OSU Wine and Grape Research and Extension Newsletter

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In this issue:

- Welcome
- Vineyard Pest Scouting Workshop
- Spring-time Short Shoots and Grape Rust Mites
- Powdery Mildew Spore Trapping Project Update
- Spotted Wing Drosophila Control Strategies-Update
- 2010 Vineyard Floor Management Survey
- New viticulture diagnostic and learning modules online
- Brettanomyces
- New Viticulture & Enology scientific research articles
- OSU Alumni spotlight: Brett Weis
- Upcoming Events

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Welcome to the May 2010 Newsletter

As the season progresses and all anticipate bloom, we hope you take the opportunity to read this issue. It is full of useful information on some pests of concern—both old and new—in the vineyard and winery: powdery mildew, grape rust mites, Spotted Wing Drosophila and *Brettanomyces*.

We include other useful information and resources in this issue including a list of new research publications from the researchers in our Viticulture & Enology Team at OSU and USDA-ARS, and new online diagnostic and learning modules with information on grapevine nutrition and herbicide drift. Finally, you can learn more about the OSU Viticulture & Enology degree program from the *OSU Alumni Spotlight* article.

The OSU Winegrape Team

Vineyard Pest Scouting Workshop Felps Fone Qkills, Paise ?wareness

Earlier this month, OSU Viticulture Extension offered the Vineyard Scouting Workshop at the OSU Botany & Plant Pathology Farm in Corvallis to provide industry members with the newest information and ways to monitor and trap for various pests. The focus was placed on grape powdery mildew (Walt Mahaffee, Andy Albrecht and Tara Neill, all USDA-ARS), grapevine viruses (Karen Keller, USDA-ARS), plant parasitic nematodes (Inga Zasada, USDA-ARS), and various invasive insect pests, including Spotted Wing Drosophila (Amy Dreves, OSU) , mealybugs, phylloxera and *Lobesia botrana* (Vaughn Walton, Angela Gadino and Danny Dalton, all OSU) and beneficial organisms. A total of 90 industry members from across all regions of Oregon participated in the event. For those who could not attend, please visit our website for publications and more information regarding various pests (http://wine.oregonstate.edu).

Workshop participants learn to identify an important new pest in the US (*Lobesia botrana*) on pheromone traps (left) and identify male grape mealybugs under a stereo microscope (right).

Spring-time Short Shoots and Grape Rust Mites, a Continuing Saga

Patty Skinkis, Ph.D., Viticulture Extension Specialist, OSU

Grape rust mite (*Calepitrimerus vitis* (Nalepa)), an Eriophyid mite species, has been known to be problematic in Oregon's Willamette Valley over the course of the past ten years. This pest has been found to be associated with stunted spring growth referred to as "Short Shoot Syndrome" or SSS (Walton et al. 2007). Symptoms of SSS have been associated with mite presence are most often observed early in the growing season. These

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symptoms are described as delayed, stunted and deformed shoot growth (Figure 1), stem tissue scarring, deformed leaves, zigzagged

Figure 1. A stunted shoot with high mite presence in early May 2010.

stems, and deformed flowers. During late April and early May, the first signs of shoot stunting became apparent as new leaves began to unfurl (Figure 2). Early season spray programs targeting rust

Figure 2. Early signs of stunted shoots emerge in the head of a young vine, April 27, 2010. The small shoots may appear similar to frost damage. For this reason, it is critical to sample tissues for mite populations to discern the potential causes.

mites have been put into practice in the north Willamette Valley since 2007 in an attempt to reduce mite populations and feeding from the wooly bud stage through bud-break. Despite these best management efforts, symptoms of short shoot were widely observed this season from many areas within the north Willamette Valley, some of which have not had this problem in the past.

Research on SSS in Oregon indicates that grape rust mites play a role, but the cause-effect relationship is not well understood. Although research has ensued both in Oregon and abroad, there are still fundamental questions about Eriophyid mite and host-plant attraction and interaction that are not fully understood. An update of what we know (and don't know) about grape rust mites and other Eriophyid mites from other crops and regions are provided herein.

Pest damage

Grape rust mites are obligate pests of *Vitis vinifera* grapes. Because of their economic damage potential in certain grape growing regions, the mite is now considered in a permanent pest status in many regions of the world including Germany, France, Spain, Switzerland, Italy, Brazil (Duso et al 2010), Australia (Bernard et al 2005), and here in the Pacific Northwest (Walton et al. 2007).

Grape rust mites are phytophagus (feed on plants), preferring to feed on young grape tissues in early spring, causing damage that leads to deformation of shoots and leaves and results in surface necrosis described as scarring. It is believed that mites prefer newly developed tissues, potentially due to higher nutrient concentrations; however, this preference is not verified. The mite feeds on plant cells by inserting its stylet into a cell and consuming its contents, taking 10-20 minutes to feed. The saliva of some Eriophyid mite species studied are found to contain plant hormones such as auxin and cytokinin analogs, and when inserted onto or into plant cells can result in deformed tissue growth (Petanović and Kielkiewicz 2010). Generally, rust mite species are found to feed on the epidermal layers (surface layers of cells) of tissues only, not reaching into the phloem.

Although these tiny pests may seem like they will not do damage by feeding on individual cells, large populations in small areas of tissues such as buds and young shoots may cause significant tissue damage, reduced photosynthetic efficiency, reduced gas exchange and further stunted growth. The wound signal responses of the plant trigger feeding by other Eriophyid mites and activity of predatory insects and mites (Petanović and Kielkiewicz 2010). The effect of rust mite feeding has been found to be most damaging in cool springs when mite populations are active and there is little shoot growth is occurring, as observed in the Willamette Valley. The faster spring growth rate of vines in other grape-growing regions of the state likely prevent major grape rust mite stunting (Walton et al. 2007).

Infestation Patterns

Over the past years of research in Oregon, grape rust mite presence and damage across the Willamette Valley has largely been associated with young vineyards (<3 years old). Similarly, presence of the rust mite has been linked to young vineyards of only 1-2 years old in Europe (Zandigiacomo and Frausin 1998). This suggests that nursery stock may already be infested with Eriophyid mites. This is not uncommon as researchers identified nursery plant materials as a source of Eriophyid mites in Linden shade trees (Vaughn Walton, personal communication).

The other mechanisms of dispersal for grape rust mites include

their own movement within the vine canopy, by wind, and by human carriers. Mites are most effectively moved in wind (Michalska et al. 2010) and the second most efficiently moved by vineyard laborers (Michalska et al. 2010, Duso et al. 2010). We are uncertain as to the extent of mite infestations caused by either of these two factors in Oregon.

At this time, we hypothesize that plant materials used to establish vineyards in Oregon may contain low populations of grape rust mites or other Eriophyid mites at small enough populations that they do not cause visible damage in the first year of growth. However, with adequate environmental conditions, the mite can increase in population to a degree which allows for increased numbers of female mites going into overwintering sites in late summer. The spray programs often used in young vineyards may also help explain why populations may go unmanaged as sulfur (or other pesticides) is not being applied as frequently in that first year. Secondly, warmer vine microclimates can allow for greater suitability for mite reproduction. These combined factors can lead to higher mite populations going into overwintering sites and thus higher populations feeding on young tissues in the following spring. While this is a running hypothesis, further research needs to be conducted to better understand other vine physiological factors that cause stunting and how they relate to mite behavior and the cause-effect relationship of stunted shoot growth.

The question often arises as to the factors that may be attracting mites to particular vines or vineyards. Mite behavioral research indicates that mites have the ability to distinguish between host and non-host plants. If grape rust mites are on a host vine, they are able to feed and move slower than if they were on a non-host plant (Michalska et al. 2010). However, the ability to "choose" a particular vine or vineyard is not understood. The problem may be related more to a population magnification in the proper environment, loss of natural predators, and other vine physiological factors in combination with mites rather than the attraction of large populations of mites to a given vineyard site.

Identifying mite presence and management

It is critical to determine if mites are present and causing damage before management practices can be implemented. There are many different factors that can contribute to stunted growth during spring, including apical dominance, frost, cold damage, thrips, phylloxera, nutrient deficiency, compromised graft union, damaged roots and low carbohydrates in storage reserves.

Grape rust mites are very tiny, and at only 0.15 mm (1/100 inch), they are best viewed with magnification of 40X or greater. There are a number of time points at which to sample tissues: dormant wood, shoots in early spring, and leaves in summer. Because the pest is mobile and tissues are growing in early spring, trying to understand the link between mite presence, population density and tissue surface area can be somewhat inconclusive. In addition, mite populations can decrease after miticide or other pesticide applications.

Dormant wood samples can be collected to determine mite presence during winter. However, it may be very difficult to identify mites and to determine a density when searching buds or large areas of bark on cordons or canes. In addition, there is little data available to indicate economic threshold levels of the mites from in-season or dormant season monitoring. However, approximately 400-1000 mites per spur were found to cause significant damage to leaves and shoots in Australia (Bernard et al. 2005), based on sticky tape traps halting mite migration from overwintering sites in spring. To date, we do not have confirmed thresholds of mites per dormant wood or leaves to indicate damage for Oregon.

During summer, rust mites inhabit leaves and feed on cell contents. Significant mite populations on leaves can cause leaf discoloration (Figure 3) that is sometimes referred to as "bronzing." There is no critical threshold identified for mite presence per leaf in summer scouting. However, this can be an indicator of potentially high populations of mites moving into overwintering sites which may cause damage the following spring.

Managing rust mite populations

Vineyard managers and viticulturists of sites plagued by stunted growth and mite infestations question whether they can ever get rid of the problem. For some, they may not be aware of the problem in the vineyard due to the short time frame to easily see damage before the canopy develops further. While some of the most experienced vineyard managers can identify the symptoms shortly after bud-break, the time at which symptoms are most visible is at the stage where 8-9 leaves are unfolded (stage 18-19 based on the extended BBSCH scale). If the window is missed or symptoms are not very severe, the problem may remain undetected.

Mites present early in the growing season have been effectively managed in years past with sulfur applications at the delayed dormant (wooly bud) stage and another spray applied at bud-break (Bernard et al 2005, Walton et al. 2007b). However, this spray strategy is not effective if cool, rainy weather persists, making sprays ineffective. During these conditions of cool weather, growth of tissues is very slow, potentially leading to compounded effects of the mite feeding.

Mite presence later in the growing season may not cause visual symptoms unless canopy populations are high and conditions are conducive for leaf symptoms to appear. In summer, high infestations can lead to leaf bronzing, but these symptoms can be highly variable depending on the mite infestation density, temperatures and conditions of vine water stress. The first signs of infestation can start out as darker green leaves. The color then progresses to a blackish surface color, and in some (not all) cases leaves turn a brown-red color in late August and into September (Figure 3). During summer, the mites feed on the leaves and then make their way into overwintering sites (bud scales of canes and bark of vine heads, cordons and trunks) before the end of summer. Identifying if you have potentially damaging mite populations and finding the correct window to manage mite populations is tricky.

According to Dr. Vaughn Walton, Horticultural Entomologist at Oregon State University, "We are looking at the impact of mid-summer sprays but no reliable data have yet been obtained due to the difficulty finding optimal trial sites and the shifting nature of the issue."

Management of grape rust mites continues to be a dilemma in the variable springs of the Willamette Valley. We hope with further research efforts in Oregon and abroad, better approaches may be found. Particularly, Dr. Walton is interested in the potential for systemic controls to be employed for mite population control

without damage to vineyard predators.

Figure 3. Summer leaf discoloration caused by mite feeding may start out as a dark greenish-black (middle) and progress to a red-brown color (right) and are easily distinguished from normal, healthy leaves (left).

Other shoot stunting insects

Other insects can cause stunted growth of vines. The presence of thrips in high populations during bud-break and early in the season may cause stunted shoot development. Stunting of grape shoots due to thrips has been observed in other winegrape production regions such as California (Western Flower Thrips), Germany, Switzerland and France (Grape Thrips). Research in California indicated only 1.6 thrips/shoot tip caused significant stunting of the shoot (McNally et al. 1985). The nymphs and adults of thrips can be easily distinguished from rust mites by the size and shape.

The two different thrips species generally found in vineyards of the Pacific Northwest include both the Western Flower Thrips (*Frankliniella occidentalis*) and Grape Thrips (*Drepanothrips reuteri*). These pests are not controlled by sulfur sprays which are typically applied during the early season for rust mite management. Rather, thrips are more adequately managed early season with other products. For more information on thrips management, see the Pacific Northwest Insect Management Guide (http://uspest.org/pnw/insects?20SMFR07.dat).

Conclusions

Short shoot syndrome has been a perennial problem in some areas of the Willamette Valley. The extent of the mite damage and the short shoot growth response is likely linked to other factors in addition to mite feeding, such as frost damage, apical dominance and cold weather. Other insect feeding such as thrips also coincides with mite feeding, particularly after bud-break. Other complex site-associated factors have also been explored to explain the extent of mite infestation and damage. The high use of sulfur is postulated to cause decreases in predatory mites while the lack of sulfur use in a largely systemic fungicide program may lead to rust mite population increases in some vineyards. The best advice to growers in preventing and managing rust mite damages include scouting young vineyards for presence in the first few years, monitoring symptomatic blocks for mite presence in-season (leaves) and in dormancy (cuttings), and using appropriate management methods. All these efforts will better allow growers to assess the problem and assist researchers in identifying causal relationships.

For more information and questions regarding mite-related research in Oregon, contact the vineyard mite research team:

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Dr. Amy Dreves, Entomology, OSU Crop & Soils (amy.dreves@oregonstate.edu)

Further Reading

- Grapevine Growth Distortions – A Guide to Identifying Symptoms, OSU Extension, http://extension.oregonstate.edu/catalog/pdf/em/em8975-e.pdf
- Short Shoot Syndrome of Grapes in the Pacific Northwest. EM-8944-E, OSU Extension, http://extension.oregonstate.edu/catalog/pdf/em/em8944-e.pdf
- Western Flower Thrips Fact Sheet, http://uspest.org/pdf/reb114.pdf

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Zandigiacomo, P. and C. Frausin. 1998. Problemi entomologici del vivaismo viticolo in Friuli-Venezia Giulia. Informatore Fitopatologico 48:10–14.

OSU Wine and Grape Research and Extension Newsletter

May 2010 http://wine.oregonstate.edu

Powdery Mildew Spore Trapping Project Update

Dr. Walt Mahaffee, Research Plant Pathologist, USDA-ARS

A spore trapping and monitoring study has been employed in the Willamette Valley for several years to better understand when powdery mildew inoculum is moving in vineyards and when to start fungicide programs. This year, 31 traps were deployed in 12 vineyards in the Willamette Valley and are being sampled bi-weekly for presence of DNA from *E. necator* (Grape Powdery Mildew) using quantitative PCR.

At six sites, we are testing the potential of a producer-performed LAMP-PCR method to allow managers to monitor PM presence on their own. At these sites, there is one trap being monitored by the vineyard manager using LAMP-PCR and two traps are being monitored by our lab (USDA-ARS Foliar Pathology)—one processed for quantitative PCR and the other for LAMP-PCR. Currently, we are working with one vineyard cooperator to test protocols and function of LAMP-PCR before involving the other five sites. LAMP-PCR results from the first test site look very promising, and we should begin testing of LAMP-PCR with the other five managers within the next few weeks.

Another year of spore trapping data continues to support previous research results. Since 1997, we have seen indications that ascospore release occurs prior to bud break and is followed by a delay (temperature dependent) before we see any further signs of release events or spread. In the first week of April 2010, when buds were in the swell to wool stage, a couple of traps had positive detection for DNA of *E. necator*. However, there has not been a positive detection at the trial sites since then. At our research site (OSU Botany Farm), where vines are allowed to grow enough mildew to turn vines white and then black with cleistothecia, we had a positive detection on April 5 and another on May 3. The detection on May 3 was negative using quantitative PCR but positive using LAMP-PCR. The LAMP-PCR method is more sensitive than quantitative PCR, and the LAMP-PCR is likely approaching a detection limit of one spore or other particle carrying the DNA of *E. necator*. The quantitative PCR method used over the past three years has a practical detection limit between 10 and 100 spores. We have also made improvements in our trap design. With the new methods and traps, we are obtaining more sensitive information.

Since there was a positive detection in commercial vineyards after bud swell, you are probably wondering if this year's epidemic has started. I doubt that it has started in these vineyards. Successful initiation of the epidemic is dependent on more than just conditions for ascospore release. Ascospore release can occur with long wet periods when temperatures are below 50°F (several have occurred in spring 2010) but infection is limited under these conditions. Infection is limited by the wool found on buds and new tissues since they act as a physical barrier to spores landing on susceptible tissue. Also, leaf wetness is harmful to *E. necator* once ascospores have infected. Our research over the past 13 years indicates that sporulation during the current conditions does not occur. For the last three years, we have had flagshoots with no spread or airborne spores detected until daily low temperatures remained above 45°F. Similarly, David Gadoury's research group at Cornell showed that temperatures below 43°F kill parts of or entire colonies and extends the time period before sporulation occurs by several days. Therefore, I think it is unlikely that we have seen measureable disease development at this point. In addition, any early season sulfur applications for eriophyid mites (bud/rust) should also have activity on any powdery mildew present on the emerging shoots.

This project will continue through the 2010 growing season and into the future to better understand when powdery mildew is present and how to best design spray programs and timing around spore releases.

Spotted Wing Drosophila Control Strategies – Update

Dr. Paul Jepson, Professor & Director, OSU Integrated Plant Protection Center

The purpose of insecticide sprays applied against SWD is to protect the fruit that they attack during the period when it is susceptible to oviposition. For damage to occur, fruit needs to be in a susceptible stage and flies need to be present. Please consult http://swd.hort.oregonstate.edu/ for the latest information about management and control recommendations in individual commodities.

THERE IS NO JUSTIFICATION WHATSOEVER FOR TREATING CROPS THAT DO NOT HAVE SUSCEPTIBLE FRUIT PRESENT.

Many Oregon commodities are weeks away from this susceptible stage at the moment, some even in the pre-flowering stage. None of these commodities should be treated against this pest until fruit are present.

SWD may be detectable in monitoring traps that are placed in orchards and farms before susceptible fruit are present, as is the case with many insect pests. It is certainly worth continuing to monitor, but there is no justification for treatment simply because flies are found. The flies are mobile, they may exhibit so-called marauding behavior where populations move between locations over time, and the factors that attract them to particular fields and farms now may be absent by the time fruit is at a susceptible stage.

The goal of spraying is not to suppress populations prior to fruit set and this will not be an economic or effective strategy for anyone to undertake. In order to have any impact at all on fly populations over the scale of Oregon counties, spray application would have to be on a massive scale and combined with a number of other practices. On individual farms and fields, sprays before susceptible fruit are present will be wasted and will only incur unnecessary costs and potential risks to operators, farm workers, the environment and for the onset of pest resistance.

Foliar sprays are to be applied ONLY to the target commodity that is written on the pesticide label. Unless explicitly permitted, spray application to the ground or surrounding vegetation is illegal

and not permitted under any circumstances.

Finally, where susceptible fruit are present, and flies are detected in monitoring traps, there may be significant risks of damage to fruit and in that case, it is essential to be prepared and to follow the recommendations available from county extension offices, supported by up-to-date information on the SWD website: http://swd.hort.oregonstate.edu/.

2010 Vineyard Floor Management Survey

The use of vineyard floor management methods can have a significant impact on vineyard productivity and fruit quality by influencing water availability, nutrition and vine vigor, and the impacts of management are being studied by researchers at OSU and the USDA-ARS in the Viticulture and Enology Research Team. An industry-wide survey has been developed to identify the current vineyard management practices with regard to vineyard floor management. All industry members are encouraged to complete the survey online here. Results of this study will help identify current management practices and decision trends to help guide Extension and research efforts.

New viticulture diagnostic and learning modules online

Two online modules have been created by OSU Viticulture Extension to provide information to the winegrape industry. These modules are interactive and serve as online diagnostic tools to help understand and diagnose problems in the vineyard. Check out the modules online today by clicking the titles!

Understanding Grapevine Nutrition—This online module leads you through information on nutrient needs of the grapevines, symptoms of deficiency and/or toxicity, and provides information on vineyard nutrient management. A section of this module also provides information for diagnosing non-nutrient problems such as damage from diseases, insects, drought, sunburn, herbicides and more! Authors: Patty Skinkis, OSU and Paul Schreiner, USDA-ARS.

Preventing and Managing Herbicide Drift—This is an online module designed to provide information on grapevine sensitivity to certain herbicides and how to prevent damage and drift exposure. It is highly recommended that new grape growers read this information, but information is useful to all levels of industry. This module should be shared with other communities and industries surrounding vineyards to raise awareness of the grapevine sensitivity to herbicide drift and volatility. Author: Patty Skinkis, OSU.

Brettanomyces

James Osborne, PhD., OSU Extension Enologist

The most common and important spoilage yeasts encountered during winemaking are of the *Dekkera/Brettanomyces* genus. These yeasts cause serious economic losses worldwide in the wine industry and have therefore been the subject of recent intensive research efforts. *Brettanomyces* is the asexual, non-sporulating form while *Dekkera* is the sexual, sporulating form. The species *B. bruxellensis* is most commonly found in wine and most frequently identified in *Brettanomyces* spoiled wines. *Brettanomyces* was named because of its connection to the English brewing industry (British brewing fungus) and is still utilized in the production of lambic beers. The reason that *Brettanomyces* is such a problem in the wine industry is its ability to produce spoilage products that can give wine a distinct aroma described as wet dog, horsey, or barnyard (to name a few). Understanding how and when *Brettanomyces* causes wine spoilage as well as appropriate preventative and corrective actions can help you minimize or prevent *Brettanomyces* issues in your winery.

Brettanomyces has been isolated from wineries around the world and the safest approach to preventing spoilage is to assume that you have some level of *Brettanomyces* in your winery already. From this basis you can actively take steps to minimize or prevent the growth of *Brettanomyces* and greatly reduce the risks of spoilage. So how is *Brettanomyces* getting into your winery? Until recently, it was thought that *Brettanomyces* was not present on the surface of wine grapes and that contamination occurred in the winery through importation of spoiled wine, poor sanitation of hoses, tanks, and barrels, and contamination by fruit flies. However, a recent study utilizing specific enrichment medium reported that *B. bruxellensis* can be present on wine grapes but usually in very low numbers. Still, the most frequently cited place where *Brettanomyces* is found in the winery is wood cooperage. *Brettanomyces* is not usually found in brand new barrels as the toasting process is an effective sanitizer. However, if infected wine is placed in these barrels, *Brettanomyces* can quickly become established and has been found up to 8 mm deep in staves. This is not to say that *Brettanomyces* is only found in barrels. If conditions are conducive, *Brettanomyces* will happily grow in wine stored in tanks.

The major spoilage problem associated with *Brettanomyces* is the production of the volatile ethylphenols 4-ethylphenol (4-EP) and 4-ethylguaiacol (4-EG). In addition to producing volatile ethylphenols, *Brettanomyces* can produce large amounts of acetic acid when growing on glucose and can also produce isovaleric acid, a compound described as 'rancid'or 'vomit'. Depending on the concentrations of these various compounds, numerous descriptors have been used to describe *Brettanomyces* character. Elevated levels of 4-ethylphenol in red wine are associated with aromas described as 'horsey', 'smoky', 'medicinal', or 'leather' while 4-ethylguiacol has been described as 'clove' or 'spice'. At low concentrations many winemakers consider *Brettanomyces* character a positive attribute and one that adds complexity to a wine. However, we do not fully understand why *Brettanomyces* spoilage in one wine may result in subtle aromas while in another overpowering barnyard and Band-Aid smells are produced. Part of this may be due to the difference sensory thresholds of compounds in wine. Although the reported sensory threshold of 4-EP and 4-EG in red wine (check order for thresholds) is 605 µg/L and 110 µg/L respectively, the concentration at which these compounds become objectionable in wine can vary greatly. This variation is due primarily to the type of wine and the relative concentrations of the two compounds. For example, the detection threshold of 4-EP in a Tempranillo wine is reported

to be much lower than a Cabernet Sauvignon wine. In addition, researchers have noted that the sensory threshold of 4-EP is lower when both 4-EP and 4-EG are present in the wine together. There is also an increasing amount of evidence that strain differences may be responsible for much of the sensory variation reported. For example, studies in California and Australia have reported large differences between the amount of 4-EP and 4-EG produced by different strains of *Brettanomyces*. Aside from the production of objectionable spoilage products, *Brettanomyces* infection can also result in the loss of varietal and fruity/floral aromas, a loss of color, and an increase in bitterness.

So what as a winemaker can you do to control *Brettanomyces* in the winery? The first thing is to understand what conditions or parameters stimulate *Brettanomyces* and use this knowledge to prevent or control them. Although the vineyard does not appear to be a serious contamination source, damaged grapes may contain higher microbial loads than healthy grapes and the removal of damaged grapes should be considered along with SO2 use. In addition, long maceration periods have been demonstrated to encourage *Brettanomyces*, perhaps due to increased amounts of substrates and pre-cursors (hydroxycinnamic acids) for Bretty off flavors. During alcoholic fermentation *Brettanomyces* populations usually remain low as they are poor competitors for nutrients with *Saccharomyces cerevisiae*. However, if you have a problematic or stuck fermentation, *Brettanomyces* can grow to high numbers and cause spoilage at this stage. This is also true during the malolactic fermentation (MLF) and one reason ensuring a quick and problem free MLF will reduce the risks of *Brettanomyces* spoilage.

Compared to most other wine microorganisms, *Brettanomyces* is very slow growing and is usually only detected in significant numbers during the aging or storage of a wine. However, significant variations in growth rates and population changes have been noted amongst strains. Very low levels of sugars (glucose, fructose, galactose, and trehalose) are required for its growth. Researchers report that as little as 275 mg/L of sugar was sufficient to support the growth of the yeast and cause spoilage of wine. In addition, *Brettanomyces* can use ethanol as a sole carbon and energy source. *Brettanomyces* also produces a B-glucosidase enzyme that enables it to utilize cellobiose, a disaccharide present in wood barrels. New barrels have much higher levels of this wood sugar than older barrels, one reason why *Brettanomyces* infections in new barrels can result in more intense spoilage. In addition, new barrels can bind more free SO_2 than older barrels. Because of its ability to utilize these alternative carbon sources, *Brettanomyces* is capable of surviving and causing spoilage even in wines that are considered dry, although ensuring your ferments complete to dryness will help minimize the risk of *Brettanomyces* spoilage. Furthermore, nutrient additions during fermentation should be appropriate as excess nutrients may result in nutrients being available for *Brettanomyces*.

Some common ways to control *Brettanomyces* growth are maintaining proper pH, SO_2, and temperature during the wine aging process. Like most wine spoilage microbes *Brettanomyces* prefers higher pHs and so a pH below 3.50 is recommended to discourage their growth. pH also impacts the effectiveness of SO_2 with SO_2 being much more potent at lower pH values. The sensitivity of *Brettanomyces* to SO_2 seems to be strain dependent but in general this antimicrobial is effective at controlling growth. An additional tool for the control of *Brettanomyces* is the compound dimethyldicarbonate (DMDC) sold under the trade name Velcorin™. This has shown to be very effective against *Brettanomyces* and is usually used close to bottling. Care needs to be taken when using this compound and a special dosage unit is necessary for its application. Maintaining wine at lower temperatures will also help minimize growth of *Brettanomyces*. Temperatures below 55°F are recommended. Some factors that can encourage *Brettanomyces* include delayed racking and oxygen. Oxidative conditions encourage *Brettanomyces* growth and excessive amounts of oxygen have been shown to enhance the production of acetic acid while racking as well as fining have been shown to reduce *Brettanomyces* populations. *Brettanomyces* is spread around the winery through poor sanitation practices. Topping wines should always be confirmed free of *Brettanomyces* and sampling thieves should be rinsed in ethanol between barrel sampling to prevent cross-contamination.

In addition to these preventative measures, a regular monitoring protocol should be in place so at the earliest possible time you detect when you have a problem and can take appropriate action. There are many different tools a winemaker has access to when it comes to monitoring for *Brettanomyces* infection. The most common tool is his or her nose. Sensory detection of *Brettanomyces* taint may take some training if you are not familiar with the aromas. However, just relying on sensory detection means that by the time you can smell a problem the wine is probably already spoiled. Therefore, you should employ other methodologies that will allow you to catch the problem early enough to prevent spoilage. The most common method to detect and identify *Brettanomyces* is the plating of a wine sample on a media containing the fungicide cycloheximide. *Brettanomyces* is resistant to cycloheximide and so if colonies grow on these plates it is most likely *Brettanomyces*. Inspection of the cells using a microscope can help confirm that the colony is *Brettanomyces* although microscopic determination of *Brettanomyces* can be difficult given that *Brettanomyces* can have much different morphology (shapes). It is generally described as a boat shaped cell, but during growth in wine it often appears as other shapes. Plating allows for identification and estimation of population but it does not tell you the level of 4-EP that the yeast has produced. Quantification of 4-EP requires more complex analysis and is usually performed by an outside lab. Monitoring of both viable cells (plating) and 4-EP levels on a regular basis can give you a good understanding of the level of *Brettanomyces* infection and the amount of spoilage product produced.

One problem with the plating method is that it only detects viable cells. Some microorganisms can enter a state called viable but not culturable (VBNC) where microorganisms do not grow on conventional microbiological medium but still remain intact and viable. At a later stage when growth conditions are favourable these microorganisms may begin growing again. This state is usually brought about in response to stresses such as temperature, oxygen concentration, and exposure to antimicrobial agents such as SO_2. It is now known that *Brettanomyces* can enter this state. In order to detect *Brettanomyces* in a VBNC state you need to use genetic based detection methods that utilize molecular biology techniques such as PCR. One of the most common based detection methods is the Scorpion® probe system although others are now becoming

available. This is a very specific test for *Brettanomyces* but is more expensive then many of the other methods. Each of these detection methods has their advantages and disadvantages. A combination of these methods works best to give you the most complete overall picture. However, the most important step is to ensure you have a plan of action in which you determine when and how you will sample and analyze wines for *Brettanomyces*. When developing this plan, decide what is the appropriate monitoring and analysis method based on your budget and needs. Careful sampling is crucial. For example, *Brettanomyces* tends to be found near the bottom of barrels and so sampling from the top of the barrel may not give an accurate analysis of the *Brettanomyces* population. Once you have decided when and how to sample you need to determine what actions you will take to control or prevent spoilage and verify that these actions are effective. A simplified example of a *Brettanomyces* control plan is included with this article. At each step of the winemaking process examples of what to monitor as well as critical limits, corrective actions, and verification steps are given. This is by no means comprehensive but should be a good framework for you to develop your own winery specific plans. Please contact me if you have any further questions about this topic: james.osborne@oregonstate.edu

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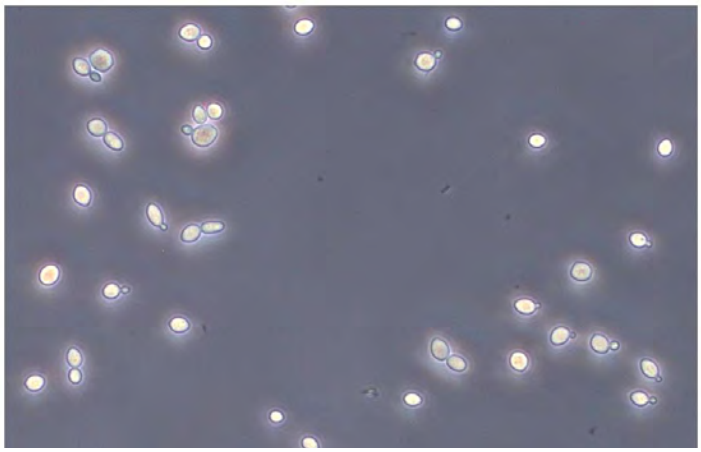

Brettanomyces (x400) (Also see flow chart at end of newsletter.)

New Viticulture & Enology Scientific Research Articles

Viticulture and Enology research is conducted for Oregon and the Pacific Northwest by Oregon State University (OSU) and the United States Department of Agriculture's Agriculture Research Service (USDA-ARS). The most recent research articles are outlined below by unit. Unit authors are indicated in bold text. Contact the journal and/or the author to obtain these articles for further reading.

USDA-ARS Horticulture Crops Research Unit

- **Lee, J.** 2010. Degradation kinetics of grape skin and seed proanthocyanidins in a model wine system. Food Chem. In press. DOI: 10.1016/j.foodchem.2010.03.126
- **Lee, J.** and Martin, R.R. 2010. Analysis of grape polyamines from grapevine leafroll associated viruses (GLRaV-2 and -3) infected vines. Food Chem. In press. DOI: 10.1016/j.foodchem.2010.03.118
- **Lee, J.**, Keller, K.E., Rennaker, C., and Martin, R.R. 2009. Influence of grapevine leafroll associated viruses (GLRaV-2 and -3) on the fruit composition of Oregon *Vitis vinifera* L. cv. Pinot noir: free amino acids, sugars, and organic acids. Food Chem. 117:99-105.
- **Lee, J.** and Martin, R.R. 2009. Influence of grapevine leafroll associated viruses (GLRaV-2 and -3) on the fruit composition of Oregon *Vitis vinifera* L. cv. Pinot noir: phenolics. Food Chem. 112:889-896.
- **Lee, J.** and **R.P. Schreiner**. 2010. Free amino acid profiles from 'Pinot noir' grapes influenced by N-status and sample preparation method. Food Chem. 119: 484-489.
- Alabi, O.J., **Martin, R.R.** and Naidu, R.A. 2009. Sequence diversity, population genetics and potential recombination events in *Rupestris stem pitting-associated virus* in Pacific Northwest Vineyards. J. Gen. Virol. doi:10.1099/vir.0.014423-0
- Mekuria, T., Gutha, L.R., **Martin, R.R.** and Naidu, R.A. 2009. Genome diversity and intra- and inter-species recombination events in *Grapevine fanleaf virus*. Phytopathology 99:1394-1402.
- Mekuria, T.A., Karasev, A.V., **Martin, R.R.** and Naidu, R.A. 2009. First report of *Grapevine leafroll-associated virus-3* in wine grape cultivars in Idaho. Plant Dis. 93:1218.
- **Schreiner, R.P.** and K.L. Mihara. 2009. The diversity of arbuscular mycorrhizal fungi amplified from grapevine roots (*Vitis vinifera* L.) in Oregon vineyards is seasonally stable and influenced by soil and vine age. Mycologia 101(5): 599-611.
- **Schreiner, R.P.** 2010. Foliar sprays containing phosphorus (P) have minimal impact on 'Pinot noir' growth and P status, mychorrhizal colonization, and fruit quality. HortScience 45: 815-821.
- Blom, P.E. and **J.M. Tarara**. 2009. Trellis tension monitoring improves yield estimation in vineyards. HortScience 44:678-685.
- **Tarara, J.M.** 2009. Estimating high rates of transpiration

in woody vines with the heat-balance method. [7th International Workshop on Sap Flow]. Acta Hort. 846:193-200.

- **Tarara, J.M.**, P.E. Blom, B. Shafii, W. J. Price, and M. Olmstead. 2009. Modeling seasonal dynamics of canopy and fruit growth in grapevine for application in trellis tension monitoring. HortScience 44:334-340.

Oregon State University

- **Qian, M.C.**, Y. Fang, and K. Shellie. 2009. Volatile composition of Merlot wine from different vine water status, J. Agric. Food Chem. 57: 7459–7463.
- Rowe, J.D, Harbertson, J.F., **Osborne, J.P.**, Freitag, M., Lim, J., **Bakalinsky, A.T.** 2010 Systematic identification of yeast proteins extracted into model wine during aging on the yeast lees. J. Ag. Food Chem. 58:2337-2346.
- **Skinkis, P.A.**, B.P. Bordelon, E.M. Butz. 2010. Effects of Sunlight Exposure on Berry and Wine Monoterpenes and Sensory Characteristics of 'Traminette.' Am. J. Enol. Vitic. 61:147-156.
- Sweetman, C., **L.G. Deluc**, G.R. Cramer, C.M. Ford, K.L. Soole. 2009. Regulation of malate metabolism in grape berry and other developing fruits. Phytochem. 70 (11-12):1329-1344.

OSU Alumni Spotlight: Brett Weis

Since graduating with a MS in Enology & Viticulture from the OSU Department of Food Science & Technology 2008, Brett has

been enjoying life in the Napa Valley wine industry. He currently works as Assistant Vineyard Manager for Antica Napa Valley, owned by the famous Antinori Family whose Italian vineyards and winemaking date back to 1385. He farms about 400 acres of Cabernet, Merlot, Pinot noir, and many other cultivars.

Brett says that the reason he chose this job was that "it provides a unique viticulture experience, as we are located in… the Foss Valley at an elevation of 1500 ft [that] is planted to nine different varieties on eight different soil types." Brett chose to study at OSU because of the small, intimate program that would allow him to work closely with faculty and local vineyards and wineries. In fact, he worked for Tyee Wine Cellars in Corvallis while completing his graduate research thesis focusing on the effect of screw caps on Pinot noir wine quality. The graduate program at OSU helped Brett get the experience he would need to land his dream job, but his upbringing around family already in the industry first caught his interest. "I really enjoyed the lifestyle it provided them. Great food and wine, beautiful country, nice people, and the opportunity to work outside..." Even before starting grad school, Brett travelled to Chile to work harvest and toured several Argentinean wineries while in the area.

Brett plans to continue working for Antica Napa Valley while learning Italian, traveling, and making great wine with his family and friends.

For more information on the OSU Viticulture & Enology Degree Program options, visit http://wine.oregonstate.edu/programs.

Upcoming Events

Umpqua Valley Grape Day
June 10, 2010, Roseberg, OR
This event will feature various sessions in the vineyard and winery. Topics will include: Spotted-Wing Drosophila Fruit Fly Update, Increasing Biodiversity in the Vineyard, Marketing Ideas to Improve your Bottom Line, Moving to Higher Density Vineyards, Sensory Evaluation of Wine Flaws, a wine social, dinner and music. Southwestern Oregon grape growers and winemakers are encouraged to attend. See the following link for more information: http://wine.oregonstate.edu/files/files/GRAPE%20DAY.pdf.

7th International Cool Climate Symposium
June 20-22, 2010, Seattle, WA
This international conference on enology and viticulture research is back in the Pacific Northwest in June! We encourage Oregon industry members to attend, as this symposium had its beginning in Oregon in 1984 and was first organized by OSU and the Oregon Wine Advisory Board! Since then, this symposium has been hosted in New Zealand, Germany, New York, and Australia. For more information about the event, visit: http://asev.org/annual-meeting-2010/.

American Society for Enology & Viticulture Annual Conference
June 23-24, 2010, Seattle, WA
The annual conference will be held immediately following the International Cool Climate Symposium in June. A number of great seminar sessions, including an industry-based seminar series is provided. For more information about this event, visit: http://asev.org/national-conference-2010/

BRETTANOMYCES QUALITY CONTROL FLOW CHART

	MONITORING/ ANALYSIS	CRITICAL LIMITS	CORRECTIVE ACTION	VERIFICATION/ DOCUMENTATION
Grapes	pH Fruit condition	< 3.50 Visible rot, off odors	Acid addition Sorting and increase SO_2	Re-check pH after cold soak
Crush Destem Maceration	SO_2 Temperature YAN	30-50 mg/l total SO_2 5-10°C, 40-50°F 140-250 mg/L	Add SO_2 Chill Nutrient addition only if needed	Monitor SO_2 levels Monitor temperature
Alcoholic Fermentation	Residual sugar	< 0.5 g/L	Ensure complete fermentation	Measure R.S
Malolactic Fermentation	Microscopic and sensory examination PCR analysis Temperature	non-Saccharomyces yeast Barnyard/medicinal aroma 15-20°C, 60-68°F	Take steps to ensure successful MLF Nutrients only if needed Adjust temp	Malic acid Microscopic examination PCR analysis Monitor temperature Sensory
Post Fermentation & Aging	Microscopic and sensory examination SO_2 New barrels Plating PCR analysis Temperature 4-EP, 4-EG Oxygen	Barnyard/medicinal aroma non-Saccharomyces yeast 20-30 mg/l free SO_2 Colonies on WLC media 13-16°C, 55-60°F Increase over baseline < 0.25 mg/L	Clarification, fining, pad filtration Add SO_2 Barrel management and sanitation Adjust temp Reverse osmosis Minimize O_2 pick-up	Microscope examination Monitor SO_2 Plating PCR analysis Monitor temperature Monitor 4-EP, 4-EG Monitor O_2 Sensory
Clarification Stabilization Filtration	SO_2 Oxygen	20-30 mg/l free < 0.25 mg/L	Adjust SO_2 Fining, polish filtration Velcorin (DMDC) Minimize O_2 pick-up	Monitor SO_2 PCR analysis Monitor O_2 Sensory
Bottling	Plating PCR analysis SO_2	20-30 mg/l free	Sterile filtration Velcorin (DMDC) Adjust SO_2	Plating PCR analysis SO_2 analysis