Year 2 Report– May 2010

Impacts of early season fruit zone leaf removal on disease control, fruit set, vine growth and grape and wine quality of Pinot noir

Patricia A. Skinkis & Walter F. Mahaffee

Summary

Early season basal leaf removal can be used as a cultural management component of integrated pest management. Currently, the practice of leaf removal is done at anytime after fruit set and before véraison to increase sunlight penetration for fruit development. Leaf removal at stages prior to bloom may be beneficial in increasing spray penetration for disease control in the grape inflorescence. The study investigates leaf pulling at principle growth stages: flower swelling and separation, bloom (50% cap fall), fruit set, pea–sized berries, bunch close, and no leaf pulling. These stages are represented by the Principle Growth Stages of 57, 65, 71, 73, and 79 respectively (Meier 2001). This second season of this study indicates that there is a benefit to leaf pulling early in the season to increase spray penetration for fungicide sprays and to help alter the microclimate to prevent botrytis or powdery mildew infestation. Leaf removal at principle growth stages 57, 71, and 73 had significantly less disease incidence and severity than the no leaf pull control. There were no differences in fruit set (%), cluster weight or basic fruit composition at harvest with early season leaf removal. This indicates that basal leaf removal employed early season to reduce disease incidence does not compromise flowering and fruitset or fruit ripening. It was previously thought that early season leaf removal could reduce fruitset and yields. However, this is not the case in this first two years of the study. During both the 2008 and 2009 seasons, a warm, dry bloom period lead to a rapid bloom and also reduced the potential for disease infection in that timeframe. Nevertheless, results of fruit analysis in 2009 show that leaf pulling pre-bloom lead to the lowest incidence and severity of grape powdery mildew across all treatments. There was no fruit sun-burning observed in any of the leaf pull treatments in 2008 and 2009 despite complete exposure of clusters from pre-bloom though harvest. Continued research on these trial plots is required to determine the impact of early season leaf removal on disease incidence and secondary effects on fruit quality.

Project Title: Impacts of early season fruit zone leaf removal on disease control, fruit set, vine growth and grape and wine quality of Pinot noir

Principal Investigators

Patricia A. Skinkis, Ph.D., Associate Professor, Viticulture Extension Specialist, Oregon State University, 4017 ALS, Corvallis, OR; Email: skinkisp@hort.oregonstate.edu; Ph: 541-737-1411

Walter F. Mahaffee, Ph.D., Research Plant Pathologist, USDA-ARS-HCRU, 3420 NW Orchard Ave, Corvallis, OR; Email: Walt.Mahaffee@ars.usda.gov; Ph: 541-738-4036

Cooperators:

Leigh Bartholomew, Archery Summit Vineyards, PO Box 85, Dundee, OR 97115; ph: 541-864-4954, e-mail: leighb@archerysummit.com

Dai Crisp, Temperance Hill Vineyard & Lumos Winery, ph: 503-572-0266, e-mail: pkanddai@mac.com

Ted Casteel, Bethel Heights Vineyard, ph:503-991-6631, e-mail: ted@bethelheights.com

Objectives and Experiments Conducted to Meet Stated Objectives:

Research plots were used at the OSU Botany Farm in Corvallis, OR on 10-year old grafted Pinot noir 2A on 420A rootstock trained to a bilateral vertical shoot position (VSP) system at 5' vine and 6'row spacing in rows oriented N to S. Trials were replicated across three commercial vineyards, Temperance Hill, Archery Summit, Bethel Heights, and Stoller in 2009; all sites have Pinot noir of production age (>5 years old) trained to VSP system.

The OSU research block was set up in 2008 as a randomized block design, where each treatment was replicated six times with five vines per treatment replicate. Commercial sites were set up with one replicate of each treatment of a minimum of 10 vines per treatment and replication occurred across vineyard sites. For each treatment, leaves were removed from shoots at specified phenology stages: flower separation, bloom (50% cap fall), fruit set, BB to pea–sized berries, bunch close, and no leaf pulling. These stages are represented by the Lorenz 1994 Principle Growth Stages of 57, 65, 71, 73, and 79 respectively (Meier 2001). All leaves were removed from the base of the shoot upward to the node above the top-most inflorescence/cluster. The cluster zone was cleared and free of leaves and laterals from the time-point of the first leaf pulling through the remainder of the season.

All fungicide (micronized sulfur at 6lbs/100 gallons) applications at the Botany Farm were made with ducted-over-the-row air-blast sprayer with venturi nozzles according to either the Gubler/Thomas Model modified with the Pearson/Gadoury ascospore release model for grape powdery mildew and/or the Broome model for Botrytis bunch rot. In each commercial vineyard block, treatments were managed according the growers standard practices with the exception of leaf pulling. Timing of leaf pulling was conducted as described above.

Objective 1. Evaluate the effectiveness of early flower/cluster zone leaf removal on disease incidence.

Once powdery mildew was detected in the OSU vineyard, each plot was inoculated by placing an infected grape plant every five plants throughout the block. The commercial

vineyards were scouted for disease and plots established near regions were disease was found two weeks prior to initiating the first leaf pulling treatment. Incidence of powdery mildew on leaves was monitored weekly by examining 10 leaves for incidence from each plant. In addition, 20 inflorescences per plot were visually examined for disease incidence every 3 weeks until bunch closure, beginning 2 weeks after inoculation of the vineyard. At harvest, percent cluster infected was determined by freezing 20 clusters per plot at -20°C, then randomly examining 25 berries per cluster for disease incidence.

Inflorescences and clusters were assess for the potential of Botrytis bunch rot by collecting 20 inflorescences per plot 2 weeks after bloom, at bunch close, during lag phase, and at harvest. They were brought back to the lab and placed in a moist chamber for 48 h and microscopically assessed for the number of flowers/berries with signs of Botrytis colonization or infection. All clusters sampled were removed across treatment vines so as not to significantly impact crop level per vine.

Objective 2. Determine the impacts of early leaf removal on floral, fruit and vegetative growth and development.

Treatments that received leaf pulling prior to full bloom were monitored for flowering and fruit set and compared to the control. The protocol for fruit set quantification outlined by Poni et al. 2006 was followed using digital images of inflorescences pre-bloom and clusters after fruit set to quantify percentage set.

In some situations, cluster zone leaf removal can have an impact on total assimilation of carbohydrates, thereby affecting shoot growth, vine size and fruit set. To monitor vine growth responses to leaf pulling early in the season, shoot length was measured for each treatment at bloom and again at fruit set, measuring three shoots from each vine per treatment replicate. Because commercial operations utilize hedging, shoot measurements were not taken from early July onward. Point quadrat analysis (Meyers and Vanden Heuvel 2008) was used to determine the canopy density of each vineyard during véraison. Shoot number per vine, leaf area, and clusters per shoot were recorded during véraison. At harvest, clusters were analyzed for their morphology and size. At the final sampling date (harvest), cluster number and yield per vine was recorded. Clusters were collected for measurements of cluster weight, berry weight, berry size and berries per cluster. Berry size was determined based on diameter separation through sieves of various sizes (4, 8, 12.5, 13.2 and 16 mm). Berries were separated and weighed for each size class.

Objective 3. Evaluate the effect of mechanical leaf pulling in early season applications on flower damage, bloom and fruit set.

As proposed in 2009, we implemented an additional treatment at the commercial sites to determine the efficacy of using mechanical leaf pulling early in the season. Mechanical leaf pulling could be completed no earlier than the BB to pea-size stage to prevent significant damage to the inflorescence or cluster. Using a *Pellenc 4380* over-the-row tractor with leaf puller attachment (courtesy of Devine Vineyard Management), leaves were pulled on both sizes of the cluster zone at the stage when berries were 3-5 mm in diameter. Using this method, we were able to remove leaves without damaging the newly formed cluster. Any earlier applications would have resulted in significant damage to the inflorescence by the leaf puller. This treatment was

applied once to each commercial vineyard site and monitored for the same parameters as outlined in objective 2.

Objective 4. Analyze fruit and wine for chemical components that provide for wine quality.

Harvested fruit quality was analyzed for each treatment and location. Upon harvest, a 30 clusters were collected from each treatment. A ten-cluster sample was frozen for later assessment of cluster and berry size/weights (objective 2). Another ten-cluster sample of fruit was analyzed immediately after harvest for soluble solids, pH and TA. Cluster samples were also measured for SS, pH and TA at three time points prior to harvest. Finally, a ten cluster sample of each treatment rep is being reserved and stored in -80ºC for compositional phenolic analyses (anthocyanins, resveratrol, flavanols, and flavonols) by an analytical lab in the future. In the meantime, samples of fruit are being analyzed for berry skin polyphenol and anthocyanin concentration using the Folin-Ciocalteau (Singleton and Rossi 1965) and pH-differential (Giusti and Wrolstad 1996) methods, respectively by the Skinkis lab to get preliminary data with respect to these treatments and to narrow down total treatments for further investigation. Currently, total phenolics and anthocyanins have been determined for the commercial vineyard locations sampled in 2009 (Jan 2010). Within the next month, the remainder of the samples will have been analyzed. Compositional analysis (HPLC) of fruit for polyphenolic composition will ensue in spring/summer 2009 for the 2008, 2009 fruit.

Summary of Major Research Accomplishments and Results by Objective:

Objective 1. Evaluate the effectiveness of early flower/cluster zone leaf removal on disease incidence.

There were significant differences in disease incidence on clusters among treatments in small plot trial at OSU Botany Farm in 2008. Samples from 2009 for this site are still being analyzed for disease infestation and severity as of January 2010. However, data from the commercial vineyard sites indicate a lower incidence and severity of grape powdery mildew infections in fruit at harvest for the pre-bloom leaf pulling (Figure 1). Leaf removal at principle growth stages 71 (fruit set), and 73 (BB-sized) had significantly less disease severity than the no leaf pull control in 2009. These data mirror results found in 2008, although incidence was very low across all sites in both years. Inflorescences developed very rapidly in both 2008 and 2009 due to a warm, dry bloom period which effectively reduced the period of time which they are susceptible to infection by either Botrytis or powdery mildew. These factors probably negated much of the expected advantage of inflorescence exposure and increased pesticide coverage for reducing disease development during bloom.

Data collected on botrytis bunch rot incidence and severity in berries is currently being analyzed for the 2009 season. In general, the Botrytis pressure was much higher in 2009 due to late season weather, and we expect to see some differences across treatments.

Objective 2. Determine the impacts of early leaf removal on floral, fruit and vegetative growth and development.

Flower and berry number were measured on ten flagged inflorescences per treatment replicate to determine the impacts of leaf pulling on fruit set. Based on flower number per cluster

and individual berry number per cluster after set, there was no difference in total flower number, berry number or percentage fruit set by treatment across all experimental sites (Figure 2). In fact, 2009 was a high fruit set year, and all sites yielded greater than 50% fruit set. Similarly, in 2008 there were no fruit set differences by treatment (n=100, P>0.05) and set was normal at ~50%. By harvest, there were no treatment differences in berry or cluster weight in 2008. In 2009, differences in cluster and berry weight were observed by treatment (Table 3). The results are not due to variability in fruit set. These data combined indicate that carbohydrates were not limiting enough in the early part of the growing season to limit flower development and reduce fruit set for the pre-bloom and bloom leaf pulling treatments. Vine vigor is high in the Willamette Valley and across all sites used for this study. It is likely that carbon is not limiting enough to create problems with fruit set when leaves are pulled early in the season. Furthermore, shoot growth can be quite significant early season with some vineyards reaching the top trellis wire by bloom. Although there is high vigor in the vines, this has not created a problem with sink competition during the two years of this study.

Vine vegetative growth was monitored to determine any potential impacts of treatments on growth and to determine vine size in relation to fruit quality parameters. Shoot lengths were measured for the pre-bloom, bloom and no-pull treatments at fruit set. There were no differences with treatment for shoot lengths just after fruit set (Table 1) or leaf areas by the latter part of the season (Table 2). This helps quantify that there were not disparate leaf areas that could play a confounding effect on other vine or fruit measures. Shoot lengths and leaf areas of the OSU vineyard site (BPP) were not included with the commercial sites due to significantly shorter shoot lengths by the same phenological stages. This was due to frost damage in spring 2009 resulting in secondary shoot growth in the block. Point quadrat analysis, a method for determining vine canopy density, was conducted in 2008 and 2009 and there were no difference with treatment.

Dormant pruning weights are being collected early February 2010 for each trial location. This will help determine any differences in vine size due to leaf pulling. Based on the lack of differences in leaf areas, shoot lengths and ripening of fruit during the growing season, we do not believe pruning weights will differ by treatment.

Objective 3. Evaluate the effect of mechanical leaf pulling in early season applications on flower damage, bloom and fruit set.

As proposed in 2009, we implemented an additional treatment at the commercial sites to determine the efficacy of using mechanical leaf pulling early in the season. Mechanical leaf pulling could be completed no earlier than the BB to pea-size stage to prevent significant damage to the inflorescence or cluster. Using a *Pellenc 4380* over-the-row tractor with leaf puller attachment (courtesy of Devine Vineyard Management), leaves were pulled on both sizes of the cluster zone at the stage when berries were 3-5 mm in diameter. Using this method, we were able to remove leaves without damaging the newly formed clusters. Leaf pulling was not done by machine after the initial treatment. Results show that there was no difference in disease incidence or severity of the mechanical treatment when compared with the control or hand-leaf pulling at bunch closure. The hand pulling at the same stage (phenological stage 73) had less incidence of disease likely because the hand pulling ensured that all leaves were removed from the cluster

zone while the mechanical pulling removed between 60-100 percent of the leaves in that same zone. Also, leaves were not re-pulled mechanically beyond that first time point.

Objective 4. Analyze fruit and wine for chemical components that provide for wine quality.

Fruit analysis was conducted at harvest for each location and treatment. There was no treatment effect on basic berry composition of soluble solids, pH or TA in 2008 or 2009 (Table 3). The results of anthocyanin and phenolic concentration are available for the commercial sites (Table 3) and OSU BPP site fruit is currently being analyzed. There are no differences by treatment from the commercial vineyard sites. Data obtained from the OSU Botany Farm site may yield more clear results in comparison of treatments replicated at one location. Results of this will help us determine which treatments to carry into the 2010 season and narrow down samples for HPLC compositional analysis in the future to work within the budgeted funds for this project.

Outside Presentations of Research:

The preliminary data obtained from years one and two were presented at the Oregon Winegrape Symposium in February 2009 and at various regional meetings within Oregon. During the 2009 season, a field day was held in the southern Willamette Valley on canopy management where preliminary data of this research trial was discussed and demonstrations of mechanical leaf pulling were conducted. Reports of the research results will be posted online at the OSU winegrape website (http://wine.oregonstate.edu) under Viticulture Research. At the completion of this multi-year study, results will be compiled for peer-reviewed publication in the *Journal of the American Society of Enology and Viticulture, HortScience* or other relevant scholarly journal.

Results of this work will also be used in an Extension outreach publication for canopy management. Some innovative methods of delivering this information are being developed and currently utilized by my program. This includes online modules that are designed as new-age "reference books" that include information on how, why and when to use certain vineyard management practices. This will be made free-access online and will be peer-reviewed. I plan to develop such a module for canopy management in Oregon (or cool climates).

Research Success Statements

Results of the first two seasons suggest that Pinot Noir in the cool climate of the Willamette Valley does not experience reduced fruit set with early season leaf pulling. During 2009 we increased pulling intensity and kept the cluster zone free and clear of leaves for the duration of the season. Although current results do not indicate reduced fruit set that we were hypothesizing, this information is encouraging given the lack of fruit set problems with these early season leaf pulling. Early results suggest the practice may be implemented without any problems with fruit set and/or fruit quality later in the season. Alternatively, we are uncertain whether early season leaf pulling can be used as a way to reduce fruit set through carbon reduction to further cut cluster thinning costs later in the season. Further studies would need to be conducted to determine the viability of that practice for Oregon Pinot Noir. Results from 2008 and 2009 indicate that early season leaf removal with mechanical leaf pullers is possible but only

once the clusters have reached approximately 5 mm in diameter. At that time point, there was little to no damage to the clusters.

Fund Status:

This study was funded by the Oregon Wine Board during August 2008 – June 2010. A research proposal has been submitted in January 2010 to the Unified Grant Management System for the third and final year of this project's funding cycle. Research plans are currently being prepared for the 2010 growing season and viticulture measurements will commence in May 2010 for the third funding cycle, pending award.

Literature Cited:

Giusti, M.N., and R.E. Wrolstad. 1996. Characterization of red radish anthocyanins. J. Food Sci. 61:322-326.

Meier, U. 2001. Grapevine. In Growth stages of mono- and dicotyledonous plants. BBCH Monograph. Federal Biological Research Center for Agriculture and Forestry, Berlin, Germany.

Meyers, J.M., and J.E. Vanden Heuvel. 2008. Enhancing the Precision and Spatial Acuity of Point Quadrat Analyses via Calibrated Exposure Mapping. Am. J. Enol. Vitic. 59:425-431.

Singleton, V.L., and J.A. Rossi. 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am. J. Enol. Vitic.16:144-58.

Smart, R.and M. Robinson. 1991. *In* Sunlight into Wine: A Handbook for Winegrape Canopy Management, pp. 21. Ashford, South Australia: Winetitles.

Figures and Tables

The tables and figures shown below do not reflect all data collected in 2009. A sampling of tables and figures from 2009 were selected for the summary and brevity in this annual report.

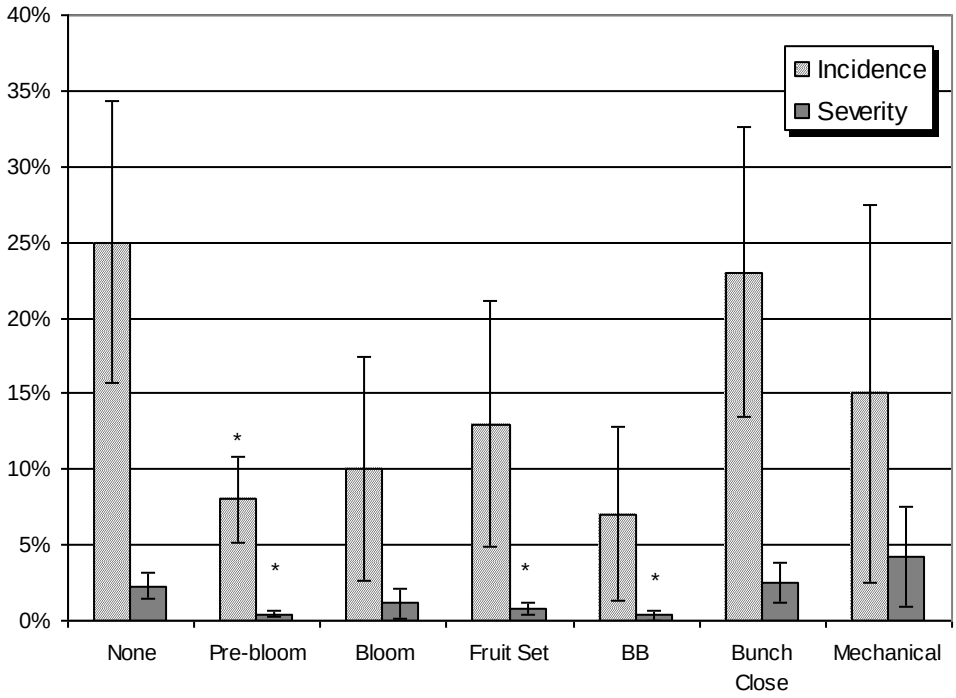

Figure 1. Mean (± SEM) percent incidence and severity of powdery mildew infections of 25 berries selected randomly from harvested clusters. Difference from the control at $P<0.05$ indicated by (*). The control is "none" which did not have leaves pulled. Data gathered from experimental plots at commercial vineyard sites.

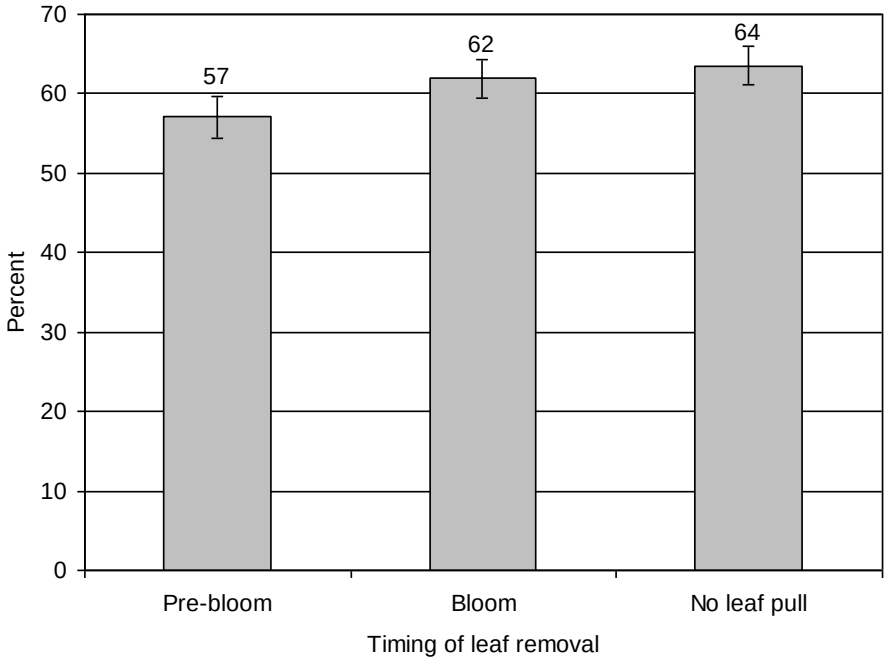

Figure 2. Mean (± SEM) percentage of fruit set of early season leaf pulling treatments and control (no leaf pulling) across all experimental sites in 2009. Timing of early season leaf pulling did not effect fruit set (P>0.05).

Table 1. Shoot lengths at Fruit Set (cm)

Treatment	BPP	Commercial Sites
Pre-bloom	49.9	110.6
Bloom	50.8	118.6
No pull	41.1	110.5
P	*0.0659* [a]	*0.4038* [b]

Means presented. No treatment effect was found at BPP (OSU trial site) or across commercial sites. P-values shown are a result of [a]ANOVA and [b]Kruskal-Wallis analysis.

Table 2. Leaf areas (m^2) of vines at véraison, 2009

Timing	Commercial Sites	OSU BPP
Pre-bloom	2.75	1.60
Bloom	3.36	1.65
Fruit Set	2.81	1.67
BBtoP	2.89	1.70
Closure	2.84	1.54
Control	3.54	1.51
Mechanical	3.31	–
P	*0.1905*	*0.7356*

Mean (m2) leaf area presented. No treatment effect at any location.

Table 3. Fruit harvest measurements and fruit composition, 2009

	Treatment	Cluster wt (g)	Berry wt (g)	SS (°Brix)	pH	TA (g/L)	Total skin phenolics (mg/g)	Total Anthocyanins (mg/g)
Commercial Vineyard Sites	pre-bloom	113.77	1.20 ab	21.7	3.13	10.5	9.95	2.44
	bloom	128.03	1.14 ab	21.2	3.11	10.9	9.83	2.44
	fruit set	102.57	1.00 b	21.9	3.13	9.9	9.83	2.33
	BB to P	117.07	1.03 ab	20.9	3.12	10.2	11.04	2.47
	Closure	128.76	1.32 a	20.7	3.14	11.0	9.55	2.48
	Control	143.18	1.08 ab	21.5	3.15	10.1	9.38	2.38
	Mechanical	119.38	1.02 b	20.9	3.16	10.3	10.93	2.73
	P	*0.4803*	***0.0263***	*0.8219*	*0.9859*	*0.9832*	*0.5383*	*0.9924*
OSU Botany Farm	pre-bloom	67.24	1.23 ab	20.6	3.11	9.3		
	bloom	61.14	1.12 b	20.2	3.08	9.4		
	fruit set	72.81	1.28 ab	20.0	3.03	10.4		
	BB to P	100.90	1.33 a	18.2	3.07	9.2		
	Closure	72.45	1.17 ab	20.2	3.07	10.1		
	Control	86.63	1.27 ab	20.2	3.05	10.8		
	P	***0.0003***[a]	***0.0209***	*0.3076*	*0.1850*	*0.1514*		

Means presented. Statistical analysis conducted by ANOVA, REGWQ means separation with exception of [a]Kruskal-Wallis.