

**Mid-Year Progress Report
Virginia Wine Board, 30 January 2015**

Optimized wine quality potential through fruit-zone management practices in red varieties

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Objective: Evaluate the effectiveness of canopy management practices such as fruit-zone leaf and lateral removal and early season carbon source limitation as means of optimizing grape composition and wine quality of Bordeaux red varieties.

Introduction/background: Leaf removal from fruit zones is a common viticulture practice, especially in humid grape growing regions, as a reduction in disease incidence and severity is often achieved. Leaf removal effects on fruit composition can be highly variable, with many factors confounding results, such as magnitude and timing of leaf removal, variety, growing region, seasonal weather patterns, and compound of interest in grapes/wines, to name a few. Consequently, leaf removal recommendations are often generalized with no mention of how or if a specific grape compound or compound class will change between varieties. Thus, it is our intention to evaluate if changing the magnitude and timing of leaf removal would change Cabernet Sauvignon, Cabernet franc, and Petit Verdot fruit/wine composition and/or consumer acceptability of wines. The compounds or compound classes of interest in fruit and wines have all been shown to be affected by either light, temperature, or both: carotenoids, norisoprenoids, anthocyanins, and total phenolics. Carotenoids are precursors to norisoprenoids, which are key aroma impact compounds in both red and white wines due to their low olfactory perception threshold. Anthocyanins and other flavonoids are important for red wine properties such as color and mouthfeel. Taken together, the value of these compounds in determining optimal leaf removal practice is a consequence of their role in aroma, color and mouthfeel of wines and, thus, consumer acceptability.

Design/Methods: The main project is being conducted in a commercial vineyard in Shenandoah County, with two smaller experiments conducted with Cabernet Sauvignon grown at the AHS Jr. AREC near Winchester. The main project, evaluating pre-bloom and post-fruit set leaf removal effects on yield, cluster architecture, and primary and secondary fruit composition in Cabernet franc and Petit Verdot, was discussed at length in last year's mid-year report. Many of the same responses presented in last year's report were repeated during the 2014 field season. In addition, berry temperatures were collected on green, pink, and red berries during

véraison and the carry-over effects of pre-bloom leaf removal on fruitfulness, yield components, and primary chemistry were evaluated. The majority of 2015 will be spent evaluating total phenolics and anthocyanins in berries as well as berry carotenoid synthesis and degradation patterns and norisoprenoid levels at harvest. In the AHS, Jr. AREC Cabernet Sauvignon vineyard, two separate randomized complete block designs, consisting of one- and two-vine experimental units and each replicated six times, were used to evaluate the effects of pre-bloom and post-fruit set leaf removal on several vine responses. The pre-bloom leaf removal experiment, using one-vine experimental units, evaluated a no leaf removal-control (“P-B-NO”) and pre-bloom leaf removal of four (P-B-4) and eight (P-B-8) basal leaves and laterals from primary shoots (at approx. two to three days before bloom). The post-fruit set leaf removal experiment, using two-vine experimental units, evaluated a no leaf removal-control (“PFS-NO”) and post-fruit set removal of six basal leaves and laterals (“PFS-6”). Fruit zone architecture and light environment was characterized by enhanced point quadrat analysis. Berry temperatures were collected in the morning, around solar noon, and again in the afternoon on both east/west sides of the canopy on seven different dates. One experimental unit of the pre-bloom leaf removal experiment had the following responses logged every 15 minutes from 25-Jul through 4-Sep and every one minute from 5-Sep through harvest (20-Oct). Berry samples were collected, weighed, and frozen at appropriate temperatures for future compositional analyses. Yield data was collected by vine and cluster compactness was evaluated on 10 clusters per experimental unit at harvest. Soluble solids, pH, and titratable acidity were determined from 60 berry samples at harvest. Grape anthocyanins and total phenolics were determined from a composite sample of berries from the cluster compactness assessment (pre-bloom experiment) and from 60 berry samples from both the east and west sides of the canopy at four different dates, including harvest (post-fruit set experiment).

Results: Cluster exposure flux availability (CEFA), a measure of the amount of light received by clusters at véraison, was increased equally by pre-bloom leaf removal of four and eight leaves and post-fruit set leaf removal of six leaves (Fig. 1a). The average number of leaf layers (LLN) in the fruit-zone was around zero for all leaf removal treatments, regardless of leaf removal stage or magnitude; no leaf removal resulted in an average of 2.6 leaf layers (Fig. 1b). These treatments and EPQA results represent unique situations of both timing and magnitude of fruit-zone leaf removal: conventional timing (post-fruit set) and non-conventional timing (pre-bloom) and greater (2.6) and lesser (0.0) fruit-zone leaf layers than conventional recommendation (1.5).

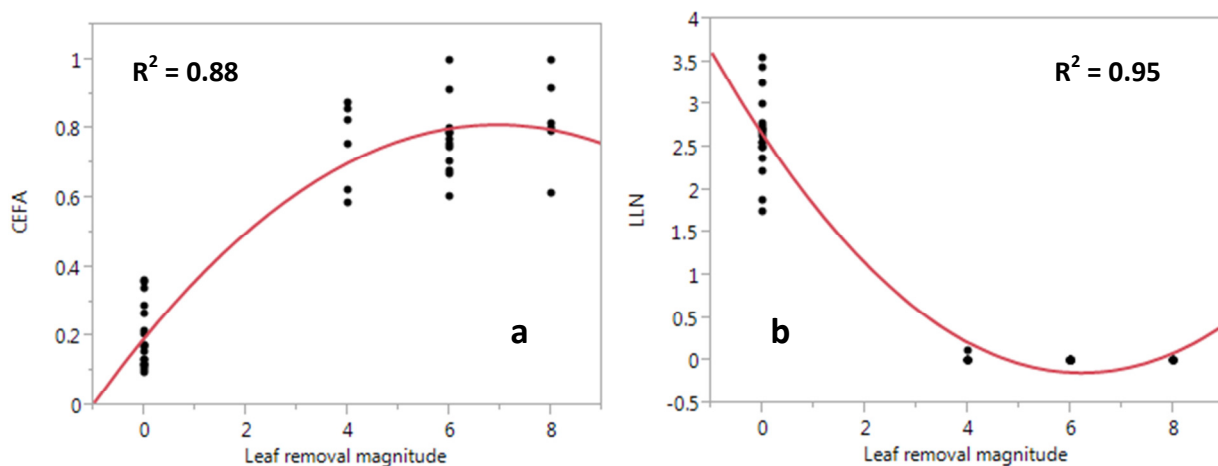


Fig. 1. Pre-bloom (0, 4, and 8 leaves) and post-fruit set (0 and six leaves) leaf removal magnitude effects on cluster exposure flux availability (CEFA, a) and leaf layer number (LLN, b) on 31-Jul-2014.

Berries experienced more time above critical temperatures for limiting anthocyanin accumulation (30 and 35 °C) when leaves were removed to greater magnitudes (Table 1). In 2013, berries spent more time above these temperature thresholds on the west side of the canopy compared to the east side; the case was opposite in 2014. Regardless of year or canopy side, refraining from removing leaves in the fruit-zone resulted in berries never reaching the upper critical temperature of 35 °C.

Table 1. Berry temperature hours spent at or above 30 and 35 °C as influenced by leaf removal treatment and canopy side in 2013 and 2014.

2013*						
	Average		East canopy		West canopy	
	Hours > 30	Hours > 35	Hours > 30	Hours > 35	Hours > 30	Hours > 35
P-B-No	39.50	0.00	38.50	0.00	43.25	0.00
P-B-4	95.75	0.00	97.25	5.25	106.75	7.25
P-B-8	103.00	0.00	107.75	3.00	118.50	9.25
2014**						
	Average		East canopy		West canopy	
	Hours > 30	Hours > 35	Hours > 30	Hours > 35	Hours > 30	Hours > 35
P-B-No	18.00	0.00	18.75	0.00	18.00	0.00
P-B-4	65.40	0.00	108.00	10.50	87.00	4.75
P-B-8	107.00	2.75	141.50	14.00	98.75	10.25

*Logged from 30-Jul through 9-Oct (harvest); **logged from 25-Jul through 22-Oct (harvest); P-B-NO, P-B-4, and P-B-8 = pre-bloom leaf removal of no, four, and eight leaves, respectively

Berry temperature is affected by ambient air temperature in a linear fashion such that as air temperature goes up berry temperature goes up (Fig. 2). Intuitively, berries in the shade are explained by ambient air temperature more so than berries in the sun, which are explained by ambient light conditions more so than berries in the shade (data not shown). Berry temperature is affected by light conditions in a quadratic fashion with a saturation point around $1700 \mu\text{mol m}^{-2} \text{s}^{-1}$, above which berry temperatures appear to no longer be increased. Evaluating the number of data points that fell in the lower and upper 25% of the ambient light range presented in Fig. 1, there were 978 data points lower than $639 \mu\text{mol m}^{-2} \text{s}^{-1}$ and only 224 data points greater than $1916 \mu\text{mol m}^{-2} \text{s}^{-1}$. This implies that conditions were more often cloudy than sunny and berry temperatures spent more time around 24 °C than 28 °C. It is also of note that few of the data points are at or above 30 °C and no data points are at or above 35 °C, the lower and upper critical temperatures for limiting anthocyanin accumulation mentioned above.

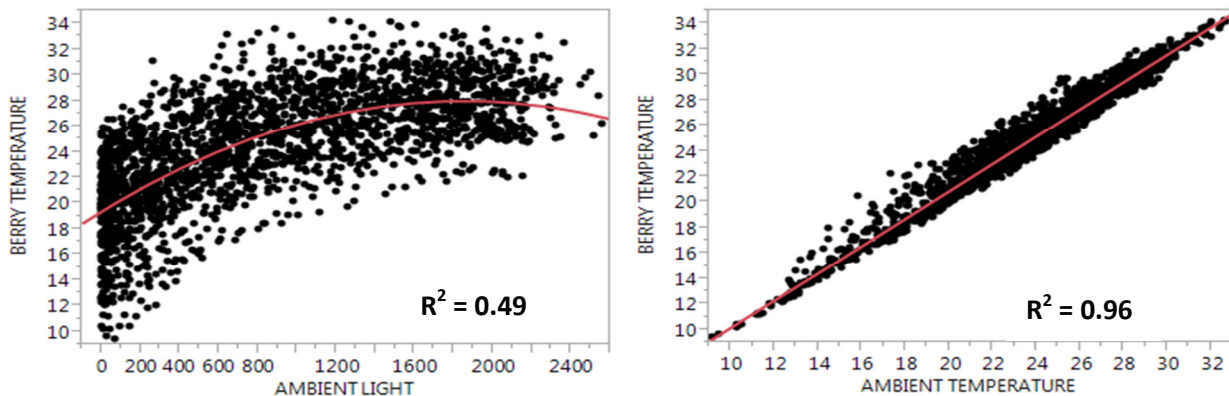


Fig. 2. The effect of ambient light and temperature on berry temperature, as averaged across all leaf removal treatments on both sides of the canopy from 25-Jul through 4-Sep 2014.

Pre-bloom removal of eight leaves reduced soluble solids when compared to not removing leaves (Table 3). Removing leaves in the pre-bloom period in the second year did not affect primary fruit chemistry. Post-fruit set removal of six leaves increased pH and decreased acidity compared to not removing leaves. No leaf removal treatment, pre-bloom or post-fruit set, affected berry anthocyanins. However, removing leaves to greater magnitudes improved total berry phenolics regardless of when implemented during berry development. There was no significant difference in anthocyanins when collected differentially by canopy side (east, west) in the post-fruit set leaf removal experiment (data not shown).

Table 3. Pre-bloom and post-fruit set leaf removal effects on primary fruit chemistry and total berry anthocyanins and phenolics.

Pre-bloom*	Soluble solids** (° brix)	pH**	TA** (g/L)	Anthocyanins*** (mg/g berry)	Total phenolics*** (au/berry)
P-B-NO	21.8 a	3.50	7.91	0.72	67.35 b
P-B-4 (2013)	21.4 ab	3.50	7.30	n/a	n/a
P-B-4	21.4 ab	3.48	7.39	0.72	77.70 ab
P-B-8 (2013)	21.1 ab	3.46	7.18	n/a	n/a
P-B-8	20.8 b	3.49	7.08	0.73	83.10 a
Post-fruit set					
PFS-NO	21.38	3.53 b	8.11 a	0.61	49.38 b
PFS-6	21.38	3.60 a	6.90 b	0.62	55.71 a

*NOTE: model was run with 2013 “re-implement” treatments for comparison purposes – significance can change if removed; removing **collected in 2014 at harvest (TA = total titratable acidity); ***collected in 2013 for pre-bloom leaf removal (from sub sample of berries from cluster compactness assessment at harvest) and collected in 2014 for post-fruit set leaf removal (from berries randomly sampled at harvest); P-B-NO, P-B-4, and P-B-8 = pre-bloom leaf removal of no, four, and eight leaves, respectively; PFS-NO and PFS-6 = post-fruit set removal of no and six leaves, respectively.

Crop yield was reduced by removing eight leaves in the pre-bloom period compared to not removing leaves (Table 2). Re-applying pre-bloom leaf removal treatments in the second year did not change crop yield or cluster weights but increased berry weights when compared to pre-bloom treatments implemented for the first time in 2014. Vine fruitfulness was not affected by pre-bloom leaf removal. Cluster compactness was significantly reduced by removing four and eight leaves in the pre-bloom period compared to not removing leaves. No component of yield was changed by removing leaves in the post-fruit set period.

Table 2. Pre-bloom and post-fruit set effects on components of yield, vine fruitfulness, and cluster compactness in 2014.

Pre-bloom*	Crop Yield (lbs/vine)	Cluster weight (g)	Berry weight (g)	Vine fruitfulness (clusters/shoot)	Cluster compactness
P-B-NO	8.40 a	139.48 a	1.44 abc	0.98	3.78 a
P-B-4 (2013)	6.43 ab	82.70 b	1.62 a	n/a	n/a
P-B-4	5.80 ab	81.00 b	1.36 bc	1.11	2.45 b
P-B-8 (2013)	3.92 b	58.04 b	1.46 ab	n/a	n/a
P-B-8	4.85 b	77.51 b	1.25 c	1.01	1.70 b
Post-fruit set					
PFS-NO	7.44	158.35	1.47	n/a	n/a
PFS-6	7.38	138.75	1.45	n/a	n/a

*NOTE: model was run with 2013 “re-implement” treatments for comparison purposes – significance can change if removed; P-B-NO, P-B-4, and P-B-8 = pre-bloom leaf removal of no, four, and eight leaves, respectively; PFS-NO and PFS-6 = post-fruit set removal of no and six leaves, respectively.

Discussion: Many growers in the industry are skeptical about removing too many leaves in the fruit-zone; this is perhaps a function of an increase in sunburn incidence, but may also be due to recommendations based on popular literature suggesting that too much fruit exposure can limit anthocyanin accumulation in fruit. Thus, conventional leaf removal practice is to remove leaves in the post-fruit set period and to a magnitude of 1.5 leaf layers in the fruit-zone, on average. These experiments aimed to evaluate if removing leaves earlier or to a greater magnitude than convention would improve fruit quality. In both the pre-bloom and post-fruit set leaf removal experiments, removing leaves resulted in zero leaf layers and an equal and greater amount of incident radiation reaching the fruit-zone. In turn, leaf removal resulted in berries that were subjected to critical temperatures for limiting anthocyanin accumulation for longer periods of time when compared to not removing leaves. However, removing leaves to zero leaf layers did not reduce total berry anthocyanin levels, regardless if implemented pre-bloom or post-fruit set. This is potentially because the time that berries in this experiment spent above the upper critical temperature limit for anthocyanin accumulation (35 °C) was as much as 75 hours less than the time that exposed fruit spent at or above this temperature in a particular study conducted in eastern Washington that suggested that too much fruit exposure can limit anthocyanin accumulation. The cloudy nature of a typical growing season in a humid climate, like Virginia, may be partially responsible for limiting the time spent above these critical temperatures as lower ambient radiation values are associated with lower berry temperatures; it is hypothesized that clouds are infrequent during growing seasons in eastern Washington, an arid climate. In addition to not limiting anthocyanins, removing leaves improved total berry phenolics, a proxy for wine quality potential and a category of compounds so broad that it is difficult to make a succinct statement about how the environment could have affected it. Soluble solids tended to be a bit lower with pre-bloom leaf removal; post-fruit set leaf removal increased pH and reduced total titratable acidity. Pre-bloom leaf removal of four and eight leaves reduced crop yield by 31% and 42%, respectively. Cluster weights were reduced by as much as 44% by pre-bloom leaf removal which was likely due to a reduction in berry number per cluster, as cluster compactness was reduced by as much as 55% by pre-bloom leaf removal. Re-applying pre-bloom leaf removal treatments in the second year resulted in no further reduction in crop yield or cluster weight. In a premature conclusion, as not all fruit quality indices have been measured to date, post-fruit set leaf removal to an average of zero leaf layers in the fruit-zone appeared to be a good practice to improve total berry phenolics and reduce acidity without having the deleterious effects on crop yield that pre-bloom leaf removal did.