Enology Notes # 143 **September 9, 2008**

To: Winemakers and Perspective Winemakers

From: Bruce Zoecklein, Professor and Head, Enology-Grape Chemistry Group

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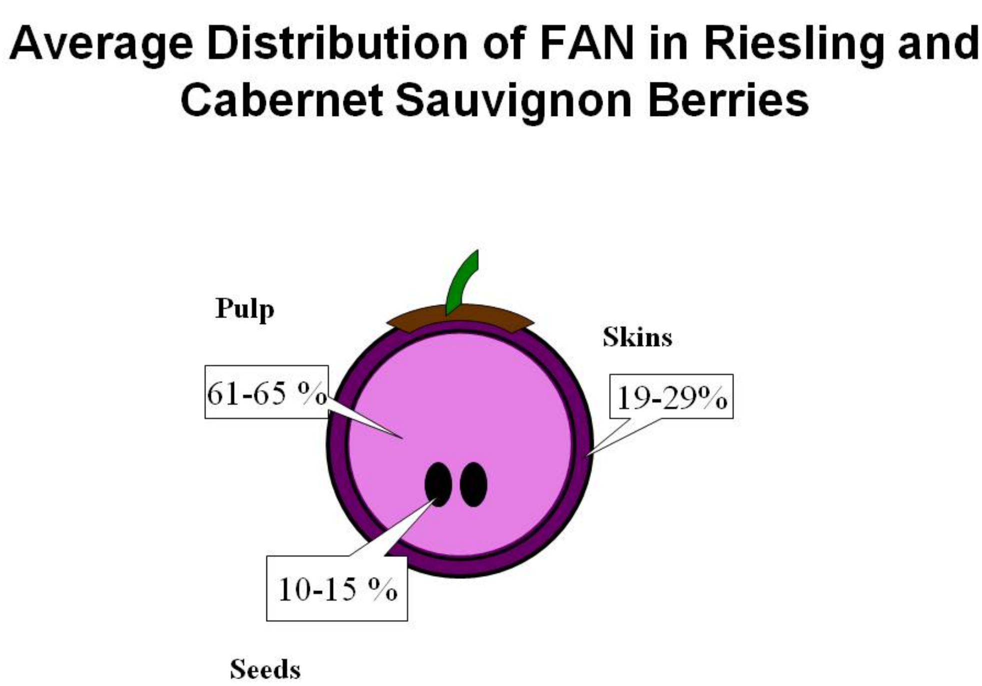
1. *Enology Notes* Index. All past *Enology Notes* are posted on-line. An extensive subject index is available at www.vtwines.info. Click Enology Notes Subject Index.

2. Measuring YAN, NH_3 and Arginine. The Enology Service Laboratory is offering pre-harvest and harvest fermentable nitrogen analysis again for the 2008 season. The sample submission procedure is as follows:

1. Email the laboratory at Enology.Services@vt.edu to request processing bags and/or bottles (fruit analysis vs. juice).
2. Include name, company, mailing address, and the number of sampling kits required.
3. Collect juice samples at harvest, or grape berry samples from the vineyard pre-harvest. Representative vineyard samples can be collected up to 5 days pre-harvest.
4. Fill sample bottles (if for juice samples) to the indicated level, and mix thoroughly to dissolve preservative.
5. Ship samples next day/early morning delivery to the Enology Service Laboratory (shipping delays impact analysis results).
6. Results will be posted on client's secure website within 24 hours.

Fermentable Nitrogen Analysis is $25. Juice panels (Brix, nitrogen, malic acid, pH, and TA) are $60 per sample. For additional information on other available analyses and analytical panels, please see the Enology-Grape Chemistry website at www.vtwines.info. As the figure below demonstrates, different parts of the fruit contain different concentrations of FAN amino acids. Make sure you process samples to reflect your winery processing methodology.



a. Measuring YAN at the Winery. While fermentable nitrogen is but one important yeast nutritional component, it is perhaps the easiest for winemakers to measure. The easiest way for most wineries to measure fruit nitrogen (which is different from other plant tissue or soil N) in-house is to conduct a Formol titration on the juice (see On-Line Publications at www.vtwines.info). In a study conducted several years ago, we demonstrated the ease and viability of this assay for determining fruit nitrogen status (Gump et al. 2002).

3. New Virginia Tech Enology Service Lab Offering: Sanitation Monitoring. The Virginia Tech Enology Service Laboratory has added sanitation monitoring to the list of available analyses. The purpose of this service is to allow winemakers to monitor the effectiveness of their sanitation procedures on tanks, barrels, winery equipment surfaces, bottling lines, etc. Such monitoring should be part of the winery HACCP procedures (see previous editions of *Enology Notes*). The laboratory will provide the necessary materials which include environmental samplers and the directions for their use. Surfaces are swabbed with the samplers provided and sent back to the lab. At the laboratory, the swabs are cultured and a determination of the concentration of enologically-important organisms is made, as well as the effectiveness of the overall sanitation procedure used. Request forms and monitoring details are available at www.vtwines.info.

4. Asynchronous Ripening. Variation is an inherent part of biological systems. Variation in the vineyard occurs among berries, in bunches and vines. A crop with asynchronous clusters or berries has a mixture of developmental stages, resulting in berries with optimal qualities diluted by berries which may be inferior.

Vines that produce the best quality wines are usually believed to be those with less variability. The lower the vineyard uniformity, the greater the negative influence on the resultant wine, and the greater the vineyard sample volume needed to get an accurate assessment of maturity (see On-Line Publication: Maturity Evaluation for Growers).

Vine-to-vine variability of morphologically-uniform vines, expressed as % coefficient of variation (standard deviation expressed as a percentage) was reported by Gray (2006), indicating the inherent nature of vineyard variability.

- Brix 4-5%
- pH 3-4%
- TA 10-12%
- Berry weight 6-20%
- Color 13-18%

a. Berry Variation. Although variation between berries is poorly understood, it potentially has the greatest impact on grape and wine quality. A typical berry follows a double sigmoid growth curve during its post-flowering development, but two berries on the same cluster may follow quite different paths. The divergence of the growth curves becomes apparent shortly after flowering, and the timing of this divergence is responsible for the extent of the difference between the two berries at harvest. The variables that contribute to variation between berries include berry size, berry composition, seed number, seed size, and berry position (Gray 2006).

5. New On-Line Publications: The following have been posted on the Enology-Grape Chemistry Group website. Click On-Line Publications.

- **Electronic Nose Evaluation of Cabernet Sauvignon Grape Maturity**
- **Electronic Nose Discrimination of Wines Produced from Ethanol Sprayed Vines**
- **Sulfur-Like Off Odors, A Winemaker HACCP Plan**

6. Herbaceous Character in Red Wines – A Review. The following is a review from previous *Enology Notes*. As a result of the rains in some areas this season, it is likely that some of our fruit may have excessive herbaceousness. This can result in wines with a reduction in fruit intensity, and detrimentally impact palate structure and texture.

This sensory feature is mainly derived from a group of nitrogen-containing compounds, pyrazines, which are present in green plant tissues, including grapes. One important methoxypyrazine, IBMP (2-methoxy-3-isobutylpyrazine), imparts a vegetal aroma at relatively low concentrations in the fruit, ranging from zero to 35 ng/L.

Several factors have confounded our understanding of methoxypyrazines, including their very low concentrations (and, therefore, difficulty in quantification) and their association with other compounds.

A concentration of 1 ng/L is 1 part per trillion. Allen (2006) put this in perspective in a presentation outlining the difficulty in monitoring methoxypyrazines: if the earth's population is 6,493,359,729, then measuring IBMP would be roughly equivalent to measuring one part in 154 earth populations.

An additional confounding factor is that herbaceousness can be caused by methoxypyrazines, certain monomeric phenols, and polyunsaturated fatty acid derivatives. These include compounds such as hexanal and hexenal, which we have measured in relatively high concentrations in some regional wines.

IBMP (2-methoxy-3-isobutylpyrazine) imparts a vegetal aroma to Cabernet Sauvignon, Cabernet franc, and Sauvignon blanc, described as bell- or green pepper-like. The detection level of IBMP is 2 ng/L in water, and about 15 ng/L in red wines. IBMP may contribute to leafy-type aromas, even in concentrations as low as 2 ng/L (Allen

2006). Too much of this character disbalances the wine and results in the overall loss of complexity. This is a notable problem with some Cabernet franc wines produced in our region.

In addition to the problems of low concentrations and association with other compounds, odor synergism and antagonism also confound our understanding of methoxypyrazines in a matrix as complex as that of wine. Sulfur-containing compounds can complement the vegetative odor, and some have green-type odors themselves. This is why the oxidation of some thiols to disulfides, which occurs with splash racking and/or microoxygenation, can change the perception of herbaceousness.

Methoxypyrazines not only contribute to odor, but also impact palate balance (see Enology Notes #94).

a. Viticultural Factors Influencing Herbaceousness. There are two pathways to production of methoxypyrazines, one dependent on grape maturity, climate, and fruit exposure, but the other, not (Allen 1998). Important viticultural factors influencing wine herbaceousness include the following:

- Vegetative growth
- Soil moisture
- Leaf maturity
- Fruit exposure to light
- Crop load and rate of fruit maturation
- Uneven fruit ripening

In the fruit, the major methoxypyrazine (IBMP) is formed early and breaks down following véraison. The level in ripe fruit is related to the prevailing weather conditions, which lead to the initial IBMP concentration. The breakdown is initially very rapid, then slows as fruit maturity increases (Roujou de Boubee 2004).

Does this drop represent photo-degradation? It is not likely. It appears that the decrease in pyrazines is the result of temperature (Allen 2006). The decrease in concentration is directly correlated to the decrease in malic acid. Malic acid decreases at a faster rate during warm nighttime temperatures, as do methoxypyrazines, like IBMP.

Photosynthesizing green leaves, and the conditions that promote the persistence of odor-active compounds (such as high soil moisture), contribute to harsh green aromas/flavors in the fruit. As such, several have reported a correlation between leaf maturity (progression towards colors expected at senescence) and reduction in berry green-fruit character (Delteil 2003, Roujou de Boubee 2004). Therefore, the timing of leaf senescence, and the associated changes in plant hormones, may be important with regard to green fruit aroma/flavor.

High soil moisture can increase vegetative growth, and may delay fruit maturation, and the reduction of methoxypyrazines. Increased sun exposure increases the rate of grape maturation and the reduction in methoxypyrazines. Therefore, there is a potential for a lower concentration in leaf-pulled vines, and in vines grown on training systems that may promote more light exposure (and therefore heat) to the fruit.

Excessive crop-to-leaf area can delay the rate of fruit development. If this occurs, the breakdown of methoxypyrazines would be impacted. In a study evaluating the impact of fruitful buds per vine, those properly balanced, but with higher bud counts, had fruit with lower concentrations of IBMP (Allen 2004).

Because pyrazines are in higher concentrations in unripe fruit, the greater the degree of asynchronous ripening, the greater the pyrazines' concentration in the resultant wine. The degree of uneven ripening is an important wine quality limiting factor (see Enology Notes #58 and 81 and above).

b. Processing Factors Influencing Herbaceousness. Processing steps influencing herbaceous compounds include:

- MOG (materials other than grapes) removal
- Stem separation
- Cap management
- Délestage
- Microoxygenation
- Thermovinification

All green grapevine tissues contain methoxypyrazines. The concentration of IBMP in basal leaves is reported to be very high, three to five times that found in the grape clusters. Therefore, leaves in the fermentor can be a source of herbal character.

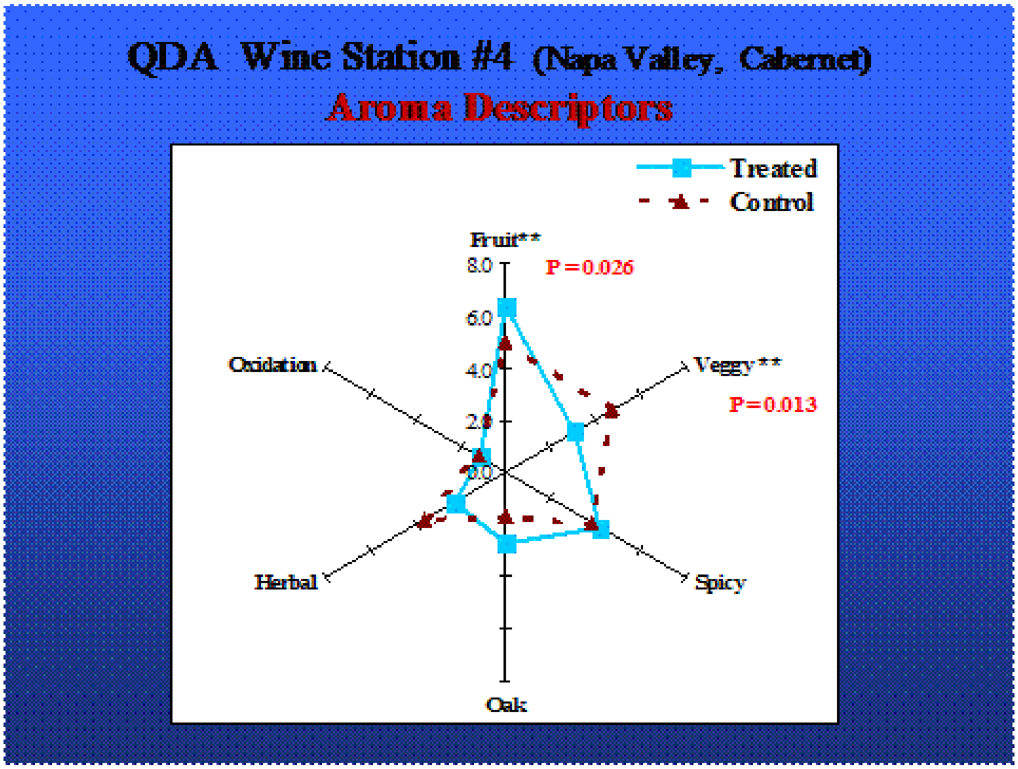
Pyrazines, such as IBMP, are found in Cabernet Sauvignon stems (53%), seeds (31%), skins (15%), and flesh (1%). As such, green pepper-type character in some wines may be the result of stem contact. Many premium wine producers use post-destemming sorting of some red fruit varieties. This may be a critical step, if destemmers leave a significant concentration of cap stem fragments (jacks) in the must. There are now commercially available post-destemmer sorting tables. During a Winemakers Roundtable meeting several years ago, we presented wines that were made with and without jack stem removal. The sensory differences were dramatic.

Herbaceous compounds are also found in the fruit. In Cabernet Sauvignon, the skins contain about 72% of the fruit IBMP, and the seeds about 24% of the total (Roujou de Boubee 2004).

Compounds like IBMP are easily liberated into the juice. Therefore, cap management protocol may not be an important factor in controlling the liberation of these compounds from the fruit, depending upon the length of cuvaison. In some instances, however, press wine will have a higher concentration than free-run wine.

The concentration of methoxypyrazines liberated from the seeds during fermentation depends on several factors, including seed maturity and uniformity of maturity. We have conducted a number of studies using délestage with seed deportation (see Enology Notes #8, 23, 69, 76, 78, 80, and 90). In many instances, there is less herbal character in the resultant wine. This may be the result of seed removal, oxygenation, or other factors.

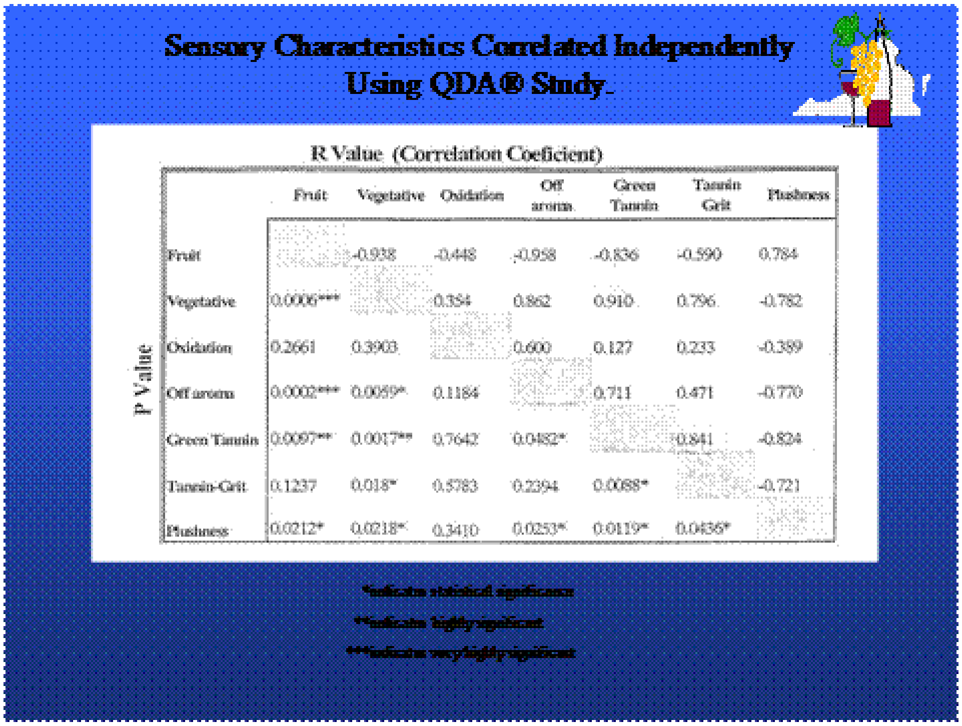
c. Wine Oxygenation and Methoxypyrazines. The effect of microoxygenation on methoxypyrazines is not well understood. It appears that the reduction in the herbal character may not be the result of changes in methoxypyrazines, but in changes in thios or sulfur-containing compounds, that help to reinforce the herbal or vegetative sensory perception. Some thio compounds complement the odor of methoxypyrazines. Sulfur-containing compounds, unlike methoxypyrazines, are not stable. During microoxygenation, it is the oxidation of some sulfur-containing compounds that may result in the muting of the vegetal character of treated wines.



Source: Zoecklein et al. (2002), from the MS thesis of Patrick Sullivan

In evaluating the changes occurring with microoxygenation, we noted that there was a change in the perception of SLO or sulfur-like odors. This change was correlated with a lower perception of herbal character, as a result of the oxidation of thiols to disulfides:

$$2\ R\text{-}S\text{-}H + \tfrac{1}{2}\,O_2 \rightarrow R\text{-}S\text{-}S\text{-}H + H_20$$



	Fruit	Vegetative	Oxidation	Off aroma	Green Tannin	Tannin Grit	Plushness
Fruit		-0.938	-0.448	-0.958	-0.836	-0.590	0.784
Vegetative	0.0006***		0.354	0.862	0.910	0.796	-0.782
Oxidation	0.2661	0.3903		0.600	0.127	0.233	-0.389
Off aroma	0.0002***	0.0059*	0.1184		0.711	0.471	-0.770
Green Tannin	0.0097**	0.0017**	0.7642	0.0482*		0.841	-0.824
Tannin-Grit	0.1237	0.018*	0.5783	0.2394	0.0088*		-0.721
Plushness	0.0212*	0.0218*	0.3410	0.0253*	0.0119*	0.0436*	

Source: Zoecklein et al. (2002), from the MS thesis of Patrick Sullivan

Excessive herbal and vegetative character results in aromatically unbalanced wines. It is essential that winemakers carefully evaluate their young wines (at the proper temperature, not at cellar temperature) to determine the aromatic profile. It is equally essential that premium winemakers understand the environmental, viticultural, and winemaking factors that produce and impact methoxypyrazines.

7. Student Paper Award. Denise Gardner, an MS student in the Enology-Grape Chemistry Group at Virginia Tech, won the award for the Best Student Presentation in Enology at the annual meeting of the American Society for Enology and Viticulture, eastern section.

Her presentation was titled: Electronic nose monitoring of the effects of clusters per shoot and cold soak on Merlot grape and wine volatiles.

8. Winery Planning and Design, Edition 14, Available. This publication, in CD format, is the result of a number of short courses and seminars covering various aspects of winery planning in several wine regions around the country. While not regionally specific, the information provided is from a number of authoritative sources covering such diverse topics as sustainable design, winery equipment, and winery economics. Winery Planning and Design, Edition 14, is available through the industry trade journal *Practical Winery and Vineyard* (phone 415-479 5819), email: donpwv@aol.com or tlv100@sonic.net. Additional information is available at www.vtwines.info.

All past *Enology Notes* technical review are posted on the Wine/Enology – Grape Chemistry Group's website at: http://www.vtwines.info

To be added to (or removed from) the *Enology Notes* listserv, send an email message to rakestra@vt.edu with the word ADD or REMOVE in the subject line.

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