, deep winter temperatures.

Once sufficient chilling has occurred and the vine enters ecodormancy it become responsive to increases in temperature in the late winter and spring. This exposure will cause grapevines to break dormancy and begin vegetative growth in the spring. The duration of the chilling requirement is essential to the success of the vine, especially early in the season. If

the winter is mild and the chilling requirement is not only met but surpassed, the vines will emerge from dormancy quickly in response to warm weather. The new growth may then be vulnerable to damage due to spring frosts. Although some cultivars are capable of producing a crop on secondary buds, reduced productivity and vine health are still significant concerns. If a vine's chilling requirements are not met by the time warm weather arrives, budbreak will be erratic, desynchronized, and extended in the spring.

Growers can, to some extent, influence the ability of their vineyards to survive low temperature events in the winter and spring. Tactics such as choosing cultivars adapted to local conditions, making better choices in vineyard site selection, using cultural practices such as canopy and cropload management, burying portions of vines, or possibly applying certain fertilizers or other materials prior to or during dormancy may contribute to improved survival. While these mechanisms meet with varied levels of success, the ability to identify markers linked to genes that influence low temperature survival and the timing of budbreak could have a much more significant impact on the ability of grapevines to thrive in many parts of the world.

Grapevines' responses and acclimation to low temperature events is a complex process that is influenced both by the environment where they are grown (and how they are grown), and by their genetic makeup. *VitisGen* scientists are developing a better understanding of the genetic mechanisms involved in acclimation, dormancy and freezing tolerance, with the goal of giving breeders better information to help them develop new varieties that will be able to survive and thrive in an increasingly wider range of climatic conditions. This work will allow scientists to more objectively evaluate a vine's cold hardiness or resistance to early bud break independent of environment, and develop new varieties that can thrive under climatic circumstances that previous made grape growing a major challenge, or even impossible. ■

Reference:

Martinson, T. and G. White. *Estimate of Crop and Wine Losses Due to Winter Injury in the Finger Lakes*. <http://www.fruit.cornell.edu/grape/pdfs/Cost of Winter Injury- Finger Lakes 2004.pdf>

VitisGen Vocabulary

Bud resilience

The ability of a bud to recover from low temperature.

Genotype

The combination of genes/alleles of an individual.

Chilling requirement

The minimum period of low temperature required before dormancy can be broken.

Vegetative growth

Growth of the vine, leaves, roots; the period of growth between germination and flowering.

Low Temperature Exotherm (LTE)

Temperature at which a tissue freezes, causing a release of heat energy.

Node

The structure, generally on the main stem, out of which a leaf or shoot will develop.

Paradormancy

Temporary cessation of growth due to conditions within plant but outside bud; for example, apical dominance.

Endodormancy

Temporary cessation of growth due to conditions within the bud. Period when the vine is accumulating chilling hours.

Ecodormancy

Temporary cessation of growth due to conditions outside plant; winter dormancy. Period after fulfillment of chilling requirements.

VitisGen Team Updates

By Elizabeth Takacs, Bruce Reisch and Lance Cadle-Davidson

Genetics

The genetics team has made extensive progress on identifying association of DNA markers with traits of importance using “next generation” DNA technology. Much of this progress can be attributed to the two postdoctoral associates, Shanshan Yang and Jonathan Fresnedo, who joined the project at the end of Year 3. Yang and Fresnedo have been generating genetic maps of key parents used by breeding programs, and performing analyses to identify marker-trait associations using trait data from the *VitisGen* trait evaluation centers and from local efforts within *VitisGen* breeding programs. As of the project meeting, high-resolution genetic maps had been completed for most of the targeted grape varieties and 52 marker-trait associations had been identified. The next step is to develop easy-to-screen detect genetic markers from these marker-trait associations, which can then be applied in marker-assisted selection, so that the most desirable seedlings can be identified based upon a simple DNA test.

Breeding

The breeding team has continued to maintain the core *VitisGen* grapevines used in genetic mapping and collaborate with the genotyping and trait evaluation centers to develop new genetic markers. As part of this cooperation, U.S. breeding programs have engaged in a large local trait evaluation effort screening 154 trait-population combinations. In addition to the contributions made toward developing new genetic markers, the breeding programs at Cornell University, the University of Minnesota, and the USDA-ARS have used previously identified markers for important traits to implement marker-assisted selection of seedlings (MAS) and marker-assisted parent selection (MAPS). To date, approximately 12,000 seedlings have been processed and an additional 2,000 vines representing germplasm and parental material were screened. The breeding team is now developing new populations to build future research endeavors on the current *VitisGen* infrastructure.

Trait Evaluation

The three trait evaluation centers continue to cooperate with breeders and the genotyping center to evaluate high priority traits necessary to develop new genetic markers. The powdery mildew center has optimized protocols and is focusing on three core populations to publish findings. Data from the phenotyping center was used to identify a novel qualitative source of strong resistance against a highly virulent powdery mildew isolate. The fruit quality center has analyzed over 30 fruit quality traits since 2011. Data sets generated by the fruit quality center from 2011–2013 have been used to pinpoint 16 quantitative trait loci (QTLs). Additionally, the fruit quality center has developed new measurements to evaluate tannin extractability and “blueness.” They have determined that pathogenesis related proteins, produced from species other than *V. vinifera* as well as hybrid grapes, are a likely factor limiting tannin concentrations in wines. The low temperature responses center has identified vines with slow and fast bud break responses, early acclimation responses, and de-acclimation dynamics in relation to chilling fulfillment. Thus far, data have been used to detect putative locations for genes controlling freezing tolerance, dormancy, bud break, and early acclimation responses. Additionally, a candidate gene for dormancy and freezing tolerance has been identified.

Extension and Outreach

The extension and outreach team has focused efforts on the public, industry, and the scientific communities. The project website, distribution of the project brochure, circulation of the project newsletter ‘The *VitisGen* Voice,’ and the publication of two short YouTube videos have served as outreach to all three audiences. The grape industry has been specifically targeted by six articles published in trade magazines or newsletters and 10 presentations delivered across the U.S. and Australia. *VitisGen* project participants have piloted much of the outreach to the scientific and research community. Overall, 50 presentations have

been delivered nationally and internationally, 10 articles have been published, and over 200 scientists have been trained in seven workshops.

Trait Economics

The trait economics team is continuing to work toward identifying top priority traits and documenting their impact. To identify consumer, grower, and market attitude to and demand for new varieties, the trait economics team developed two surveys. The first survey assessed researcher perceptions of top priority traits and the feasibility of incorporating those traits into new varieties. The top trait identified in this survey was powdery mildew resistance. The second survey shifted the emphasis on identifying grower attitudes toward adopting new varieties and their willingness-to-pay for specific varietal innovations. Currently, the results from this survey are being analyzed. The trait economics team has also been documenting the economic value of the adoption of new grape varieties

with either Pierce's disease resistance or powdery mildew resistance. For powdery mildew resistance, the trait economics team performed an aggregate analysis of pecuniary and non-pecuniary cost of powdery mildew management in California. They also produced budgets for representative vineyards to illustrate the potential benefits gained by transitioning from established to resistant varieties within three sectors of the industry (i.e., wine grapes, table grapes, and raisins), and regional benefits based on assumptions about adoption rates and timing.

The *VitisGen* team has surpassed some of the original project goals, and looks forward to continued contributions toward grape improvement efforts. Looking forward to another summer of data collection and analysis, DNA technology will again be employed to select the most promising seedlings in the weeks following germination. The team is actively planning a successor 5-year project to build upon the progress already made and to deepen our national impact. ■

Recent Publications and Presentations

Publications

Fellman S, Walter-Peterson, H. *VitisGen: Mapping the Way to the Next Generation of Grapes*. *Appellation Cornell*. Cornell University. March 2015. <http://grapesandwine.cals.cornell.edu/sites/grapesandwine.cals.cornell.edu/files/shared/Research%20Focus%202015-1.pdf>

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Stafne ET. 2014. Online Grape Production Information via eViticulture. ASEV-ES News. Spring 2014: 4-5.

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Adhikari P, Chen X, Sapkota SD, Chen L, Hyma K, Cadle-Davidson L, Hwang C-F. 2014. Identification of True Hybrids of *Vitis aestivalis*-derived 'Norton'-Based Populations Using Molecular Markers. 65th American Society of Enology and Viticulture (ASEV) National Conference, June 25–26, Austin, Texas.

Alston JM. 2014. Evolving varietal distinctiveness of US wine regions: comparative evidence from a new global database. Presented at American Association of Wine Economists Annual Conference. June, Whitman College, Walla Walla, Washington.

Barba P, Takacs EM, Hyma KE, Sun Q, Cadle-Davidson L, Reisch BI. 2014. Application of Genotyping-By-Sequencing in Crosses of Heterozygous Grapevines: Tools for Map Construction and Marker-Trait Association Testing. *VitisGen* Annual Meeting, February 20–21, Geneva, New York.

Barba P. 2014. Using next-generation sequencing for the genetic dissection of resistance to *Phomopsis viticola* in North American *Vitis*. 11th International Conference on Grapevine Breeding and Genetics, August, Beijing, China. (Abstract)

Best A, Gadoury DM, Seem RC. 2014. Temporal changes in concentrations of airborne inoculum in vineyards and effects of light quality on conidial production in *Erysiphe necator*. Proc. Geneva Summer Scholars Research Experience Program, New York State Agric. Exp. Station. Presented at Hobart and William Smith Colleges August, Geneva, New York.

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- Nowogrodzki A, Schaub M, Lagoz LC, Seem RC, Gadoury DM, Cadle-Davidson L. 2014. Challenges and Rewards of Powdery Mildew Phenotyping. *VitisGen* Annual Meeting, February 20–21, Geneva, New York.
- Ryona I, Sacks GL. 2014. Quantification of Green-leaf Volatiles in Horizon X Illinois 547-1 Population using Stable Isotope Dilution Analysis. *VitisGen* Annual Meeting, February 20–21, Geneva, New York.
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- Sapkota SD, Gadoury DM, Cadle-Davidson L, Seem R, Hwang C-F. 2014. Sporulation and Propagation of Downy Mildew of Grapes. *VitisGen* Annual Meeting, February 20–21, Geneva, New York.
- Sapkota S, Chen L, Hwang C-F. 2014. Norton (*Vitis aestivalis*) Grape Breeding for Powdery Mildew Disease Resistance. 65th American Society of Enology and Viticulture (ASEV) National Conference, June 25–26, Austin, Texas.
- Springer LF, Defnet E, Sacks GL. 2014. Mapping Wine Mouthfeel: Determining Phenotypic Measures of Tannin Extractability. *VitisGen* Annual Meeting, February 20-21, Geneva, New York.
- Springer LF, Sacks GL. 2014. Tannin Management. Proceedings of the B.E.V. NY Conference, Waterloo, NY. (Abstract)
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- Springer LF, Sacks GL. 2014. Understanding tannins in hybrid wines, an explanation of why extraction and additions present particular challenges. Midwest Grape and Wine + Craft Brew Conference, St. Charles, MO. (Abstract)
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