



Impact of cluster thinning on productive characteristics and wine phenolic composition of cv. Merlot

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Abstract

The effects of cluster thinning on productive characteristics and wine phenolic composition of a red grapevine variety Merlot (*Vitis vinifera* L.) was evaluated in this study. Cluster thinning consisted of removing upper clusters on shoots in order to remove 30% and 60% of clusters at veraison. Control treatment without cluster thinning was also applied. Two levels of cluster thinning reduced yield by 19% and 40%, respectively. Average cluster weight and leaf area/fruit weight ratio were increased and fruit maturity was advanced with cluster thinning. Wines produced from cluster thinned treatments had higher alcohol content, lower acidity, higher concentration of total phenolics, total anthocyanins and higher color intensity comparing to the control treatment. Cluster thinning affected not only the total anthocyanin concentration but also the anthocyanin profile in wines. It significantly increased the levels of delphinidin-3-monoglucoside, cyanidin-3-monoglucoside, petunidin-3-monoglucoside and peonidin-3-monoglucoside, while the concentration of the most abundant individual anthocyanin malvidin-3-monoglucoside and acylated anthocyanins did not differ considerably among treatments. The highest increase in the concentration of phenolic compounds occurred in the treatment with 60% of clusters removed. It was concluded that more than 1.5 m² of leaf area per kg of fruit was needed for the production of high quality Merlot wine, which is especially adequate for aging due to high concentration of phenolic compounds.

Key words: Cluster thinning, phenolics, anthocyanins, color intensity, Merlot.

Introduction

Grapevine crop level and the balance of vegetative and productive growth are considered important factors for high quality grape and wine production. Since vegetative and productive growth develop simultaneously in grapevine, competition between them occurs if they are not in balance or resources are not sufficient. Excessive crop loads delay ripening and may reduce fruit and wine quality ¹. The partial removal of reproductive sinks by cluster thinning is a canopy management practice that can be used in order to limit crop level and enhance the leaf area/fruit weight ratio. As a result, grape maturation may be advanced and potential wine quality improved, which may be especially important in vines that are overcropped and thus out of balance ².

Several studies investigated the impact of cluster thinning on phenolic compounds, especially anthocyanins and tannins, as they are responsible for bitterness, astringency and color intensity of wines. Removal of 50% of clusters one month after bloom concentrated soluble solids, berry skin anthocyanins and flavonoids of cv. Nebbiolo ³. Cluster thinning increased the phenolic composition of wines as indicated by increases in total anthocyanins, total phenolics and total resveratrol content of red winegrape hybrid Chambourcin (*Vitis* spp.) ⁴. Anthocyanin concentration expressed as berry fresh weight basis was higher in fruit from the mechanically thinned vines compared to control (unthinned) vines of cv. Cabernet Sauvignon ⁵. However, grape

and wine quality is not always improved with cluster thinning. Hand thinning of grapes of cv. Nebbiolo ⁶ and mechanical thinning of cvs. Tempranillo and Grenache ⁷ improved the phenolic composition of grapes in some years of investigation, while in other years there were no differences between cluster-thinned and control vines. In some cases cluster thinning of Cabernet Sauvignon vines failed to improve the color of grapes in all investigated seasons ^{2, 8}. There were no differences in color intensity of Cabernet Sauvignon wines produced from vines having about 60 to 70 clusters per vine or thinned to 40 and 20 clusters per vine ⁹. In the study conducted by Nuzzo and Matthews ¹⁰ different crop levels affected the timing of ripening in a high-yielding, dry-farmed Cabernet Sauvignon vineyard, but the quality of the berries based upon standard assays of fruit composition was not affected. It is considered that improvement in grape quality is not obtained with cluster thinning if vines are not overcropped ^{2, 10}.

There may be differences among cultivars in terms of their response to or tolerance of heavy crops ². Moreover, each region should investigate for each variety and training system the optimum yield that will still produce quality wine yet still return an economic crop ¹.

Merlot is the second most widespread grapevine variety in Istria, where it represents about 13% of cultivated vineyard area and it is still widely planted in recent years. Istria is an ancient

winegrowing region, located in the north-west of Croatia (the Mediterranean Basin, Central-Eastern Europe). Up to now, no research on cluster thinning of cv. Merlot has been conducted and published in Istria or other regions of Croatia.

As differences occur among different varieties and vine growing regions, the aim of this study was to investigate the effects of cluster thinning on productive characteristics and grape and wine quality characteristics of a red grapevine variety Merlot (*Vitis vinifera* L.) in Istrian agroecological conditions.

Materials and Methods

The experiment was performed in 2008 on seven years old *Vitis vinifera* L. cv. Merlot vines, clone Rauscedo 3, grafted on K5BB (*Vitis berlandieri* x *Vitis riparia*) rootstock in a commercial vineyard located near Bale (West Istrian winegrowing region, Croatia), 50 m above sea level. The soil was typical, deep, anthropogenized red Mediterranean soil (Terra rossa). Row and vine spacing were 2.5 m x 0.5 m, corresponding to 8000 vines per hectare. Vines were trained to single Guyot training system, with one renewal spur and one cane left on each vine. Shoots were vertically positioned and sustained with two pairs of catching wires, positioned 40 and 80 cm above the basal wire. Shoots thinning was performed at grapevine growth stage 16 according to the modified E-L system¹¹, in order to attain approximately 15 shoots per metre of canopy. Leaves and laterals were partially removed in the cluster zone after the end of bloom, at grapevine growth stage 27 according to the modified E-L system¹¹. When berries were at pea size, shoots were trimmed 30 cm above the upper pair of catching wires. Some additional leaves were removed at veraison. Other viticultural practices were standard for the cultivar and region.

The sum of growing-degree days in the period from April to September in 2008 was 2103, while the sum of rainfall was 298 mm. The warmest month was July with the average temperature of 24.8°C. Rainfall was highest in April (110 mm), June and August had approximately 60 mm, May 30 mm, whilst other months had somewhat less than 20 mm of rainfall¹².

In this experiment a randomized block design was used, with three cluster thinning treatments: control treatment without cluster thinning; cluster thinning of approximately 30% of clusters on vine (30% CT) and cluster thinning of approximately 60% of clusters on vine (60% CT). Each treatment was applied in three replications with five adjacent vines. Cluster thinning was performed at the beginning of veraison, at grapevine growth stage 35 according to the modified E-L system¹¹. Clusters were thinned manually. As a rule, lower clusters were left on the shoots, while upper clusters were removed.

The 60% CT treatment was harvested on 12th September, 30% CT treatment on 19th September, whilst control treatment was harvested on 26th September 2008. Harvest date was postponed one week for 30% CT treatment and two weeks for control treatment due to slower ripening of these treatments in relation to 60% CT treatment.

Yield and number of clusters per vine were recorded at harvest. Two hundred berries were randomly chosen from each treatment replicate to determine mean berry weight. Mean cluster weights were calculated from yield and clusters per vine data. Leaf area was determined as described by Smart and Robinson¹³. Samples for must analyses were taken after crushing-destemming of grapes.

Soluble solids (°Brix) were assessed by HR200 digital refractometer (APT Instruments, Litchfield, IL, USA). Titratable acidity (expressed as g L⁻¹ of tartaric acid) and pH value of must and wine and volume fraction of alcohol in wine were analyzed by O.I.V. methods¹⁴. Weight of cane prunings were measured at winter pruning.

Fermentations were carried out per each treatment in triplicate. Grapes were destemmed, crushed, treated with 50 mg L⁻¹ SO₂ and inoculated with *Saccharomyces cerevisiae* (Enoferm BDx; Lallemand, Montreal, Canada). Fermentations were conducted in 5-litre glass fermentors at a temperature of 25°C. Pomace was mixed two times per day. After seven days of fermentation and maceration pomace was pressed using a small mechanical press. Wines were racked and sulfited with 50 mg L⁻¹ SO₂. Two months after the end of fermentation wines were taken for the analyses.

Total phenolics were evaluated as stated by Singleton and Rossi¹⁵ using Folin-Ciocalteu reagent. The quantification of total phenolics was carried out using a calibration curve prepared with known amounts of gallic acid. The total anthocyanin content in wines was determined using bisulphite bleaching method¹⁶. The free anthocyanin content was determined with HPLC according to method of Berente *et al.*¹⁷. Wine color intensity was evaluated as stated by Glories¹⁸, measuring the optical density at 420, 520 and 620 nm using UV/VIS spectrophotometer (Cary 50, Varian, USA).

Data were analyzed by the analysis of variance (ANOVA) using Statistica software package (version 8; StatSoft, Tulsa, OK, USA). Duncan's multiple range test was used for *post hoc* comparison of significant treatment means.

Results and Discussion

Yield per vine was reduced in cluster thinning treatments (Table 1). The decrease in yield was lower than the rate of cluster thinning due to the increase in berry and cluster weight following cluster thinning, which is confirmed also by other authors^{19,20}. In 30% CT treatment, which had 32% less clusters than control treatment, yield reduction of 19% appeared, while in 60% CT treatment, which had 61% less clusters than control treatment, yield reduction was 40%. This was partly due to the augmentation of berry weight in cluster thinning treatments comparing to control treatment (Table 1), which was 13% and 34% for 30% CT and 60% CT treatments, respectively. The other reason of lower decrease in yield as compared to the rate of cluster thinning applied is the removal of upper clusters on shoot, which are usually smaller than basal clusters²¹. This assumption is confirmed with the extent of augmentation of average cluster weight induced by cluster thinning, which is higher than the augmentation of average berry weight. The augmentation of average cluster weight was 18% and 57% for 30% CT and 60% CT treatments, respectively.

Table 1. Effects of cluster thinning on yield components of Merlot grapevines.

Treatment	Yield/ vine (kg)	Clusters/ vine	Cluster weight (g)	Berry weight (g)	Shoots/ vine
Control	1.87 a	14.53 a	128.9 c	0.89 c	7.27
30% CT	1.51 b	9.87 b	152.7 b	1.01 b	7.33
60% CT	1.13 c	5.60 c	202.4 a	1.19 a	6.93

Means within columns designated by different letters are significantly different by the Duncan's multiple range test ($p = 0.05$).

Leaf area per vine did not vary considerably among treatments (Table 2), while the leaf area/yield ratio was inversely proportional to yield per vine, with control treatment having the lowest leaf area/yield ratio, and 60% CT treatment having the highest ratio. As it is considered that 0.8 to 1.2 m² of leaf area per kg of fruit is required for maximum level of total soluble solids, berry weight and berry coloration at harvest for single-canopy training systems²², it can be deduced that no treatment had insufficient leaf area per kg of fruit for the ripening of crop. Leaf area/yield ratio of 60% CT treatment (1.84 m² kg⁻¹) may seem to be too high, but Naor *et al.*²³ proved that total wine sensory score for Sauvignon Blanc wines decrease with decreasing the leaf area/yield ratio below ≈1.80 m² kg⁻¹. Similarly to leaf area per vine, pruning weight did not vary considerably among treatments, while the yield/pruning weight ratio was the highest in control treatment and the lowest in 60% CT treatment. Yield/pruning weight ratio for control treatment and 30% CT treatment were in the range considered optimal (4–10 kg kg⁻¹) for the production of high quality fruit and wines according to Kliewer and Dokoozlian²², and such vines are considered well balanced. Yield/pruning weight ratio for 60% CT treatment is lower than optimal, but this value is not a consequence of severe vegetative growth, but rather of extensive fruit removal.

Table 2. Effects of cluster thinning on vegetative growth and vegetative-productive indices of Merlot grapevines.

Treatment	Leaf area/ vine (m ²)	Leaf area/ yield (m ² kg ⁻¹)	Pruning weight/ vine (kg)	Yield/pruning weight (kg kg ⁻¹)
Control	1.96	1.05 c	0.32	5.83 a
30% CT	2.05	1.37 b	0.38	4.01 b
60% CT	2.08	1.84 a	0.36	3.16 b

Means within columns designated by different letters are significantly different by the Duncan's multiple range test ($p = 0.05$).

Although harvest was postponed one week for 30% CT treatment and two weeks for control treatment due to slower ripening, the highest degree Brix was obtained by the 60% CT treatment, whilst the control treatment had the lowest degree Brix (Table 3). High yield delays maturation of the fruit, so that more time is required for the sugar content to reach a given degree Brix²⁴. Soluble solids content increased as the leaf area/yield ratio increased. Leaf area/yield ratio is considered more important for quality achievement than a yield *per se*¹³, due to higher assimilate availability for the fruit if the leaf area/yield ratio is increased. Control treatment obtained higher titratable acidity than both cluster thinning treatments, which indicates faster ripening of cluster thinning treatments. Moreover, dilution of acids in cluster thinning treatments may be a consequence of larger increase in berry weight and volume from veraison until harvest. The faster ripening of grapes on vines with lower crop level was also confirmed by other authors^{10, 20, 25}. Nuzzo and Matthews¹⁰ reported that despite the latter ripening of vines with high crop

Table 3. Effects of cluster thinning on berry composition of Merlot grapevines.

Treatment	Soluble solids (°Brix)	Titratable acidity (g L ⁻¹)	pH
Control	21.9 c	7.3 a	3.23
30% CT	22.9 b	6.7 b	3.20
60% CT	24.4 a	6.5 b	3.29

Means within columns designated by different letters are significantly different by the Duncan's multiple range test ($p = 0.05$).

level, no or little impact on cluster and berry size and final soluble solids content were found among different crop levels obtained by cluster thinning on dry-farmed Cabernet Sauvignon, indicating that agroecological conditions impact the productive response of grapevine.

Alcohol concentration in wines followed the same trends as the sugar concentration in grapes, while the titratable acidity in wines was almost the same as the titratable acidity in grapes (Table 4). The concentration of total phenolics in wine was highest in 60% CT treatment. There were no considerable differences in total phenolics between control treatment and 30% CT treatment. The 60% CT treatment had also the highest total anthocyanin concentration, while control treatment had the lowest total anthocyanin concentration. Same trends were found for the color intensity of wines. It is generally considered and confirmed by some studies that cluster thinning leads to the improvement of the phenolic composition and color of red grapes and wines³⁻⁵. In some cases this hypothesis was not proved^{2, 8-10}, and it is considered that in these studies the vines were not overcropped and consequently environmental conditions allowed full ripening of grapes. Since in our study cluster thinning improved the phenolic composition of Merlot wines, it is assumed that control treatment vines were slightly overcropped, despite the fact that the leaf area/yield ratio and crop yield/pruning weight ratio of control treatment vines (Table 2) were in ranges considered optimal for fully ripening of grapes according to Kliewer and Dokoozlian²². The values of total phenolics, total anthocyanins and color intensity followed similar trends as soluble solids content and the leaf area/yield ratio, indicating that cluster thinning may have increased the accumulation of phenolics indirectly by advancing fruit maturity or directly by altering the source to sink balance⁴. Guidoni *et al.*³ hypothesize that sugar concentration may influence berry anthocyanin composition, which could be the case also in this study. It is deduced that high quality Merlot wine in the agroecological condition of Istria is obtained with the leaf area/yield ratio higher than 1.5 m² kg⁻¹, which is higher than generally recommended²², but consistent with some findings on Nebbiolo variety in Piedmont region, Italy⁶.

Table 4. Effects of cluster thinning on basic and phenolic composition of Merlot wines.

Treatment	Alcohol (vol %)	Titratable acidity (g L ⁻¹)	pH	Total phenolics (mg L ⁻¹)	Total anthocyanins (mg L ⁻¹)	Color intensity
Control	12.2 c	7.2 a	3.06 b	1600 b	559 b	1.57 b
30% CT	12.8 b	6.5 b	3.05 b	1670 b	658 a	1.74 ab
60% CT	13.9 a	6.5 b	3.29 a	2110 a	741 a	1.96 a

Means within columns designated by different letters are significantly different by the Duncan's multiple range test ($p = 0.05$).

The concentrations of individual anthocyanins were consistent with the concentration of total anthocyanins in wines of different treatments. The highest concentration of most individual anthocyanins was obtained in 60% CT treatment, while the lowest concentration of most individual anthocyanins was obtained in control treatment (Table 5). Cluster thinning significantly increased the levels of delphinidin-3-monoglucoside, cyanidin-3-monoglucoside, petunidin-3-monoglucoside and peonidin-3-monoglucoside, while the concentration of the most abundant individual anthocyanin malvidin-3-monoglucoside and acylated

Table 5. Effects of cluster thinning on individual anthocyanin concentration (mg L⁻¹) of Merlot wines.

	Control	30% CT	60% CT
Delphinidin-3-monoglucoside	2.4 c	5.1 b	8.5 a
Cyanidin-3-monoglucoside	0.1 b	0.2 b	0.9 a
Petunidin-3-monoglucoside	5.2 c	9.1 b	12.3 a
Peonidin-3-monoglucoside	5.2 c	10.7 b	13.7 a
Malvidin-3-monoglucoside	78.8	96.1	93.1
Peonidin-3-monoglucoside-acetate	0.2	0.2	0.1
Malvidin-3-monoglucoside-acetate	23.7	26.4	24.6
Peonidin-3-monoglucoside-p-coumarate	1.8 b	3.7 a	4.0 a
Malvidin-3-monoglucoside-p-coumarate	7.7 b	14.1 a	12.2 a

Means within rows designated by different letters are significantly different by the Duncan's multiple range test ($p = 0.05$).

anthocyanins did not differ significantly among treatments. Similar findings were obtained by Guidoni *et al.*³ on cv. Nebbiolo, indicating that cluster thinning affects not only the total anthocyanins content, but also the specific accumulation of individual anthocyanins during ripening, which results in different anthocyanin profile in grapes and wines.

Conclusions

Results showed that yield reduction at veraison obtained by cluster thinning reduced yield, increased the leaf area/fruit weight ratio, accelerated ripening and improved the phenolic composition of Merlot wine in Istrian agroecological conditions. High intensity of cluster removal (60%) led to the highest concentration of total phenolics, total anthocyanins, most individual anthocyanins and color intensity of wine. More than 1.5 m² leaf area per kg of fruit was needed to achieve high quality Merlot wine, which is especially adequate for aging due to high concentration of phenolic compounds. Despite the increase in quality of wines achieved with cluster thinning, concern should be put on the loss of crop obtained with this canopy management practice, and every producer should find its optimal balance according to the desired wine style.

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