

Identification & Control of Brettanomyces in the Vineyard and Winery

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A high percentage of wines, mainly reds, are contaminated with volatile phenols. These compounds, principally from from hydroxycinnamic acid metabolism by *B. bruxellensis* are responsible for “Brett Taint” odor and degrade the quality of the wines affected. The detection thresholds of these compounds are very low, the order of 440 mg / L for 4-ethylphenol, and 620 mg / Liter for sum of 4-ethyl Guayacol and 4-ethylphenol (“Band-Aid”).

Techniques used in the making of great red wines, often create favorable conditions for the development of specific spoilage yeast contaminants and the occurrence of phenolic compounds. There are no methods to fully correct this defect once appeared.

Winemakers need to develop strategies for their control at all times of the wines, especially red wines for aging.

In the Vineyard

The presence of *Brettanomyces* SPP. in the vineyard is a fact rarely evaluated by the winemakers as a potential problem. However, it is a known fact that naturally on grapes there is a significant flora; often called wild or native, within which there are different strains of yeast, often including spoilage yeasts. The most significant impact on the quality of red wine comes from *Brettanomyces*. It is estimated that as much as 50% of incoming grapes contain some level of *Brettanomyces* SPP. (Primarily *Brettanomyces bruxellensis*). The unique survival niche associated with Brett results in an apparently random contamination; likely related to its sensitivity in some environments and ability to thrive in the “challenging” environment post fermentation.

An effective control strategy for *Brettanomyces* must include an evaluation of the population levels on the grapes and the different conditions that affect each particular vineyard.

Several factors can increase the levels of these yeasts, excessive cars traffic, wind, earth movement, clusters too close to ground level and grapes removed and left to themselves can increase the level of *Brettanomyces* in clusters.

Control of this risk requires determining the level of contamination of the grapes. Lonvaud-Funat Renouf and published in the Journal Microbiological Research (2007) a paper entitled “Development of an Enrichment medium to detect *Dekkera / Brettanomyces bruxellensis*, a spoilage wine yeast, on the surface of grape berries.” This publication describes the EBB medium formulation and validation. Today the media is available in the market under the trade name *Berry Easy*® (Lebrun Labs LLC, Anaheim, CA) which allows semi-quantitatively determine population levels.



The barbera grapes were located at the inner edge of a very busy road, the Zinfandel in an isolated area. *Berry Easy*® was used and then incubated with *Easy Blue*®.

Grape Harvest and Transport

Harvesting and transport of grapes can increase the content of Brett on the grapes if there is breakage and too much time elapses from the harvest to the mill itself, which can be aggravated if the grapes are exposed to high temperatures during transport.

It is evident that the use of harvesters can increase the popu-

lation of wild yeast if not done with adequate sulfitation. Knowledge of the Brett population levels on the grapes and its evolution during harvest operations and transportation is vital to avoid fermentations that are contaminated with *Brettanomyces SPP*.

Some new investigations being carried in Spain seems to show that leaves contamination can increase *Brettanomyces* population in must.

Maceration and Fermentation

The current, fashionable trend in the development of high-end red wines requires the total phenolic maturity of skins and seeds which normally reach very high sugar content. The musts resulting from these grapes can present fermentation difficulty.

Meanwhile some vineyards, and some vintages, grapes produce very few YAN levels found in some cases, levels below 50 ppm, which requires an adequate addition of complex nutrients to ferment properly.

In addition, the amount of SO₂ used during fermentation is becoming reduced every year.

Depending on the combinations of yeast strain and if there is inadequate nutrients, it becomes likely that there can be a reduction in fermentation kinetics during the last third. In this

situation it is possible that the steps taken can oxygenate the must-wine-and the erroneous (at this moment) addition of ammonium phosphate.

This can lead to an uncontrolled growth or “bloom” of *Brettanomyces*, which comes to wine from the vineyard or is the result of chronic contamination of the winery. It is therefore important to know the Brett population level in musts at the time immediately prior to planting of commercial yeasts, which can be done simply by *Easy Sniff*[®] (Lebrun Labs LLC, Anaheim, CA).

Keys that assure proper development of the alcoholic fermentation include:

- Proper sulphite levels in the must, depending on the grapes sanitary level and pH
- In case of high Bx and low YAN, selection of yeast with a low requirement for nitrogen and capable of tolerating high concentrations of alcohol.
- Addition of complex nutrients, instead of ammonium phosphate.

If the above are achieved, the development (“bloom”) of *Brettanomyces* will be controlled/eliminated in the final stages of fermentation. Strong strains of *Saccharomyces* can also help control development (“bloom”) of Brett.

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Malolactic Fermentation

The time that elapses between the end of alcoholic fermentation and the beginning of the Malolactic is also a critical period.

As the wine is not yet stabilized there is a risk of aromatic deviations. The traditional method of inoculating the wine with bacteria or permitting the spontaneous onset of MLF may be the perfect opportunity for *Brettanomyces* to proliferate and produce volatile phenolic compounds.

Co-treatment with sulphite at the right time can be of great in reducing this risk; by minimizing the time during which the wine is unprotected and vulnerable.

Conservation

Undoubtedly, the time during when most *Brettanomyces* blooms occur is during holding in tanks and especially in barrels.

At the stage of holding wine in tanks, SO₂ is an effective tool, although some strains of Brett are highly resistant. The pH of the wine affects the activity of SO₂, making it less efficient as the pH increases.

One characteristic of these yeasts is increased levels at the bottom of the tanks. Therefore racking is a way to prevent the growth of the population of Brett.

In case of samples are removed for testing and determining

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Brett concentration, do not forget that it is absolutely necessary to sample from the bottom or mix the contents prior to sampling. An easy low cost test is the *Easy Blue Brett test*TM (Lebrun Labs LLC, Anaheim, CA)

Because of aging and the “niche”, barrels are the critical point of Brett contamination, and one must pay special attention to cleaning and decontamination of barrels, assuming it is impossible to complete sterilization Brett Decontamination should be performed immediately after each transfer and before filling. The washing temperature must be maintained at a lower temperature, longer application. Never below 60 ° C. The steam is more efficient but affects the quality of the barrels and it is advisable to reserve it for cases of suspected contamination.


The high pressure cleaning (80-110 bars) eliminates almost all Brett from tanks, provided the equipment uses rotating heads adapted to the geometry of the barrel and enough time (10-20 minutes). Washing should not be so long as to affect the integrity of the wood. The use of cleaning products, it is also advisable to reserve it for suspicious barrels.

Sulfur should be applied to wood for approximately five days. You must repeat the operation when the barrel is dry. The barrels can be evaluated using suspicious *Test for Brettanomyces in and on Oak* (Lebrun Labs LLC, Anaheim, CA)

Easy Blue[®] (Lebrun Labs LLC, Anaheim, CA) can routinely


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check the population of *Brettanomyces* in wine. A population greater than 100 yeasts per ml is unsafe and indicates a risk of potentially disastrous bloom (where levels can reach a million cells/ml within weeks and phenolic compounds can overcome the wine). In the event that a positive detection of *Brettanomyces*, one must initiate corrective measures before a bloom occurs.

Corrective measures must be done when the wine still shows no sensory disturbance and volatile phenol content is less than 400 mg/liter, the priority is to stop the development of *Brettanomyces* and therefore the increased concentration of volatile phenols before they can be detected by the human nose. One approach is to increase free sulfur levels in sufficient quantities and in accordance with the pH above 0.8 ppm SO₂ molecules. It is also advisable to perfectly limit the lot altered, avoiding mixtures and the use of barrels that have had contact with contaminated wine. With respect to these barrels, intensive and thorough cleaning and disinfection programs should be conducted: these must be especially energetic and carefull; or in many cases, the barrels should be discarded.

Bottling

The increasingly common practice of unfiltered wine packaging increases the risk of development of yeast after bottling, and the consequent appearance of volatile phenols that will ruin the product. In these cases the use of DMDC is a very effective tool to control all the yeast contaminating the product.

In addition, sterile filtration performed correctly handles all possible development of yeasts in bottled wines. Sterile filtered wines should be checked for microbial levels using either an Easy Yeast and Mold Test™ (Lebrun Labs LLC, Anaheim, CA) or Easy WL Total Microbial Counts Test (Lebrun Labs LLC, Anaheim, CA). This will ensure that not only any Brett has been filtered out but also that any filtration was effective and metabolic activity will not result in pressure that can pop the cork.

Conclusion

Brettanomyces-related problems do not result from a single cause. The wine is vulnerable from birth to glass to the destructive effects of these yeasts. Understanding the different ways and times that the wine is more vulnerable to contamination together with the availability of new tools for detection and quantification makes it possible to develop control strategies.

Winemakers have the task of analyzing which risks are important and what actions should be taken to solve related problems.

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