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

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ABSTRACT
Cool summers and wet fall in Michigan often limits technological fruit maturity at harvest especially in cultivars prone to cluster-rot. Several important wine grape varieties in Michigan have high susceptibility to harvest season cluster rot (e.g. Riesling, Chardonnay, Pinot blanc, Pinot gris, Pinot noir, all 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. 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 Michigan to assure good quality fruit at harvest. Cultivar cropping potential is determined by the genotypic bud fruitfulness and the traditional crop-control strategies are winter pruning, node count adjustment, and, for further fine-tuning, manual shoot and cluster thinning. Manual defoliation near the time of bloom could be effective in reducing fruit set and berry size, leading to looser clusters with improved must composition and reduced cluster bunch rot.

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

- **Can early leaf removal at bloom 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 at harvest**

OUTCOMES
Crop adjustment are of crucial importance to reach the targeted quality in fruit and wine. This project demonstrated that a practical canopy management (leaf removal) that has a great impact on yield per vine and fruit ripening. Manual defoliation is a time-consuming operation; however 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 clusters is also important to reduce the incidence of bunch rot and increase the quality of the fruit. Early leaf removal proved to be a valid technique in reducing crop, improving fruit quality and decreasing the incidence of bunch rot. It will be important as future development of this project try to test the hypothesis that this technique may takes the place of multiple fungicide applications, reducing the amount of chemical sprayed on a vineyard. It could reduce also 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 2015.

WORK ACCOMPLISHED DURING THE PERIOD

The experiment was conducted in 2015 at Michigan State University's Southwest Michigan Research and Extension Center in Benton Harbor (MI) on a plot of Riesling (clone 49) grafted of 3309-C rootstock. The vines were trained in a Scott Henry system at a spacing of 1.8 meter inter-row and 2.7 meter intra-row. A total of 40 vines were tagged and selected for the trials. Of these, the 24 best most uniform vines were selected as the main experiment, while the last 16 where used as extras for sampling. Two treatments were applied to each half of the selected vines, defoliation and control. For each treatment, 12 vines were of the main experiment and 8 vines were in the extra group. All selected vines where inspected and number of inflorescences and shoots on each vine was counted and length of the shoots were registered. Using the collected data the plot was divided into blocks of four, each block containing three each of main experiment vines with an additional two extra experiment vines, with eight vines per block. On each vine, five modal shoots on the upper cordon were selected and tagged for data collection. The selection criteria for the shoots were two potential clusters and an average shoot length. Selected shoots were tagged with white laminated paper tags with a number 1 through 5. All the vines where selected and inspected prior to bloom. After selection of experimental vines the defoliated group was defoliated and the six basal leaves of the shoots on the upper cordon were removed. The shoots on the lower cordon were untreated in all groups. For each shoot the defoliated leaves were collected and put into a quart size bag and the bags for one vine were collected in a gallon size bag with the defoliated leaves of non-tagged shoots. On the date of bloom, the individual inflorescences were photographed still on the vine with a digital camera (Panasonic, Lumix GH2, Tokyo, Japan) on a black cardboard background. From the extra vines, ten shoots from guard vines were collected and five shoots were selected from the extra defoliated group. The shoots were selected on the criteria of two inflorescences and average length of the shoot (ie. a modal shoot). In the vineyard, the collected shoots were separated from their respective inflorescences and the shoots and the inflorescences were tagged according to their treatment and assigned a number between 1 and 15 (1-10 for the control and 11-15 for the defoliated group). Like the inflorescences from the main experiment group, the inflorescences were pictured. In the laboratory, the flowers on each inflorescence were counted as were the flowers on the pictures. Based on the two numbers for each inflorescence a regression curve was calculated to estimate the number of flowers on the main experiment inflorescences. The leaf area of the defoliated leaves was measured using a leaf area meter (LI-3050AHS, Lambda Instruments Corporation, Nebraska) to determine how much leaf area was removed from the defoliated vines. The sampled shoots taken from the vineyard likewise had their leaf area measured in relation to shoot length. The regression curve based on this was used to evaluate the amount of leaf area each shoot on tagged vines in the vineyard had, allowing for determination of difference between defoliated group and control group. Each week the length of all tagged shoots was measured and a selection of shoot samples from the guard vines were collected from the extra vines to add observations to the regression line data. As standard practice on vines, leaves are removed from the fruit zone around the time of veraison, to reduce the impact of botrytis on the clusters. The treatment was performed on the control group of the Riesling on September 4th. On October 8, harvest took place. During the harvest, each basal and apical cluster was bagged and tagged from the tagged shoots on the vine (1-5), yielding approximately 10 sample clusters from each vine. The rest of the clusters were harvest but not registered except to note the specific yield of each cordon (upper and lower) in number of clusters and total weight of yield from each vine.

Cluster Morphology analysis. The morphology of the cluster is characterized by the following parameters: cluster weight, stem weight, number and weight of total berries, number and weight of large berries (berries that have seeds), number and weight of small berries (berries without seeds), number and weight of rotten berries (berries that were affected by a combination of the following characters: brown, broken skin, smelly, wrinkly and clear growth of mold), stem length, number of lateral branches on the stem and length of the lateral branches. Each cluster was first weighed on a one point decimal scale (Mettler PJ4000), then all large berries were picked off the cluster and counted. Each harvested cluster were cut from the shoot at the proximal end of the peduncle and the cluster was measured with a standard ruler from the point of the cut to the distal point of the rachis. The first lateral branch, often referred to as the shoulder, was also included in the full stem length and not included in the category of lateral branches. When counting the lateral branches, a branch was considered a branch if it was beyond 1 cm in length and/or with more than one group of pedicels along its axis.

Grape Berry analysis. The grapes were analyzed based on variety of characteristics that included: skin weight, pulp weight, number of seeds, Brix, total acidity and pH. Once the grapes had been processed for the morphological analysis they were transferred their respective labelled plastic bags and frozen in an -80°C. This allowed for the separation of the skin, the pulp and the seeds for their respective weighing and counting. For each cluster, five large berries (with seeds) were selected and measured. They were then peeled and the peel of all five berries collected. The pulp was then cut in two and the seeds were counted and dug out from the pulp using a small pair of tweezers. Brix was measured once for each cluster based on a sample drawn from the thawed cluster bag. All the bags were thawed and then crushed just enough to release their juice. The juice was poured from the bag in to small plastic cups and a few drop of the mixed juice was put on a refractometer (ATAGO Pocket Refractometer PAL-1). pH was measured for each sample using an electric pH-meter that had been calibrated prior to use. Total acidity was measured as an automated process using an automatic titration system that could take up to 16 samples each revolution. For each sample beaker a known quantity (standard was 10 mL, but some samples had less than 10 mL which was accounted for in the calculations later) of juice was mixed with an unknown quantity of demineralized water such that there were enough to cover the pH-meter. 16 sample beakers were placed in a Titration sample changer (SCHOTT Instruments, TW alpha plus). The samples were titrated with a 0.1 M NaOH solution by a universal titrator (SCHOTT Instruments, Titronics Universal) and the results were automatically recorded using a data collection program.

Terpene analysis. In order to verify any difference in terms of flavor, a broad analysis for terpene content of the grapes was. While this particular flavor compounds group does not cover all the potential flavor compound groups found in white wine, it may serve as a good indicator of potential differences in potential wine quality. For the determination of terpenes each block were separated and each treatment within each block was juiced into a beaker to a total amount of ~ 80 mL for a total of 4 blocks * 2 treatments = 8 juice samples. Each sample had its pH adjusted just prior to distillation to put it within the pH-range of 6,90-7,10 by drop wise addition of an acid (HCL) and base (NaOH). Each prepared sample yielded two terpene samples: free volatile terpenes and potential volatile terpenes for a total of 16 samples (Fig 1).

Fig 1*. Markham still. The upper valve (top left corner of the picture) is at the start OPEN to the outer column. The lower valve (mid left, just next to the head of the Bunsen burner in the picture) is CLOSED. Outer column is filled with distilled water. The upper valve is then turned to OPEN for the inner column (thus CLOSED for the outer column) to pour in the grape juice sample.*

RESULTS

There were only a very small difference between the number of shoots and clusters on the vines between treatments, with a 4.3% difference in the number of shoots and a 1.8% difference between the number of clusters and a 1.5% difference in clusters/shoots. Leaf area of the vines could not be precisely determined in a non-destructive fashion, so it was estimated based on a correlation between the length and the measured leaf area on a selection of medial shoots (the criteria for the shoot being average length compared to the rest of the vine it was selected from, with all leaves intact and the presence of two inflorescences). All tagged shoots were measured using a measuring tape and the leaf area of a particular shoot was determined using the determined equation. Between the two treatments there is the large difference of 52.4% in leaf area and a difference of 56.6% when in relation to the number of clusters on the vines (Leaf Area/Cluster). Between the treatments the difference in number of flowers was 11.2%, between berries was 23.2% and lastly there was a 32.2% difference in the percentage of fruit set. As with the leaf area, the average flower and berry number on the vines are an estimate, based on an equation determined through the destructive counting of flowers/berries on a cluster which was then related to counted flowers/berries on pictures of same clusters.

Treatment	Average cluster weight (g)	Average weight of large berries (g)	Average number of rotten berries	Average weight of rotten berries (g)
Control	85.64 a	55.69 a	30.47 a	17.56 a
Defoliated	66.35 b	44.37 a	16.5 b	11.39 b
Difference	22.5%	20.3%	45.9%	35.1%

Between the two treatments, there are strong differences between all of the weight characteristics. The average weight of the clusters in the two groups are different by 22.5% and the average weight of the large berries (berries with seeds) are different by 20.3%. Of the average cluster weight, 65% and 66.9% of that weight is made up from large berries in the control group and defoliated group respectively. Between the two groups the difference in rotten berries are also strong, with a difference of 45.9% in the average number of rotten berries on the clusters and a 35.1% difference in weight between these rotten berries.

Treatment	Average rachis length (cm)	Average length of laterals (cm)	Average number of laterals	Average stem weight (g)
Control	10.71 a	2.57 a	1.78 a	5.17 a
Defoliated	9.64 a	1.85 b	1.48 a	3.54 b
Difference	10%	28%	16.9%	31.5%

There were difference between the control and defoliated vines when it comes to the average length of the laterals and the average weight of the stems, with the first being 28% and the latter being 31.5%. For the length of the rachis, the difference is less, being only 10% and for the average number of laterals the difference is around 17%. To further elaborate on the cluster morphology, it can be seen from this data that in terms of weight/cm of stem the control is at (5.17 grams /(10.71 cm + 2.57 cm * 1,78)=) 0.34 grams/cm and the clusters from the defoliated vines are at (3.54 grams/(9.64 cm + 1.85 cm * 1,48)=) 0.29 grams/cm. Of the total stem length (rachis + lateral branches), the laterals make up around 30% and 22% of the total in the control and defoliated group respectively. Further, for every 10 cm of rachis length there are 1.66 and 1.54 lateral branches for the control and the defoliated group respectively.

Treatment	Average total number of berries	Average total weight of berries (g)	Average % Rotten number of berries	Average % rotten berry weight
Control	92,26 a	79,18 a	31,49 a	23,60 a
Defoliated	71,69 b	61,49 b	21,28 b	17,21 a
Difference	22,3%	22,3%	32,4%	27,1%

Of the total number of berries on the average cluster on the control and the defoliated group, 31.5% and 21.3% were rotten berries. In terms of weight, the percentage is 23.6 and 17.2 respectively for the control and defoliated group. Between the rotten berries, there is a 32.4% difference in terms of the number of berries, while the rotten berry weight has a difference of 27.1%. The data quite clearly suggests that there is more rot on the clusters from the control group than there are on the clusters from the group of defoliated vines. Given the percentages of rot in the bunches, this leaves a total number of berries with rot within each treatment to be 29 and 15.3 berries/cluster that has been affected by rot.

Looking at the total number berries/cluster and the total weight of berries/cluster, the control has more berries and weighs more than the clusters from defoliated vines with a difference at 22.3%. The average total number of berries accounts for large berries and small berries. The table below shows the numbers and differences between the average pulp weight of the large (with seeds) berries, the average weight of large berries (skin, seeds and pulp) , the average °Brix in the grapes along with the average total acidity and the total (free+potential) terpene content.

Treatment	Average pulp weight (g)	Average weight of berries (g)	Average of Brix	Average of TA (g/L)	Total Terpenes (mg/L)
Control	1.41 a	1.88 a	17.58 a	7.48 a	1.46 a
Defoliated	1.21 a	1.67 a	18.70 a	7.06 a	1.61 a
Difference	14.2%	11.2%	6.4%	5.6%	9.9%

When it comes to the berry analysis the four main characteristics where (albeit slight) differences were noticed were the average weight of the pulp, the average weight of the berries, the average of the sugar content and the total acidity. The average pulp weight indicated in the table above is the total weight of five berries including the measuring boat. This yields the pulp weight for one berry to be 1.41 grams and 1.21 grams for control and defoliated respectively. The same principle applies to the total weight of the berries, with a single whole berry weighing 1.88 grams and 1.67 grams for control and defoliated respectively. The difference between the two groups are 14.2%nd 11.2% for the pulp weight and the total berry weight respectively. For the sugar and acid content of the grapes, the control has a higher degree of acid content but lower degree of sugar content as compared to the berries from the defoliated vines.

COMMUNICATIONS ACTIVITIES, ACCOMPLISHMENTS, AND IMPACTS

1. Sabbatini P., D. Acimovic, L. Tozzini and P. Murad. 2015. Early leaf removal to reduce cluster compactness and improve fruit quality. Southwest Michigan Horticultural Days, February 4-5, Lake Michigan College, Mendel Center, Benton Harbor (MI).
2. Sabbatini P. 2015. Impact of canopy management, crop load and vine balance on fruit quality in red wine grapes. Ohio Grape and Wine Conference, February 16-17, Dublin (OH).
3. Sabbatini P. 2015. Early leaf removal: a tool for controlling yield and improving quality of grapes grown in cool and wet climate. Ohio Grape and Wine Conference, February 16-17, Dublin (OH).
4. Acimovic D., L. Tozzini and P. Sabbatini. 2015. Defoliation intensity threshold for reduction in fruit set, change in cluster morphology and increased fruit quality in small and tight cluster Vitis vinifera L. cultivar Pinot noir. Australian Journal of Grape and Wine Research (accepted)

FUNDING PARTNERSHIPS

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