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Leaf removal: a tool to improve crop control and fruit quality in vinifera grapes

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ABSTRACT

Cool and humid climate in Michigan limits technological fruit maturity at harvest as evidenced by cluster-rot and poor ripening. Economically important wine grape varieties in Michigan have high susceptibility to harvest season cluster rot. Important cultivars that are particularly susceptible are Riesling, Chardonnay, Pinot blanc, Pinot gris, Pinot noir (Vitis vinifera L.). A detrimental characteristic common to all of these varieties is the compactness of the berries held on the cluster rachis.

The aim of this work is to determine whether a quantified amount of leaf removal at bloom would reduce fruit set and consequently produce a controlled reduction in cluster compactness. Research reports have shown in a three-year survey that both pre- and post- bloom hand and mechanical defoliation are effective in limiting yield by means of reducing the number of berries per cluster on a high-cropping cultivar. Cluster size was also reduced while improving must soluble solids and total anthocyanins on a fresh-weight basis.

Our study was conducted to 1) verify whether early leaf removal can be consistently used as a tool for controlling cluster bunch rot through reducing cluster compactness on Riesling and determine the effects of leaf removal on grape quality (skin/flesh ratio, color and basic fruit chemistry parameters).

GOALS & OBJECTIVES

Crop control is a priority in many viticultural areas of the world utilized to insure high quality wines. Cultivar cropping potential is determined by the genotypic bud fruitfulness. The traditional crop-control strategies are winter pruning, node count adjustment, and, for further fine-tuning, manual shoot and cluster thinning. Recent trials reported that manual defoliation near the time of bloom is effective in reducing fruit set and berry size, leading to looser clusters with improved must composition

Objectives

Based upon the relationship between carbohydrate availability near anthesis and final yield the goals of the proposed study was to test several hypotheses:

- **Verify whether early leaf removal at bloom can be consistently used as a tool for controlling yield through reduction of fruit-set in vinifera cultivars characterized by compact clusters thereby decreasing cluster compactness and reducing the potential for bunch rot;**
- **Determine the effects of leaf removal on grape quality (fruit chemistry and skin/pulp ratio);**

OUTCOMES

Crop adjustment are of crucial importance to reach the targeted quality in fruit and wine. The development of this project is helping growers to better understand a practical canopy management (leaf removal) that has a great impact on yield per vine and fruit ripening. Although manual defoliation is a time-consuming operation, the array of its positive effects in improving quality traits is fundamentally important and may prove to far outweigh the initial expense in its pay-off by providing drastically improved quality and yield. The positive impact on formation of looser bunches is also important,

especially in wet years, which could reduce the incidence of bunch rot and increase the quality of the fruit (basic fruit chemistry, color and skin/flesh ratio). In the interest of thoroughness, this project must assess the impact of early-season leaf removal on inflorescence and cluster damage for several years over different growing seasons. This will provide valuable information on the appropriateness of the technique given the particular seasonal characteristics. A key portion of this study will be to investigate the carry-over effect of early leaf pulling on vine reserves, canopy growth and bud induction. This should be explored for several years before it becomes a standard practice for wine grape growers in Michigan to determine that there are no long-term adverse effects on overall vine health. Should early leaf removal prove to be a valid technique in reducing crop, improving fruit quality and decreasing the incidence of bunch rot it may demonstrate a method that takes the place of multiple fungicide applications, reducing the amount of chemical sprayed on a vineyard and the labor cost associated with it, as well as reduce the labor cost of manual crop thinning thereby compensating for its own expense by reducing others while at the same time increasing the value of that very fruit.

PROJECT PERIOD

This project was conducted during the summer and fall of 2014.

WORK ACCOMPLISHED DURING THE PERIOD

Plant Material and Experimental Design. The research was carried out in a 6-yr old vineyard of *V. vinifera*, cv. Riesling grafted on 101-14 rootstock at 12 Corners Vineyards & Winery (1201 N Benton Center Rd Benton Harbor, MI 49022). Vines are planted in a Spinks loamy fine soil with a spacing of 1.8 m between vines and 3.0 m between rows, and trained to a vertical shoot positioning system (VSP). Vines were spur-pruned during the winter leaving approximately sixty buds per vine. No additional shoot or cluster thinning was performed before treatment application. Recommended crop protection practices were followed and the pest management program was based on scouting, experience and weather conditions, except during bloom time to avoid potential mechanical damage to flowers by the sprayer. A combination of fungicides and insecticides used for control were rotated to avoid resistance. Pertinent temperature data were recorded during the experiment by an automated weather station from the Michigan Automated Weather Network (MAWN) located on the site at 1.5 miles from the experimental vineyard. No irrigation was used and standard summer vineyard practices were applied. The experiment was arranged as a randomized complete block design with one categorical factor, leaf removal (LR) with five levels of defoliation: no leaves removed (LR-0); leaves removed from 4 basal nodes (LR-4); leaves removed from 6 basal nodes (LR-6); leaves removed from 8 basal nodes (LR-8); and, leaves removed from 10 basal nodes (LR-10). Approximately 3 weeks before bloom, vines were organized in six blocks by the number of inflorescences, tagged, and then each treatment was randomly assigned to six vines. Additionally, a sub-sample of four shoots per vine was randomly chosen and tagged for detailed measurements of shoot length, degree of fruit set, cluster parameters, and fruit chemistry. Treatments were applied at full bloom, developmental stage 23 after Eichhorn and Lorenz (1977). The timing of budburst, bloom, pea size berries, and harvest were also recorded.

Estimation of Leaf Area. Shoot length was measured weekly from two weeks before bloom up to one month after bloom. A sample of ten shoots, collected weekly from guard vines, was used for estimation of the total leaf area (LA) per shoot. Leaves removed using each defoliation level were collected in ziplock bags and transported to the campus laboratory. In the laboratory, total LA was determined by measuring the single LA with a leaf area meter (LI-3050AHS, Lambda Instruments Corporation, Nebraska). A linear relationship between the LA (y) and shoot length (x): $y = 20.2x - 348$, $r^2 = 0.89$, was used for estimation of total LA. After defoliation, LA removed per shoot was measured and subtracted from total LA in a final calculation for the retained LA.

Estimation of Fruit Set. Each basal cluster per tagged shoot (n=120) was photographed in the field at developmental stage 20 (onset of bloom) and at developmental stage 31 (berries pea size), after Eichhorn and Lorenz (1977). Samples of twenty clusters at developmental stage 20 and twenty clusters at stage 31 from the guard vines were photographed in the field against a dark background and then

separately collected in ziplock bags and transported to the laboratory. Using the same methodology described by Poni et al. (2006), the actual number of florets and berries were destructively counted. The number of florets and berries visible in the photos were counted using Microsoft Office Paint (Windows XP). The linear relationships between the actual number of florets (y) and the counted florets (x): $y = 2.03x$, $r^2 = 0.86$; and actual number of berries (y) and counted berries (x) in the photos: $y = 1.50x$, $r^2 = 0.85$ were used to estimate the initial number of florets and set berries of each basal cluster per tagged shoot. The percentage of fruit set was expressed in two ways: percentage of fruit set at developmental stage 31 (FS-31) and percentage of fruit set at developmental stage 38, harvest (FS-38). FS-31 was calculated as the ratio between the estimated number of set berries three weeks after bloom and the estimated number of florets. FS-38 was calculated as a ratio between the number of berries at harvest and the estimated number of florets.

Cluster Parameters and Morphology. After harvest, basal clusters from tagged shoots were collected and weighed. Berries were separated from the rachis and then total berry numbers, total berry weights, and rachis weights were recorded. Rachis length was calculated as the sum of the central axis length (inner arm), lateral wing or shoulder length (outer arm), and secondary branch length (if they were longer than 5 mm). The number of secondary branches on the inner and outer were also recorded. Cluster compactness was expressed in two different ways: as the ratio between the number of berries and rachis length (compactness index, CI) and as the ratio between the number of branches and rachis length.

Basic Fruit Chemistry. Approximately 20 mL of juice was collected for soluble solids (ºBrix) analysis with an Atago PAL-1 refractometer (Kirkland, WA) and pH measurement with a Thermo Scientific Orion 370 pH meter (Beverly, MA). For total acidity (TA) determination, 10 mL of juice was titrated against a standardized 0.1*N* sodium hydroxide solution in an automatic titrator coupled to an auto sampler and control unit (Titroline 96, Schott, Germany) and expressed as g/L of tartaric acid equivalents. The remaining portions of the berries were briefly frozen and subsequently cold-ground with a tissue grinder (Model PT 10/35, Brinkmann Instruments Co, Switzerland).

Yield Components. At harvest, yield per vine and total number of clusters per vine were recorded. Harvest cluster rot was calculated and recorded as rot incidence (percentage of affected clusters per vine), where every cluster was considered to be affected if it was visually judged to have more than 2-3% of visible rot. Rot severity was calculated as a percentage of affected berries per tagged cluster. During winter pruning post-harvest, pruning weight per vine will be recorded.

Water Retention. One week before harvest, samples of ten random clusters from LR-0 vines and 10 clusters from LR-8 vines were harvested and collected in ziplock bags. Clusters were transported to the laboratory where they were weighed, dipped in water, and weighed again. Wet clusters were hung on a metal rod and air-dried at room temperature. Drying rate was calculated as the difference in cluster weight, which was taken after 5, 10, 15, 45, 60, 90, 120, 150, and 180 min. For this purpose, complete dryness was defined as the moment when clusters had returned to their initial weight. To calculate cluster compactness, berries were separated from the rachis and the total numbers of both berries and branches were counted and the rachis length was measured.

Statistical Analysis. Data were analyzed using one-way ANOVA in PROC MIXED procedure, SAS 9.3. Using the tagged shoots, measurements of shoot length were taken weekly and then analyzed using the REPEATED statement function in PROC MIXED. Normality of the residuals was assessed by visual inspection of the normal probability plot and Kolmogorov test. Whenever the distribution of the residuals was found to significantly diverge from the normal distribution, data were subjected to either logarithmic or square root transformation. Homogeneity of variances was checked using the side-by-side box plot and Levene's test. Models with equal and unequal variances as well as models with different variance - covariance structures in repeated measurements were compared using the goodness of fit indicators. The model that showed the lowest Akaike information criterion (AIC) and Bayesian information criterion (BIC) was used for further analysis. When the treatment effect was found to be statistically significant at $\alpha = 0.05$, all-pairwise comparisons among the treatments were conducted using the t-test. When the treatment effect was not statistically significant at $\alpha = 0.05$, all-pairwise comparisons among the

treatments were conducted using Tukey's HSD. Significance of linear regressions was checked using the Regression Wizard in a scientific data analysis and graphing software package Sigma Plot 11.0.

Preliminary Results

Using the linear correlation between the number of visible florets in the photos and the number of actual florets per cluster, we calculated the estimated number of florets per cluster. Treatments and control showed a minimal of estimated florets per cluster. Leaf removal at bloom and during fruit set caused significantly lower numbers of estimated berries per cluster in the treatments. However, removing leaves in LR-4 and LR-6 obviously could not cause a drastic source reduction, so these treatments had statistically similar numbers of estimated berries per cluster compared to the control; specifically, the non-defoliated control, LR-4 and LR-6 showed approximately 90 estimated berries per cluster. In contrast, FS-31 of LR-8 and LR-10 resulted in an average of 78 estimated berries per cluster, a difference of 14%. The early defoliation caused significantly reduced FS-31 in LR-8 compared to the control, LR-4 and LR-6. The defoliation of 8 basal nodes seemed to be a limitation threshold at which the vines were no longer able to compensate effectively, and this resulted in significantly lower FS-31 and, thus, less estimated berries per cluster. Comparing the estimated number of berries and the actual number of berries, we noticed an additional drop in berry numbers, which occurred between developmental stage 31 and 38.

Change in Cluster Weight Caused by Defoliation. Early leaf removal caused significantly reduced cluster weights for all defoliated vines. Treatments LR-4 and LR-6 had the same mean cluster weight, but significantly heavier clusters than LR-8. Practically, the vines with 8 defoliated nodes had clusters that were more than 40% lighter than the control. The same reduction trend was observed in total berry weight and rachis weight. In comparison to the control, all defoliated vines had lighter berry and rachis weights. LR-4 and LR-6 had approximately 30% lighter total berries and rachises than control. The more severe defoliation in LR-8 caused an approximately 60% decrease in total berry weight and rachis weight compared to the control. Tremendous reduction of cluster size was a result of the decreased number of berries per cluster and not berry size. Although berries from different treatments showed no difference in weight, a reduction in berry size increased with an increased amount of removed leaves. Hence, leaf removal on 8 nodes resulted in 10% decreased berry weight compared to non-defoliated vines. Limitation of source availability during the early stages of cluster development did not significantly affect rachis length. It was observed that LR-8 had slightly shorter rachises than other treatments regardless of the fact that statistical analysis showed no significant differences among vines.

Impact of Early Defoliation on Fruit Chemistry. All defoliated vines had significantly higher soluble solids than the control. The highest soluble solids, i.e., 24 ^{o}Brix, were found in the fruit juice of treatment LR-8. This treatment also showed significantly higher juice pH compared to other treatments and to the control. If compared to non-defoliated vines, significantly lower TA was found in al the.

Yield Components. During harvest, all clusters on a vine were counted and yield per vine was measured. Results showed that defoliated vines did not differ in number of clusters per vine from the control. Since, defoliation did not affect the number of clusters per vine, we concluded that this significant yield reduction was the direct consequence of the about 50% decrease in cluster weight that was found in treatment LR-8. While counting total clusters on the vines, we also counted the existing rot clusters. Similar to the rot severity results, defoliation did prevent rot incidence.

Water Retention. Five minutes after complete wetting, clusters of LR-0 and LR-8 retained water that resulted in a 3.2 and 3.7% increase of their initial weight, respectively. During the next 10 min, clusters lost considerable amount of water and their weight was 1.5 (LR-0) and 1.2% (LR-8) heavier than their initial weight, respectively. Forty-five and 60 min after wetting, the drying rate was significantly higher in LR-10 than in LR-0. That led to a complete drying of LR-10 in which clusters reached their initial weight after 60 min. In contrast, clusters of LR-0 reached that state after 150 min. In other words, LR-0 clusters required a period 2.5 times longer than LR-10 to dry fully under laboratory conditions.

COMMUNICATIONS ACTIVITIES, ACCOMPLISHMENTS, AND IMPACTS

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Figure 1. Vine with leaves removed from 6 basal nodes (LR-6). Photo was taken at bloom.

Table 1. Total leaf area before treatment application; removed LA with early defoliation; and, retained LA per shoot after defoliation.

Treatment[z]	Total LA[w] (cm^2)[y]		Removed LA (cm^2)		Removed LA (%)		Retained LA (cm^2)		Retained LA (%)	
LR-0	800[x]	ns	-	-	-		811.5	a	-	-
LR-4	810		232.4	c	28.5	c	576.2	b	71.5	a
LR-6	824		351.4	b	42.1	b	464.7	b	56.5	b
LR-8	832		540.1	a	65.4	a	275.2	c	34.6	c

Table 2. Early defoliation effect on number of berries per cluster and percentage of fruit set at developmental stages 31 and 38.

Treatment[z]	Estimated number of florets per cluster[y]	Estimated number of berries per cluster		FS-31 (%)[w]		FS-38 (%)	
LR-0	325	100[x]	a	30	a	26	a
LR-4	388	102	a	26	a	24	a
LR-6	341	105	a	30	a	22	a
LR-8	323	79	b	23	b	12	b

Table 3. Impact of early defoliation on fruit chemistry

Treatment[z]	Soluble solids (Brix)[y]		pH		TA[w] (g/L)	
LR-0	20.9[x]	c	3.5	a	6.1	a
LR-4	22.8	b	3.4	a	5.9	ab
LR-6	22.0	ab	3.5	a	5.5	abc
LR-8	22.9	A	3.5	a	5.4	bc