Impact of Crop Control Strategies on Performance of High-Yielding Sangiovese Grapevines

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Abstract: Climate change will require grapegrowers to develop improved viticultural practices to control vine yield and the rate of fruit maturation. The impacts of five canopy management regimens on vegetative growth, yield, and grape quality were investigated over three years, and carryover effects on vines in the fourth year were examined. Winter pruning (*Wp*, the control), shoot thinning (*St*), shoot thinning with preanthesis defoliation (*St*+*Dpa*), shoot thinning with preveraison defoliation (*St*+*Dpv*), and shoot thinning with preveraison defoliation plus cluster thinning (*St*+*Dpv*+*Ct*) were applied to Sangiovese vines from 2011 to 2013. Neither *St* nor *St*+*Dpv* changed yield or grape quality compared to *Wp*. The *St*+*Dpa* treatment reduced leaf area and yield by 33% compared to *Wp* and *St* and led to increased sugar concentrations and a carryover effect into 2014 that reduced vine capacity. A management strategy that combines shoot thinning with preanthesis defoliation, which will increase sugar concentrations and suppress yield, offers the strongest potential for long-term regulation of vine yield and grape quality. However, in a nonirrigated vineyard of medium vigor, *Wp*, *St*, and *St*+*Dpv* could be used to achieve yield and fruit quality levels that meet defined thresholds while reducing costs in respect to other additional interventions such as *Dpa* or *Ct*.

Key words: cluster thinning, defoliation, leaf removal, shoot thinning, winter pruning

Given the threat of climate change (IPCC 2014), grapegrowers in Italy and other major grapegrowing countries are focused on developing strategies to better manage grape and wine production under conditions of rising temperature (Schultz 2000, Palliotti et al. 2014). This includes characterizing varieties, clones, and rootstocks to find those that demonstrate inherent resistance to abiotic stress, such as thermal and radiative excesses and water limitation (Chaves et al. 2010). However, it is likely that at least five to ten years of effort will be needed before results are ready for the field. Near-term strategies are also required, and a new set of cultural management techniques capable of regulating the grape-ripening process has been developed for improved yield and/or fruit sensory characteristics (Palliotti et al. 2013a, 2013b, Herrera et al. 2015).

One of the most important outcomes of increasing temperatures is acceleration of the ripening process, which leads to increases in grape sugar concentration and an often-undesirable increase in alcohol levels in the resulting wines (Jones et al. 2005). This can cause deviations from the expected wine style and failure to meet consumer expectations for taste. One solution is to alter vineyard cultural practices. Shoot thinning (*St*) is one of the most common practices used to adjust canopy density and crop load via the removal of extra shoots arising from count nodes of spurs or canes. Malformed or sterile shoots are first removed, but fertile shoots are also often eliminated. However, shoot removal can cause short-term loss of leaf area followed by vegetative compensation with longer shoots and more lateral shoots (Kliewer and Dokoozlian 2005, Myers et al. 2008, Bernizzoni et al. 2011). *St* can decrease the ratio of yield to pruning weight (Naor et al. 2002, Myers et al. 2008), i.e., the Ravaz index (Ravaz 1911), which is commonly used to assess the balance between vine growth and yield. Ravaz index values between 5 and 10 kg/kg indicate a good balance between yield (kg fruit per vine) and vine vigor (dormant pruning weight) (Kliewer and Dokoozlian 2005). Investigations of French–American hybrids, such as Aurore, Chancellor, and Villard noir (Morris et al. 2004) have shown that timely *St* can improve grape quality (Keller et al. 2008, Susaj et al. 2013). In addition, when this technique is used in tandem with cluster thinning, vegetative growth increases, yield decreases, and wine quality increases, the latter as a result of improved wine fruit (Sun et al. 2012).

Defoliation is another technique that has been studied extensively, and it is generally applied at two different times during the growing season and with different objectives. The effects of leaf removal on canopy density, yield capacity, fruit characteristics, or disease resistance differ depending on the time of application. If the first six or seven basal leaves are removed before bloom (*Dpa*), which temporarily limits carbohydrate sources, then the result will likely be a reduction in the

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quantity of grapes produced in direct response to a significant fruit set decrease (Coombe 1959, Candolfi-Vasconcelos and Koblet 1990, Caspari et al. 1998, Poni et al. 2006, 2008, Intrieri et al. 2008, Bravetti et al. 2012, Gatti et al. 2015). This in turn decreases cluster compactness and, thus, tolerance to rot and rot-prone conditions (Sabbatini and Howell 2010). Previous studies also report that source limitation induced by early defoliation impacts growth of the berry skin and flesh mass, thus influencing final berry size (Poni and Bernizzoni 2010, Gatti et al. 2015), but does not lead to imbalances in leaf area-to-yield ratio, since the reduction in total leaf area is counterbalanced by decreased yield (Bravetti et al. 2012, Gatti et al. 2015). Investigations with many different grape varieties have also confirmed that removal of the first six basal leaves before bloom can limit vine yield capacity and cluster compactness and can secondarily improve grape quality (Intrieri et al. 2008, Poni et al. 2008, Bravetti et al. 2012). Poni et al. (2006) used *Dpa* with success on an array of genotypes (e.g., Sangiovese, Trebbiano, Barbera, and Lambrusco) under different growing conditions, demonstrating the capacity of *Dpa* to improve cluster morphology, control crop load, and improve grape and wine composition.

Defoliation performed prior to veraison (*Dpv*) is the partial or total removal of leaves around the cluster zone, timed between the stages of fruit set and veraison on high-density canopies. *Dpv* can increase light exposure and aeration of clusters, leading to improved sugar content and skin and flesh pigmentation as well as tolerance to fungal diseases (Zoecklein et al. 1992, Bravetti et al. 2012). Unlike *Dpa*, *Dpv* has little impact on canopy net assimilation, since basal leaves in advanced age are eliminated (Petrie et al. 2000).

Cluster thinning (*Ct*) is typically performed manually and in vineyards that are managed for premium-quality wine. *Ct* significantly reduces yield, increasing the leaf area-to-yield ratio, accelerates grape ripening, increases sugar concentrations, and decreases titratable acidity (TA) and pH, improving phenolic composition and wine color intensity. The removal of whole or parts of clusters modifies the Ravaz index by reducing crop load. This technique has been shown to improve grape quality in numerous cultivars and French–American hybrids (Bravetti et al. 2012, Susaj et al. 2013, Gatti et al. 2015). Different pruning and canopy management techniques can induce complex changes in the source/sink balance, and leaf removal or modification of shoot numbers often results in an improved canopy microclimate, which leads to improved fruit quality at harvest.

Here, we performed a four-year investigation (2011 to 2014) on a high-yielding red Italian variety and evaluated the effects of different canopy management techniques on canopy density, yield capacity, and grape quality. Our objective was to determine the best strategy for achieving the desired levels of yield and grape composition. Additionally, we characterized weather patterns during the study to compare the treatments under different environmental conditions. Lastly, during the last experimental season (2014), we evaluated potential carryover effects of several canopy management strategies on vine growth and development and fruit maturity at harvest.

Materials and Methods

Plant material, trial conditions, and experimental design. The trial was carried out from 2011 to 2014 in a hillside vineyard (~5% slope) near Ancona in the Marche region of east-central Italy (lat. 43°32′N; long. 13°22′E; 203 m asl). The vineyard was planted in 2004 with certified virus-free cuttings of Sangiovese (clone R24) grafted onto SO4 rootstock. The vines were spaced 1.20 m within rows and 2.75 m between rows, oriented NNE to SSW, with a planting density of 3030 vines/ha. Each vine was cordon-trained, vertically shoot-positioned, and winter-pruned, leaving seven spurs of two nodes each. The cordons were located 0.6 m aboveground with two pairs of catch wires providing trellising extending 0.9 m above the cordons. During the study, shoots were mechanically trimmed when the majority of their growth exceeded the top wires, usually in mid-June. A pest and disease management program was implemented according to prevailing local practices influenced by field scouting, experience, and weather conditions.

The study was carried out on 30 uniform vines chosen along one row and organized into three randomized blocks of 10 grapevines each. Each block was divided into five plots of two vines each; the same treatment was assigned to the vines in each plot to have two replicates per treatment per block and, therefore, a total of six replicates for each treatment. The canopy management techniques included: winter pruning (the control treatment), in which all vines were uniformly pruned at dormancy (*Wp*); shoot thinning at preanthesis to 14 shoots per vine (*St*); shoot thinning with preanthesis defoliation during the rapid shoot elongation phase (*St*+*Dpa*); shoot thinning with preveraison defoliation in the fruit zone with full canopy and bunch closure phase (*St*+*Dpv*); and shoot thinning and preveraison defoliation combined with cluster thinning (*St*+*Dpv*+*Ct*).

Seasonal climate variability during the trial resulted in a shift in the dates on which treatments were implemented. From 2011 to 2013, we performed the *St* treatments, reducing shoots to 14 per vine during the last 15 days of May each year, except for six control vines subjected to *Wp* only. The *Dpa* treatment, consisting of manual removal of leaves and laterals from the first six basal nodes of each shoot, was performed on six *St* vines during the last 10 days of May in each year. The *Dpv* treatment, in which leaves were removed from the first six basal nodes of the shoots in the fruit zone on 12 *St* vines, and the *Ct* treatment, in which six vines were cluster thinned to obtain a per-vine yield between 3.6 and 4.2 kg, were performed during the last 10 days of July in each year. The target yield for *Ct* corresponded to the limits imposed by the trial site's controlled designation of origin (DOC) regulations, which includes the Sangiovese cultivar, by leaving one cluster per shoot where possible, or by removing clusters partially. In the final season (2014), vines were winter pruned only, leaving seven spurs of two nodes each per vine.

Vine growth and canopy measurements. From 2011 to 2013, we recorded the number of shoots per vine before and after thinning. Seasonal evolution of the canopy was monitored using point quadrat analysis (Smart and Robinson 1991) using 100 to 120 insertions, according to the height of the canopy, at 10-cm intervals with a thin metal rod following a sampling grid. The canopy density, expressed as leaf layer number (LLN), was monitored considering the effects of treatments on LLN in the fruiting zone (at 20 and 40 cm from the cordon). Leaves removed in each treatment were subdivided into primary and lateral leaves, and leaf area was determined using a leaf area meter (LI-3100, LI-COR Inc.).

Gas exchange measurements. During the three-year trial, leaf gas exchange activity in vines subjected to the *St* and *St+Dpa* treatments was evaluated to identify any treatment-induced compensation effects. Measurements were carried out in the morning (from 0930 hr to 1130 hr) on clear days at varying intervals until harvest using a portable, open-system LCA3 infrared gas analyzer (ADC BioScientific Ltd.). The system had a broad leaf chamber with a 6.25 cm^2 window; all readings were taken at ambient relative humidity with airflow adjusted to 350 mL/min. For each treatment, three fully expanded leaves at node positions six to 10 from the base were sampled under saturating light (PAR >1400 photons $mmol/m^2/s^1$).

Vine yield and grape composition. Harvest dates were 23, 21, and 17 Sept in 2011, 2012, and 2013 respectively, when sugar accumulation (total soluble solids, Brix) began to level off as measured from grapes sampled by mid-August. In 2014, grapes were harvested and analyzed on 15 Sept. Grapes were individually picked and the total number of clusters per vine was counted and weighed. Mean individual cluster weight was calculated as the ratio of total cluster weight per vine (yield) to the total number of clusters per vine.

In 2012, data collection was expanded to include cluster morphology. Ten individual clusters per vine were weighed separately and their size (length and width), compactness (weight to length ratio, g/mm), and fruit health status (rating of *Botrytis cinerea* and sunburn) were recorded.

Each year, 100 berries per vine were collected and weighed to determine berry fresh weight. The berries were then crushed and the juice was used to determine total soluble solids, pH, and TA. Total soluble solids were measured using a temperature-compensating Maselli LR-01 digital refractometer (Maselli Misure). Must pH was analyzed with a Crison pH meter (Crison Instruments) using a glass electrode, and TA was determined with a Crison titrator (Crison Instruments) using 0.25 N NaOH to a pH 7.00 endpoint, expressed as g/L of tartaric acid equivalent.

Over the three years of the study, the yeast assimilable nitrogen (YAN) concentration, including ammonium and α-amino acids, was estimated in each year following the Ogorodnik and Merkureua (1971) procedure reported in Gump et al. (2002). Each year, total anthocyanin and polyphenol concentrations were determined according to Mattivi et al. (2004), using the samples from which juice characteristics were determined. The grapes were pressed to extract all of the juice; then, the crushed grapes (skins + seeds) were placed in a jar containing a buffer solution extractive of chloridric acid (15 mL in 1 L of water) and homogenized with an Ultra-Turrax T25 (Janke & Kunkel, Ika-Labortechnik) for 1 min at 10000 rpm, and stored in the dark for 30 min. Next, a subsample of homogenate was transferred to a centrifuge tube and centrifuged (model ALC 4218, International s.r.l.) for 10 min at 3200 rpm. The liquid phase was collected in dark glass bottles (25 mL), which were filled completely. For anthocyanin determination, the extract was diluted with ethanol hydrochloric and analyzed using 10-mm cuvettes on a spectrophotometer (UV-1601, Shimadzu Corporation) at 520 nm. Anthocyanin content was calculated as malvidin 3-glucoside chloride equivalents (mg/kg of grape).

To determine total polyphenols, the extract was diluted with water. A 1-mL portion was transferred into a 20-mL calibrated flask, and 2 mL of methanol, 5 mL of water, and 1 mL of Folin–Ciocalteu reagent was added. After 3 min, 4 mL of sodium carbonate (10%) was added and the solution was left to stand for 90 min. Then, absorbance was registered at 700 nm on the spectrophotometer using 10-mm cuvettes. Concentrations were determined using a calibration curve and expressed as (+)–catechin, mg/kg of grape.

In 2012 and 2013, subsamples of 20 berries per vine were frozen at –20°C immediately after harvest. When the berries were thawed, their individual length, width, and weight were recorded with an analytical balance (Model E42 SB, Gibertini). Each berry was sliced open using a scalpel, and the seeds, skin, and flesh were removed, separated, and weighed individually to determine if summer pruning treatments influenced the growth of berry components.

Statistical analysis. Basic statistics and analysis of variance were performed using Statistica version 4.3 (StatSoft, Inc.) and Sigma Plot version 10 (SPSS, Inc.). Results were tested for homogeneity of variance and subjected to analysis of variance (ANOVA). Treatment results were compared for each year applying Duncan's multiple range test at $p \leq 0.05$ and 0.01. Values are presented as three-year averages (2011 to 2013) reporting the significance of the treatment and, since year was considered as a random variable, the error term for the treatment factor was the mean square of the year × treatment interaction. We divided the year × treatment interaction to determine the significance of the *F*-test. Variances were homogeneous in all cases, so the effects of year × treatments were tested using the pooled mean square error as an error term (Gomez and Gomez 1984). Finally, we used data collected in 2014 to quantify any carryover effects of treatments applied in earlier growing seasons. These data were tested using means separation calculated by applying the Student-Newman-Keuls test at $p \leq 0.05$ and 0.01.

Results

Summer drought occurred in 2011 and 2012, followed by a more regular distribution of rainfall and the absence of drought in 2013 (Table 1), which enabled a comparison of canopy management methods under different meteorological conditions. Heat accumulation, expressed as growing degree-days (GDD, base 10°C) from budburst to harvest, was similar in 2011 and 2012, and was lower in 2013 (Table 1). Total rainfall from budburst to harvest fluctuated significantly and was lowest in 2011 and highest in 2012; the vast majority of rainfall in 2012 occurred in September, 10 to 20 days before harvest. The distribution of precipitation during the growing periods differed between the years (Table 1). Although heat accumulation was similar, higher summer temperatures occurred in 2011 and 2012. August was warm in all three years. In 2011, there were 53 consecutive days without rainfall. Under these conditions and because vines were not irrigated, symptoms of water stress were observed in 2011 and 2012, with most basal leaves of the shoots showing signs of chlorosis and necrosis. The increased rainfall distribution in 2013 led to a more consistent level of soil water availability during that season with a positive effect on both vine growth and yield.

All vines developed ~30 shoots each with similar total leaf area (Table 2) before shoot thinning was performed each year. This number of shoots was greater than the number of nodes left by winter pruning (seven spurs of two nodes each for a total of 14 buds per vine), largely due to the breaking of both dormant and latent buds. Vine fruitfulness (ratio of inflorescence number to shoots) before treatment was lowest in *St+Dpa* and highest in *St+Dpv+Ct* (Table 2). The *St* treatment required the removal of an average of 16 shoots per vine to obtain the target of 14 per vine and led to the removal of varying amounts of leaf area. Just after treatment, total leaf area differed significantly between *Wp* vines and the other treatments (Table 2).

Most of the shoots that were removed either had developed in undesirable positions or were malformed or fruitless. When necessary, we also removed fertile shoots by eliminating two to three inflorescences per vine. Thinning to leave mainly fruitful shoots had a positive effect on vine fertility (fruitfulness, Table 2). In 2011, the *St* treatment was applied at DOY 150, 15 days later than in 2012 and 2013, with vines characterized by more developed canopies. The *St* treatments

Table 1 Weather variables on a monthly basis, from budburst (April) to harvest (September) in Sangiovese vines. Data are from DOY 97 to DOY 266 in 2011, from DOY 110 to 265 in 2012, and from DOY 100 to 260 in 2013. GDD, growing degree-days.

GDD	**April**	**May**	**June**	**July**	**August**	**September**	**Budburst to harvest**
2011	184	287	398	452	520	425	2140
2012	129	247	448	545	526	345	2076
2013	153	229	350	481	474	333	1877
Precipitation (mm)							
2011	25	13	25	59	0	30	153
2012	17	44	11	16	38	205	329
2013	29	128	61	24	18	60	266
T > 30°C (# days)							
2011	0	1	1	8	13	10	33
2012	0	0	8	18	15	2	42
2013	0	0	4	8	9	0	21

Table 2 Vegetative and fruit characteristics before and after shoot thinning in Sangiovese vines on DOY 150 (30 May), 136 (15 May), and 134 (14 May) in 2011, 2012, and 2013, respectively (n = 18).

	Preshoot thinning[a]				**Postshoot thinning**[a]			
	Shoots/ vine (#)	**TLA/ vine (m^2)**	**Inflor/ vine (#)**	**Fruitfulness**	**Shoots/ vine (#)**	**TLA/ vine (m^2)**	**# Inflor/ vine (#)**	**Fruitfulness**
Treatment (T)[b]								
Wp	28	1.37	15	0.55 ab[c]	26 a	1.37 a	15 a	0.55 c
St	31	1.34	17	0.55 ab	14 b	1.09 b	14 ab	1.03 a
St+Dpa	32	1.36	15	0.49 b	14 b	1.07 b	12 c	0.86 b
St+Dpv	30	1.32	16	0.57 ab	14 b	1.19 b	13 bc	0.93 a
St+Dpv+Ct	29	1.36	18	0.63 a	14 b	1.09 b	14 ab	0.99 a
Signif.	ns	ns	ns	*	**	*	*	**
Year (Y)								
2011	33 a	2.20 a	15b	0.45 a	18 a	1.94 a	12 c	0.75 b
2012	30 b	0.91 b	17a	0.57 b	16 b	0.80 b	15 a	0.98 a
2013	26 c	0.94 b	17a	0.65 c	15 c	0.75 b	14 b	0.88 a
Signif.	**	**	**	**	**	**	**	**
T x Y[d]	ns	ns	*	*	**	*	ns	ns

[a] TLA, total leaf area; Inflor, inflorescence; Fruitfulness, ratio of number of inflorescences to number of shoots.

[b] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; *St+Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning.

[c] Within columns, different letters indicate significant differences between means (Duncan's multiple range test). *, ** significant at $p \leq 0.05$ and $p < 0.01$, respectively; ns, not significant.

[d] The year effect is also shown as seasonal data averaged over all treatments (n = 30).

led to an average removal of 0.26, 0.11, and 0.19 m^2 leaf area/vine in 2011, 2012 and 2013, respectively (Table 2).

Application of *Dpa* between the first and sixth shoot nodes drastically reduced total leaf area on *St* vines, by a three-year average of 0.87 m^2 per vine. This difference was maintained until emergence of the full canopy. The *Dpv* treatment, in which laterals were removed in July on vines that had already undergone the *St* treatment, further reduced total leaf area in *St+Dpv* and *St+Dpv+Ct* vines (Table 3).

St reduced the LLN, mostly in the fruiting zone (fz) 40 cm from the cordon. After *St*, LLNfz values 20 and 40 cm from the cordon were ~1.91 and 1.59, respectively, compared to 2.83 and 1.73 in *Wp* vines (Figure 1). The difference between treatments was maintained until full canopy development. The *Dpa* treatment caused a further abrupt decrease in LLNfz to 0.07 and 0.27 at 20 and 40 cm from the cordon, respectively, which at full canopy were 0.50 and 1.32 (Figure 1). The *Dpv* treatment reduced LLNfz to 1.09 and 2.24, respectively. Over the three years, LLN measured at 60 cm from the cordon did not differ significantly between the canopy treatments, and the differences continued to narrow at 80 and 100 to 120 cm above the vines until they were almost equal among the treatments (Figure 1).

In contrast to 2012 and 2013, leaves on *St+Dpa* vines in 2011 showed a significant increase in photosynthesis (Pn) compared to *St* vines just after treatment and were 8, 14, and 12% greater at DOY 160, 180, and 197, respectively (Figure 2). In 2011, photosynthetic compensation due to leaf removal began 10 days after treatment (DOY 160) and continued until DOY 197; then from mid-July, Pn decreased (likely due to drought) and remained uniform between treatments. In 2012 and 2013, Pn in the *St+Dp* treatment was comparable to that in the *St* treatment.

In Sangiovese, a significant treatment × year interaction was found for yield per vine, cluster weight, and berry weight (Table 4). Despite the elimination of ~16 shoots and two to three inflorescences per vine, the *St* treatment not only failed

Table 3 Total leaf area in Sangiovese vines before and after preanthesis defoliation (*Dpa*), preveraison defoliation (*Dpv*), and at harvest (n = 18). In 2011, 2012, and 2013, *Dpa* was performed on DOY 151 (31 May), DOY 143 (22 May), and DOY 140 (20 May), respectively; *Dpv* was performed on DOY 199 (18 July), DOY 200 (18 July), and DOY 206 (25 July); harvest took place on DOY 199 (18 July), DOY 200 (18 July), and DOY 206 (25 July).

	Total leaf area (m^2)				
	Rapid shoot elongation phase anthesis		Full canopy vine veraison		
	Predef.[a]	Postdef.[a]	Predef.	Postdef.	Harvest
Treatment (T)[b]					
Wp	1.68 a	1.68 a[c]	4.04 a	4.04 a	3.21 a
St	1.30 b	1.30 b	3.20 b	3.20 b	3.82 b
St+Dpa	1.30 b	0.43 c	2.59 c	2.59 c	3.34 c
St+Dpv	1.40 b	1.40 b	3.35 b	2.77 bc	3.50 bc
St+Dpv+Ct	1.30 b	1.30 b	3.31 b	2.67 c	3.54 bc
Signif.	**	**	**	*	**
Year (Y)					
2011	1.94 a	1.73 a	3.13 c	2.80 b	2.28 c
2012	1.08 b	0.92 b	3.48 a	3.27 a	2.73 b
2013	1.17 b	1.01 b	3.28 b	3.09 a	3.04 a
Signif.	**	**	**	**	**
T x Y[d]	ns	*	ns	ns	ns

[a] Predef., predefoliation; Postdef., postdefoliation.

[b] *Wp*, winter pruning; *St*, shoot thinning; *Ct*, cluster thinning.

[c] Within columns, different letters indicate significant differences between means (Duncan's multiple range test). *, ** significant at $p \leq 0.05$ and $p < 0.01$, respectively; ns, not significant.

[d] The year effect is also shown as seasonal data averaged over all treatments (n = 30).

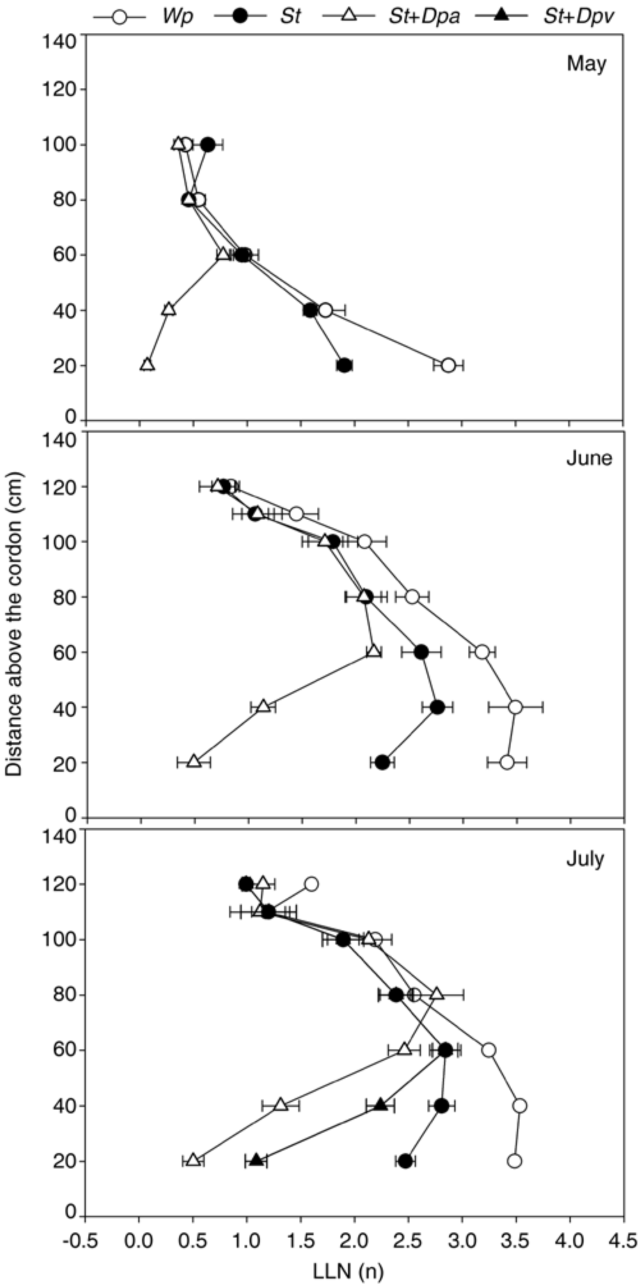

Figure 1 Seasonal evolution of leaf layer number (LLN) recorded at three stages of vegetative growth from 2011 to 2013 in Sangiovese vines subjected to different canopy management treatments. *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation. Each data point is the mean ± standard error of six vines.

to reduce yield per vine, but it generated the heaviest crop compared to the other treatments, except for *Wp* in the first year (Tables 4 and 5). Despite having lower cluster numbers than *Wp*-only vines, *St* vines had increased cluster weight (+24%) and number of berries per cluster (+19%) (Table 4). The *St+Dpa* and *St+Dpv+Ct* vines produced the lowest yield in all three years. The significant yield reduction in *St+Dpa* compared to *St* (–34%) was due to smaller clusters with fewer and smaller berries. The *St+Dpv* treatment did not induce substantial changes in yield per vine compared to *Wp* and *St* vines; however, addition of the *Ct* treatment significantly reduced the yield per vine compared to *Wp*, *St*, and *St+Dpv* (Table 4). Overall, vines showed more abundant yield in the last year (2013), with ~5 kg of grapes per vine composed of heavier clusters with larger and more numerous berries compared to the other two years (Tables 4 and 5).

The lowest yields in the *St+Dpa* and *St+Dpv+Ct* treatments were also affected by minor shortening of the first rachis branch. Cluster compactness (expressed as weight/length) was significantly higher in *St* vines, which contributed to increasing the incidence of *B. cinerea* compared to *St+Dpa* and *St+Dpv* treatments. *St* vines also had the greatest leaf area in the fruiting zone and had a lower incidence of sunburn (Table 6).

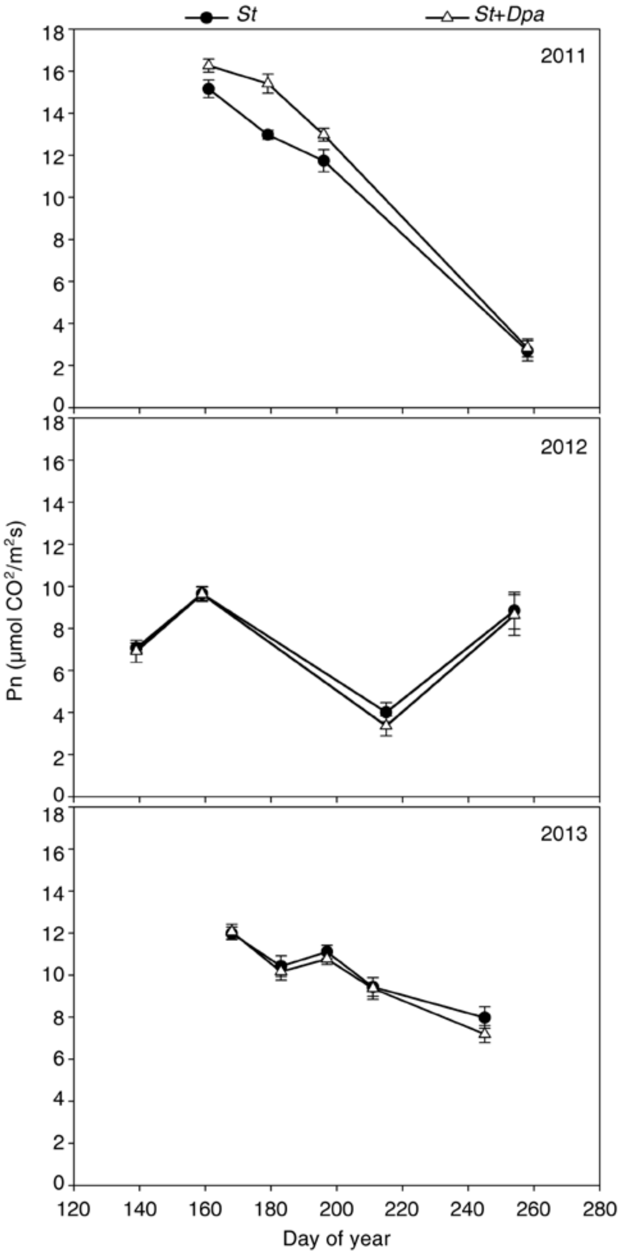

Figure 2 Seasonal evolution of net photosynthesis from 2011 to 2013 in Sangiovese vines subjected to shoot thinning (*St*) and shoot thinning and pre-anthesis defoliation (*St+Dpa*). Each data point is the mean ± standard error of six measurements.

Table 4 Yield components, cluster, and berry characteristics recorded from 2011 to 2013 in Sangiovese vines subjected to different canopy management treatments.

	Clusters/ vine (#)	Yield/ vine (kg)	Cluster weight (g)	Berry weight (g)	Berries/ cluster (#)
Treatment (T)[a]					
Wp	18 a[b]	3.9 a	219 b	2.1 ab	104 b
St	15 b	4.1 a	271 a	2.2 a	123 a
St+Dpa	14 b	2.7 b	196 b	1.9 b	103 b
St+Dpv	14 b	3.6 a	234 ab	2.0 b	117 ab
St+Dpv+Ct	12 c	2.6 b	210 b	2.1 ab	100 b
Signif.	**	**	**	*	**
Year (Y)					
2011	13 b	1.6 c	124 c	1.8 b	65 c
2012	16 a	3.2 b	197 b	1.7 c	114 b
2013	15 a	5.3 a	356 a	2.7 a	133 a
Signif.	**	**	**	**	**
T x Y[c]	ns	*	**	**	ns

[a] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 18).

[b] Within columns, different letters indicate significant differences between means (Duncan's multiple range test). *, ** significant at $p \leq 0.05$ and $p < 0.01$, respectively; ns, not significant.

[c] The year effect is also shown as seasonal data averaged over all treatments (n = 30).

Table 5 Yield and cluster weight recorded at harvest in Sangiovese vines subjected to different canopy management treatments. Harvest dates in 2011, 2012, and 2013 were DOY 199 (18 July), DOY 200 (18 July), and DOY 206 (25 July), respectively.

	Yield/vine (kg)			Cluster weight (g)		
Treatment[a]	**2011**	**2012**	**2013**	**2011**	**2012**	**2013**
Wp	2.5 a[b]	3.5 ab	5.6 a	157 a	183 b	316 b
St	1.6 b	4.1 a	6.7 a	123 ab	239 a	452 a
St+Dpa	1.3 b	2.7 b	4.1 b	91 b	175 b	321 b
St+Dpv	1.5 b	3.4 ab	5.9 a	135 ab	211 ab	355 b
St+Dpv+Ct	1.1 b	2.5 b	4.1 b	115 ab	178 b	336 b
Signif.	*	*	*	*	*	*

[a] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 6).

[b] Within columns, different letters indicate significant differences between means (Student Newman Keuls test). *, significant at $p \leq 0.05$.

Berry growth components showed that skin weight was greatest in *St+Dpa* vines, while *St* vines produced the greatest flesh weight (Table 4). Berry size and relative skin and flesh growth differed between treatments; *St+Dpa* vines produced berries with thicker skin, and consequently, the greatest skin to-flesh ratio (Table 7).

The *St* treatment did not improve berry composition at harvest; sugar concentrations, pH, TA, YAN, and polyphenol values in *St* treatments were similar to those of *Wp*, but anthocyanin concentrations were lowest. Leaf removal at preanthesis on vines that had already been treated with *St* led to greater must soluble solids (25 Brix), malic acid degradation, and anthocyanin and polyphenol concentrations compared to *St*. Compared to *Wp* vines, *St+Dpa* vines had greater soluble solids and polyphenol concentrations (Table 8).

Sugar accumulation declined from 2011 to 2013 (Table 9). *St+Dpa* and *St+Dpv+Ct* vines had greater sugar concentrations at harvest than did the other treatments in all years. Significant differences in TA occurred in cluster-thinned vines (*St+Dpv+Ct*), in which the lowest average TA (6.2 g/L) occurred. There were no significant differences among treatments in tartaric acid or YAN, and malic acid decreased in all leaf-removal treatments (Table 8). TA was greatest in 2013 for each treatment (Table 9).

A significant year × treatment interaction was found for must soluble solids and pH because of higher values in 2011 and 2012 compared to 2013; significant interactions for TA were caused by lower values in 2011 and 2012 (Tables 8 and 9).

None of the canopy management treatments had a significant influence on the leaf area-to-yield ratio (Table 10). Shoot-thinning treatments caused a predictable difference in the number of canes per vine: *Wp* vines had an average of 18 canes each; in contrast, the additional treatments reduced that number to an average of 13 per vine. During the three years, *St+Dpv+Ct* vines developed heavier canes, with average pruning weight that was 18 and 24% higher than that of *Wp* and *St+Dpa* vines, respectively. Cluster thinning, which reduced yield, increased the Ravaz index (Table 10).

In 2014, all vines were winter-pruned to seven spurs of two nodes each, and no other treatment was applied. After budbreak, the vines had an average of 25 to 29 shoots per vine, and the number of inflorescences per shoot ranged from 0.58 to 0.79 (Table 11). Carryover effects on yield were observed in vines that had been defoliated preanthesis. In 2014, these

Table 7 Berry characteristics in Sangiovese vines subjected to different canopy management treatments.

	Berry weight (g)	Skin weight (mg)	Flesh weight (g)	Seed weight (mg)	Seeds/ berry (#)	Skin-to-flesh ratio (%)
Treatment (T)[a]						
Wp	2.46 ab[b]	159 b	2.19 b	113	3.0	7.5 b
St	2.66 a	164 b	2.37 a	119	3.0	7.1 b
St+Dpa	2.47 ab	181 a	2.18 b	116	3.0	8.5 a
St+Dpv	2.33 b	147 b	2.08 b	108	2.8	7.4 b
St+Dpv+Ct	2.48 ab	165 b	2.20 b	111	2.9	7.6 b
Signif.	*	*	*	ns	ns	**
Year (Y)						
2012	1.99 b	140 b	1.75 b	106 b	2.9	8.2 a
2013	2.97 a	187 a	2.66 a	121 a	3.0	7.1 b
Signif.	**	**	**	**	ns	**
T x Y[c]	ns	ns	ns	ns	ns	ns

[a] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 12).

[b] Within columns, different letters indicate significant differences between means (Duncan's multiple range test). *, ** significant at $p \leq 0.05$ and $p < 0.01$, respectively; ns, not significant.

[c] The year effect is also shown as seasonal data averaged over all treatments (n = 30).

Table 6 Cluster morphology and incidence of rot or sunburn in Sangiovese vines subjected to different canopy management treatments.

			Cluster compactness		*Botrytis cinerea* (%)		Sunburn (%)	
	Cluster length (mm)	Branching length (mm)	Weight/length (g/mm)	OIV rating[a]	Diff.[a]	Inc.[a]	Diff.[a]	Inc.[a]
Treatment (T)[b]								
Wp	154 ab[c]	79 b	2.0 bc	7.4	24.2 ab	4.6 ab	16.7	3.6 ab
St	163 a	94 a	2.3 a	7.9	30.0 a	6.7 a	19.2	1.6 b
St+Dpa	148 b	61 c	1.8 c	7.3	11.7 b	1.7 b	35.8	6.1 a
St+Dpv	155 ab	84 b	2.1 b	7.8	13.3 b	3.7 ab	22.5	4.1 ab
St+Dpv+Ct	150 b	53 c	1.8 bc	7.7	19.2 ab	2.5 ab	19.2	3.4 ab
Signif.	*	**	**			*	ns	*
Year (Y)								
2012	144 b	69 b	1.6 b	6.7 b	20.3	4.4	18.0	4.2
2013	164 a	79 a	2.4 a	8.5 a	19.0	3.3	27.3	3.3
Signif.	**	**	**	**	ns	ns	ns	ns
T x Y[d]	ns	ns	*	ns	ns	ns	ns	ns

[a] OIV, cluster compactness visually estimated using Organisation Internationale de la Vigne et du Vin (OIV) code 204 (OIV 1983); Diff., *Botrytis cinerea* and sunburn diffusion on clusters; Inc., *B. cinerea* and sunburn incidence on clusters.

[b] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 12).

[c] Within columns, different letters indicate significant differences between means (Duncan's multiple range test). *, ** significant at $p \leq 0.05$ and $p < 0.01$, respectively; ns, not significant.

[d] The year effect is also shown as seasonal data averaged over all treatments (n = 30).

vines had the lowest yield (5 kg). Their yield reduction (–29%) compared to ex-*St* (i.e., former or previously [in 2011-2013] *St*-treated vines) was mainly due to lower cluster numbers and fewer berries per cluster. There were no significant differences in average berry weight (Table 11).

Even in 2014, the ex- *St+Dpa* vines produced clusters with a significantly shorter first rachis branch, thereby maintaining the differences found in the earlier trial years, and generating decreased cluster compactness (Table 12). In 2014, the ex-*St* and *Wp* vines had similar total soluble solids, pH, TA, and polyphenols, and *St* had lower anthocyanin concentrations. Compared with ex-*St* vines, ex-*St+Dpa* produced more soluble solids and anthocyanins in 2014 (Table 13).

Discussion

Prior research suggested that shoot thinning, regardless of timing, can reduce the number of leaf layers in the canopy (Reynolds et al. 1994). Here, effects of shoot thinning were generally consistent with that finding, in that leaf area at anthesis and at full canopy development was restricted in *St* treatments. These effects were observed mainly in the fruit zone, where the density values (LLNfz), especially at full canopy, were significantly lower than those of the vines treated only with *Wp*. The *St* treatment also had positive effects on canopy microclimate as a result of increased sunlight penetration. This is consistent with the findings of Reynolds et al. (2005) who reported that later-season *St* treatments allowed for more sunlight penetration to the canopy interior compared to earlier thinning in Pinot noir and Cabernet franc. In contrast, in another trial on young Sangiovese vines subjected to shoot thinning (Myers et al. 2008), the primary leaves and laterals provided significant growth compensation. Bernizzoni et al. (2011) reported that shoot-thinning treatments produced full compensatory vegetative growth, reduced yield, and improved must composition in potted and mature field-grown Barbera vines compared to control vines. In our study, compared to *Wp*, *St* failed to reduce yield capacity, improve juice composition (soluble solids, TA, and pH), or increase polyphenol concentrations. These results are similar to those of Bravetti et al. (2012), who performed a similar study using

Table 8 Grape composition at harvest in Sangiovese vines subjected to different canopy management treatments.

	Soluble solids (Brix)	pH	Titratable acidity (g/L)	Tartaric acid (g/L)	Malic acid (g/L)	Anthocyanins (mg/kg)	Polyphenols (mg/kg)	YAN[a] (mg/L)
Treatment (T)[b]								
Wp	24.1 b[c]	3.46	6.4 ab	8.53	1.95 ab	721 ab	2208 b	121
St	24.0 b	3.43	6.5 ab	8.58	2.06 a	615 c	2191 b	108
St+Dpa	25.0 a	3.45	6.9 a	8.74	1.86 bc	769 a	2488 a	113
St+Dpv	24.0 b	3.42	6.8 a	8.90	1.77 cd	694 b	2193 b	109
St+Dpv+Ct	25.3 a	3.49	6.2 b	8.12	1.64 d	695 b	2317 ab	111
Signif.	**	ns	*	ns	*	**	*	ns
Year (Y)								
2011	27.6 a	3.69 a	5.9 b	10.36 a	1.96 b	681 b	2706 a	182 a
2012	23.8 b	3.35 b	6.0 b	8.10 b	1.255 c	854 a	2754 a	77 b
2013	22.0 c	3.30 c	7.7 a	7.27 c	2.35 a	561 c	1378 b	78 b
Signif.	**	**	**	*	*	**	**	**
T x Y[d]	*	*	*	ns	ns	ns	ns	*

[a] YAN, yeast assimilable nitrogen.

[b] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 18).

[c] Within columns, different letters indicate significant differences between means (Duncan's multiple range test). *, ** significant at $p \le 0.05$ and $p < 0.01$, respectively; ns, not significant.

[d] The year effect is also shown as seasonal data averaged over all treatments (n = 30).

Table 9 Total soluble solids, pH, and titratable acidity (TA), recorded at harvest in Sangiovese vines subjected to different canopy management treatments. Harvest dates in 2011, 2012, and 2013 were DOY 199 (18 July), DOY 200 (18 July), and DOY 206 (25 July), respectively.

	Soluble solids (Brix)			pH			Titratable acidity (g/L)		
Treatment[a]	**2011**	**2012**	**2013**	**2011**	**2012**	**2013**	**2011**	**2012**	**2013**
Wp	26.7 c[b]	23.7 ab	21.8 b	3.69	3.42 a	3.28 b	5.5 b	5.8 ab	8.0 a
St	27.2 bc	23.7 ab	21.1 b	3.68	3.35 ab	3.26 b	6.0 ab	5.7 b	7.9 a
St+Dpa	28.0 a	24.1 a	23.0 a	3.68	3.31 b	3.35 a	6.4 a	6.4 a	7.8 a
St+Dpv	27.8 ab	23.1 b	21.0 b	3.69	3.33 b	3.25 b	5.9 ab	6.4 a	8.0 a
St+Dpv+Ct	28.4 a	24.3 a	23.1 a	3.73	3.36 ab	3.38 a	5.9 ab	5.9 ab	6.9 b
Signif.	*	*	*	ns	*	*	*	*	*

[a] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 6).

[b] Within columns, different letters indicate significant differences between means (Student Newman Keuls test). *, significant at $p \le 0.05$; ns, not significant.

Montepulciano vines; however, in contrast to Bravetti et al. (2012), our *St* clusters were more compact than those of *Wp* vines (Table 3).

Photosynthesis decreased over time and in inverse proportion to increasing summer temperatures and water stress (Figure 2). In 2012, low temperatures occurred during the last 10 days of May and in the early summer, resulting in limited photosynthetic capacity. Compared to 2011 and 2013, the first measurement in 2012 was very low because it was taken in May on very young leaves (~20 days old). The canopy in the *St+Dpa* treatments did not respond to the loss of vegetative growth and photosynthetic capacity enough to compensate and generate a final canopy similar to that of the *St* and *Wp* vines. This is contrary to observations in regularly irrigated, potted Sangiovese grapevines in which stronger lateral formation led to significant leaf compensation (Poni et al. 2006). Earlier work on non-irrigated, field-grown Sangiovese (Intrieri et al. 2008) and Trebbiano (Poni et al. 2006) similarly showed lower rates of lateral development in shoots subjected to *Dpa*; in both studies, water availability was thought to be the crucial factor in determining the grapevine response to preanthesis defoliation. Here, the lack of compensation cannot be attributed to water availability; the soil water status was especially optimal in 2013, and conditions were favorable for photosynthesis in all years.

Table 10 Vegetative and pruning characteristics recorded from 2011 to 2013 in Sangiovese vines subjected to different canopy management treatments. Harvest dates in 2011, 2012, and 2013 were DOY 199 (18 July), DOY 200 (18 July), and DOY 206 (25 July), respectively.

	Leaf area/ yield (m^2/kg)	Cane/ vine (#)	Pruning weight (g)	Ravaz index (kg/kg)
Treatment (T)[a]				
Wp	1.25	18 a[b]	47.5 b	4.49 a
St	1.20	13 b	56.6 ab	5.26 a
St+Dpa	1.81	13 b	44.3 b	4.51 a
St+Dpv	1.23	13 b	55.9 ab	4.69 a
St+Dpv+Ct	1.78	13 b	58.1a	3.26 b
Signif.	ns	**	*	*
Year (Y)				
2011	2.46 a	14 b	48 b	2.45 c
2012	1.19 b	15 a	48 b	4.47 b
2013	0.72 b	14 b	62 a	6.40 a
Signif.	**	**	**	**
T x Y[c]	ns	ns	ns	ns

[a] *Wp*, winter pruning; *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 18).

[b] Within columns, different letters indicate significant differences between means (Duncan's multiple range test). *, ** significant at $p \leq 0.05$ and $p < 0.01$, respectively; ns, not significant.

[c] The year effect is also shown as seasonal data averaged over all treatments (n = 30).

Despite having no effect on final berry size, the *St+Dpa* treatment improved skin development, as reported for Barbera by Poni and Bernizzoni (2010). In the *St+Dpa* treated vines, skin growth was favored by the improved fruiting zone microclimate produced by defoliation, indicating that cell division in the pericarp and exocarp may be sensitive to temperature, as reported by Poni et al. (2009). The improved climatic condition in the fruiting zone of *St+Dpa* vines also led to a significant reduction in the incidence and diffusion of *Botrytis* on clusters (Palliotti et al. 2011, Gatti et al. 2015), but here, these clusters were more susceptible to sunburn.

The *St+Dpa* treatment led to a 48% reduction in yield in 2011 and to a ~25% reduction in 2012 and 2013 compared to *Wp* vines, showing that it is an effective tool for regulating yield. The *St+Dpa* treatment also led to a significant increase in must total soluble solids, as previously shown by Poni et al. (2006, 2009), Intrieri et al. (2008), Palliotti et al. (2011), Bravetti et al. (2012), and Gatti et al. (2015). The soluble solids concentration in *St+Dpa* grapes may have been influenced by compensatory photosynthesis in the retained leaves, laterals, and newly developing shoots, as shown by Poni et al. (2006, 2009) in potted grapevines. However, in our study, the capacity of leaves to temporarily compensate for reduced leaf area (Candolfi-Vasconcelos and Koblet 1990) was observed only in the first year and for only ~40 days after defoliation.

TA and tartaric acid were unaffected by the treatments. In contrast, malic acid concentrations were lower in all defoliated vines (*St+Dpa*, *St+Dpv*, and *St+Dpv+Ct*) (Table 6); this was probably related to increased exposure of fruit to sunlight, causing high berry temperatures as a result of leaf removal and lower LLN in the fruiting zone.

Table 11 Vegetative and yield components recorded in 2014 in Sangiovese vines subjected to winter pruning only (*Wp*) (n = 6).

Treatment		Shoots/vine		Clusters/vine	Yield/vine	Cluster wt	Berry wt	Berries/cluster
2014	2011–2013[a]	(#)	Fruitfulness	(#)	(kg)	(g)	(g)	(#)
Wp	*Wp*	28	0.69 ab[b]	19 a	7.0 a	351	3.0	117 ab
Wp	*St*	29	0.63 ab	18 ab	7.0 a	385	3.1	124 a
Wp	*St+Dpa*	26	0.58 b	15 c	5.0 b	321	3.2	100 b
Wp	*St+Dpv*	25	0.65 ab	16 bc	5.2 b	331	3.0	110 ab
Wp	*St+Dpv+Ct*	25	0.79 a	20 a	7.1 a	355	3.2	111 ab
Signif.		ns	*	*	*	ns	ns	*

[a] *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 6).

[b] Within columns, different letters indicate significant differences between means (Student Newman Keuls test). *, significant at $p \leq 0.05$; ns, not significant.

It is likely that lower temperatures in 2012 and 2013 between budburst (DOY 110 and 100, respectively) and flowering (DOY 149 and 147, respectively) impeded canopy photosynthesis, and that shoot growth was sustained by carbohydrate reserves. Additionally, in April and May of 2012 and 2013, more precipitation, lower than normal weekly temperatures, and decreased available sunlight may have occurred, although these parameters were not measured.

Preveraison defoliation reduced total leaf area but, consistent with Bravetti et al. (2012), did not reduce yield per vine. It did, however, cause a lowering of the leaf-to-fruit ratio, which nonetheless remained above the threshold considered sufficient to ensure optimal grape ripening (Kliewer and Dokoozlian 2005). Preveraison defoliation reduced the canopy density in the fruiting zone, but with different results compared to those observed in cv. Ortrugo (Gatti et al. 2015). The *St+Dpv* treatments failed to produce significant differences in juice composition and berry color compounds compared to *Wp* vines (Table 5), matching observations in Montepulciano by Bravetti et al. (2012).

According to earlier studies (Susaj et al. 2013, Gatti et al. 2015), preveraison cluster thinning significantly lowers yield and increases sugar concentrations. Here, the *St+Dpa* and *St+Dpv+Ct* treatments produced those same results, but the significant increase in anthocyanin and polyphenol concentrations in *St+Dpv+Ct* did not occur in *St+Dpa*. Cluster thinning in combination with shoot thinning and preveraison defoliation led to further yield reduction, and these vines had the lowest Ravaz index value (yield/pruning weight).

The vegetative and reproductive responses of high-yielding red grape varieties such as Sangiovese change according to water availability during the season. Environmental conditions in 2011 influenced vine yield capacity, but significant reductions in yield occurred, mainly as a result of reduced fruit set due to preanthesis defoliation. The lack of available water in the soil coupled with high temperatures during berry development, as occurred in 2011, likely reduced cell expansion and triggered berry dehydration as a result of greater transpiration rates (Greenspan et al. 1994), producing increases in soluble solids as reported in other environments (Lanari et al. 2014).

The lowest levels of TA, observed in 2011 and 2012, were due to excessive degradation of malic acid caused by summer drought conditions that occurred during the period of cluster ripening. In contrast, high levels of TA were measured in 2013, when abundant, frequent rainfall and lower maximum air temperature occurred.

Carryover effects in year one post-trial. We were interested in examining whether any effects might carry forward from the three years of treatment to the following year (2014). Shoot numbers and vine fruitfulness were similar among treatments in 2014, except for vines subjected to *St+Dpv+Ct*, in which these values were greater (Table 11).

In vines that had been subjected specifically to preanthesis defoliation (ex-*St+Dpa*), yield and cluster morphology

Table 12 Cluster morphology and incidence of rot recorded in 2014 in Sangiovese vines subjected to winter pruning only (*Wp*) (n = 6).

				Cluster compactness		***Botrytis cinerea* (%)**	
2014	**2011–2013**[a]	**Cluster length (mm)**	**Branching length (mm)**	**Weight/length (g/mm)**	**OIV rating**[b]	**Diff.**[b]	**Inc.**[b]
Wp	*Wp*	184	103 a[c]	1.60 ab	8.1	33.3	4.4
Wp	*St*	184	97 a	1.90 a	8.2	16.7	3.4
Wp	*St+Dpa*	167	69 b	1.52 b	7.9	23.3	4.4
Wp	*St+Dpv*	172	92 ab	1.54 b	7.3	23.3	3.1
Wp	*St+Dpv+Ct*	180	103 a	1.65 ab	7.6	21.7	4.1
Signif.		ns	*	*	ns	ns	ns

[a] *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 6).

[b] OIV, cluster compactness visually estimated using Organisation Internationale de la Vigne et du Vin (OIV) code 204 (OIV 1983); Diff., *Botrytis cinerea* and sunburn diffusion on clusters; Inc., *B. cinerea* and sunburn incidence on clusters.

[c] Within columns, different letters indicate significant differences between means (Student Newman Keuls test). *, significant at $p \le 0.05$; ns, not significant.

Table 13 Grape composition at harvest recorded in 2014 in Sangiovese vines subjected to winter pruning only (*Wp*) (n = 6).

Treatment						
2014	**2011–2013**[a]	**Soluble Solids (Brix)**	**pH**	**Titratable acidity (g/L)**	**Anthocyanins (mg/kg)**	**Polyphenols (mg/kg)**
Wp	Wp	20.1 ab[b]	3.28 ab	7.9 ab	579 ab	2862 a
Wp	*St*	18.9 b	3.36 b	8.1 a	433 d	2600 ab
Wp	*St+Dpa*	20.3 a	3.34 a	7.5 b	582 ab	2832 a
Wp	*St+Dpv*	20.4 a	3.29 ab	7.6 ab	604 a	262 a
Wp	*St+Dpv+Ct*	19.1 ab	3.31 b	8.1 a	466 cd	2178 b
Signif.		*	*	*	*	*

[a] *St*, shoot thinning; *St+Dpa*, shoot thinning and preanthesis defoliation; *St+Dpv*, shoot thinning and preveraison defoliation; St+*Dpv+Ct*, shoot thinning, preveraison defoliation, and cluster thinning (n = 6).

[b] Within columns, different letters indicate significant differences between means (Student Newman Keuls test). *, significant at $p \le 0.05$.

continued to be positively influenced in 2014, consistent with the trial results. These vines showed a strong carryover effect on primary branching length, which was statistically shorter in these vines than in the other treatments. This difference can be attributed to removal of the first six basal leaves at preanthesis, which triggered several related events. First, it modified the cluster microclimate, resulting in increased solar radiation to the inflorescence and likely leading to degradation of gibberellin, the hormone responsible for cell expansion. This, in turn, partially inhibited elongation of the rachis. In addition, the leaf removal treatments were performed in late May, when the initial flowers are forming inside the hibernating bud, which remains latent until development in the following year. It seems reasonable to assume that the removal of a significant proportion of leaf area can cause metabolic stress to the vines, compromising this critical step and cluster morphology in the following season.

Conclusions

Shoot thinning can be an effective tool for limiting canopy density in medium-vigor grape varieties such as Sangiovese, and in environments like that of central Italy with variable precipitation. However, the use of *St* in combination with *St+Dpa*, gives growers a more powerful strategy that can also modify yield and the fruiting zone microclimate, leading to decreased loss from disease and improved grape quality.

Here, we show that when defoliation is applied preanthesis, it can successfully limit yield not only in the year of application but also in the following year. In fact, our investigation conducted in 2014 on Sangiovese vines that were defoliated from 2011 to 2013 and that were winter pruned only in 2014 confirmed that defoliation limits yield capacity (cluster/vine, yield/vine, and the first branching length). *St+Dpa* also results in a significant increase in sugar concentration, which may or may not be desirable.

St alone, or performed with *St+Dpv*, could be a useful tool for achieving lower yield and improved grape composition. However, using *Wp* only, if done correctly relative to the vigor of the variety, may be sufficient to ensure good yield and adequate quality without the additional costs of summer pruning and thinning interventions.

The *St+Dpa* treatment appears to be the most effective tool when it is necessary to limit crop load, especially of characteristically vigorous grape varieties. With its high potential to reduce yield, including in the following year, this treatment could be used by growers in alternate years to reduce excess yield to a desired or mandated threshold, while containing intervention costs.

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