Bulgarian Journal of Agricultural Science, 19 (No 1) 2013, 96-102
Agricultural Academy

EFFECT OF TIMING OF BASAL LEAF REMOVAL ON YIELD COMPONENTS AND GRAPE QUALITY OF GRAPEVINE CVS CABERNET SAUVIGNON AND PROKUPAC (*VITIS VINIFERA* L.)

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Abstract

BESLIC, Z., S. TODIC and S. MATIJASEVIC, 2013. Effect of timing of basal leaf removal on yield components and grape quality of grapevine cvs Cabernet Sauvignon and Prokupac (*Vitis vinifera* L.). *Bulg. J. Agric. Sci.,* 19: 96-102

The examination of timing of partial defoliation was performed on cultivars Cabernet Sauvignon and Prokupac - Serbian autochtonous wine cultivar. Leaf removal treatments were manually applied at full bloom, at fruit set (3-5 mm berry diameter) and before veraison. The treatments consisted of defoliation of the first six nodes of all the shoots. Results indicate that defoliation carried out during the flowering and 3-5 mm berry diameter considerably influence the cluster structure and grape quality. Partial defoliation in these periods reduced the number of berries per cluster and berry size, resulting in a reduction of yield in both cultivars. Early defoliation increased dry matter content of the must. Defoliation treatments increased the content of total phenols in comparison with control, while the content of total anthocyanins was not significant changed in Canernet Sauvignon. In cv. Prokupac, early defoliation increased content of total phenols (in the full bloom and fruit set treatments) and total anthocyanins (in full bloom treatment). The increase in the content of total phenolics and anthocyanins occurred only in treatments applied at flowering and fruit set.

Key words: defoliation, cluster structure, must quality, anthocyanins, phenols.

Abbreviations: BBCH - Biologische Bundesanstalt, Bundessortenamt and CHemical industry; OIV - Organisation Internationale de la Vigne et du Vin

Introduction

The main aim of viticultural production, especially in growing high quality wine cultivars, is obtainining a desirable composition of grapes. Manipulation of canopy density which will lead to improving of vine microclimate conditions and vine balance is one of the main ways for improving grape quality. Partial removal of leaves from the shoots in the clusterfruiting zone is the standard measure which should primarily improve cluster exposure which prompts accumulation of dry matter in must, anthocyanins and polyphenol compounds in berry skins (Kliewer, 1970; Hunter et al., 1991). Andrade et al. (2005) found that the removal of the basal leaves had a favourable effect on light microclimate in the cluster zone with positive consequences on polyphenol synthesis. The erlier basal leaf removal leads to higher accumulation of aromatic compounds and their precursors in graip (Bubola et al., 2009; Karoglan et al., 2008).

In addition to that, better airing of clusters lowers the degree of damage caused by grey rot (Smart et al. 1990; Gubler et al., 1991). Effect of defoliation mainly depends on its intensity and the time of application. Most commonly, defoliation is carried out just before or after veraison. If carried out within this phase, there

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is no considerable influence on the berry number and berry size, nor the yield per vine.

Early defoliation, carried out within the intensive shoot growing phase, causes the total shoot photosynthesis level to decrease by up to 70%, due to the removal of the photosynthetically active surface. The photosynthetic shock caused in such a manner causes a halt in the sink organs development, which shows in a decreased number of berries within clusters, smaller berry size and change in the skin to pulp ratio (Poni et al., 2005, 2006). Such grape structure changes are related to an increased accumulation of dry matter in must and phenolic content in berry skin. The most prominent changes in the grape and berry structure occur when defoliation is carried out during the phase of intensive berry cell division after the end of flowering. Within that phase, the number of pericarp cell layers is determined and each halt in assimilator inflow results in a decreased cell number. Especially beneficial effects are achieved with the wine cultivars characterized by large clusters and berries. These cultivars, after defoliation, produce smaller and looser clusteres, less yield and more beneficial berry skin to pulp ratio (Intrieri et al., 2008). Prokupac is a vigorous and high yielded cultivar, with medium clusters and medium to large berries. It has large leaves and clusters are often developed in the shadow, witch can influence on berry colour. Control of vigor and finding the proper balance between leaf area and yield are of great importance for the quality of grapes and wine. Grapes are harvested about seven days after cv. Cabernet Sauvignon. Wine is medium bodied to bodied (dependes of grape yeild), ruby colored and rich in tannins.

The objective of this study was to investigate the effects of timing of basal leaf removal on yield components and berry composition of Cabernet Sauvignon and Prokupac in non-irrigated conditions in a Cfb climate.

Materials and Methods

Investigations were conducted during three growing seasons 2008 and 2010. in cvs. Cabernet Sauvignon and Prokupac (*Vitis vinifera* L.). Plantage with Cabernet Sauvignon clone R5 grafted onto Kober 5BB was planted in 1993 in an experimental vineyard at the Experimental Station “Radmilovac”, which belongs to the Faculty of Agriculture, University of Belgrade. The location is in the Sumadija-Velika Morava wine region, characterised as a Cfb climate (Kottek et al. 2006). The vine spacing in plantation was 3 × 1 m and were trained as a double Guyot with a trunk height of 90 cm, foliage height about 130 cm and width about 30 cm. The vines were pruned to a mix of canes and spurs. Cultivar Prokupac (*Vitis vinifera* L.) was examined in the production vineyard in the village Rivica near Irig, Serbia. According to the location, the cultivar belongs to Fruška Gora wine region, characterized as a Cfb climate. The vineyard was established in 2003 with the planting space of 3 × 0.5 m. The trellising system was a spur-pruned Lyra, composed of eigth one-node spurs. The foliage height was about 150 cm and width about 50 cm. Prokupac was grafted onto Kober 5BB.

In both trial, all lateral growth were trimmed consequently during vegetation period.

The experiment was set as a random block design with 20 vines per experimental treatment, each vine representing an observation unit. The vines were tagged and randomly assigned to the following treatments: (a) non-defoliated (control) labelled as K; (b) hand removal of the first six basal leaves at the phenological stage 65 (full flowering: 50% of flowerhoods fallen according to BBCH scale, Lorenz et al. 1994) labelled as I; (c) hand removal of the first six basal leaves at the phenological stage 73 (berries groat sized, ovary diameter varying from 3-5 mm according to BBCH scale, Lorenz et al. (1994) labelled as II; (d) hand removal of the first six basal leaves at the stage 79 (majority of berries touching, according to BBCH scale, Lorenz et al*.,* 1994) labelled as III.

Berry size was monitored by measuring equatorial diameter of 30 randomly chosen berries per treatment using an electronic caliper.

Cluster compactness was visually estimated using OIV (Organisation Internationale de la Vigne et du Vin) code 204 (OIV 1983), which ranks “berries in grouped formation with many visible pedicels” as 1 and “misshaped berries” as 9.

After the grape harvest, a representative sample of 3-5 kg of grape was taken from each experimental version and used for must and berry epidermis chemical analysis. Must quality was determined from representative samples during the grape harvest. Concentration of Total Soluble Solids (°Brix) was determined by a hand-held refractometer, Milwaukee MR200ATC, USA. Titratable acidity (TA) was measured by titration with 0.1 N NaOH to a pH 8.2 end point. Total anthocyanins and phenolics were determined by UV/Vis-spectrophotometry (T60 V, PG Instrument Limited). Six samples of grapes from each treatment were collected when soluble solids in must reached 20-22%. Each sample consisted about 200 berries.The samples were collected in black plastic begs and immediately stored at 4°C. All berries from collected cluster were removed and placed in sealed plastic vessels and stored at -20°C. Before analysis, frozen berries were thawed in a refrigerator at 4°C. The berry skins from randomly selected 50 berries from each sample were then peeled with tweezers, freeze-dried and ground with a laboratory mill. For sample extraction, 1 g of ground berry skin was placed in 10 ml tube with 10 ml of 50% v/v aqueous methanol which was adjusted to pH 2.0 with 1.0 M HCl, mixed and incubated for least 3 hours. The homogenate/methanol mixture was centrifuged at 9500 rpm for 20 minutes and a supernatant was decanted for absorbance. Total anthocyanin content was determined at 530 nm and total phenolic content at 740 nm, after a reaction with a Folin-Ciocalteu reagent.

Results and Discussion

Both cultivars' berry development followed a typical double sigmoid curve. This means that berries develop in two phases, separated by a phase of slow growth – lag phase (Coombe, 1992) (Figure 1a, b). It may be noted that the reduction in the assimilative quantity during the first 3-4 weeks of berry development, caused by early defoliation, reduced berry size in treatments I and II compared to treatment III and control. This was maintained until the harvest. Irreversible reduction of berry size occurred due to a decreased assimilative inflow in the period of rapid berry development (Mullins et al., 1992). This stage lasted between 3 and 4 weeks after flowering and during this phase berries grew both through a cell division and cell enlargement (Coombe, 1992). No further cell division occurs after this period. At this stage, berry enlargement is very sensitive to assimilative supply because it requires intense biosynthesis of structural and osmotic compounds and enzymatic machinery, which are energy dependent (Ollat and Gaudillere, 1998) (Figure 1a, b).

Defoliation carried out in flowering phase (stage 65) on cv. Prokupac, and flowering (stage 65) and fruit set (stage 73) on Cabernet Sauvignon caused statistically significant differences in the number of berries per cluster due to the decrease in the degree of fully developed berries (Table 1). This caused a decrease in cluster compactness during the first two defoliation terms in Prokupac and Cabernet Sauvignon. Number of berries in cluster was significant decreased for 56% (treatment I) and for 32% (treatment II) in comparison with non defoliated vines (K) in cv. Prokupac. Partial defoliation – treatment III had no effect nor the number of berries in cluster nor tde cluster weight.

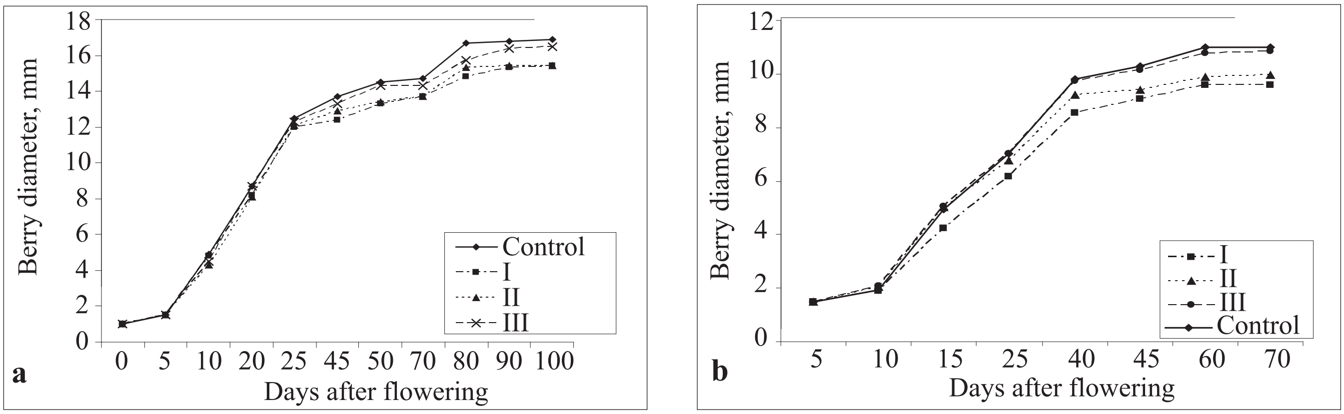

Fig. 1a, b. Changes in berry diameter (mm) on cvs Cabernet Sauvignon and Prokupac depending of timing of partial defoliation during 2009 growing season

The carbohydrate supplay at anthesis is a primary determinant of fruit set (Caspari and Lang, 1996). The decreased assimilation surface during the phenological stages of flowering and frut set resulted in a small number of set berries, as the consequence of the increased degree of flower abortions which has also been confirmed by other authors (Clingeleffer et al., 2001; Poni et al., 2005; Intrieri et al., 2008).

Since the yield per vine depends on the number of clusters, as well as on the berry number and size, yield fluctuations per experiment treatment were expected. In treatments I and II, when the berry number and berry weight decreased the yield and average cluster weight (Table 1) also decreased, which was in accordance with similar examinations of Petrie et al. (2003) and Poni et al. (2006). In cv. Cabernet Sauvignon, the cluster weight reduction was due to reduced berry number rather than berry weight in treatments I and II. In cv. Prokupac, the reduction of cluster weight and yield was due to both factors, berry number and berry weight. In this cultivar, partial leaf removal in treatments I and II reduced berry weight by 18% and 8.8% respectively, when compared to control.

Influence of defoliation time on change in the skin to pulp ratio was not observed within the present experiment. This is in accordance with the study of Matthews (2007), where the berry tissue grows simultaneously and proportionally to the berry size during their developement.

The content of soluble solids in must was significantly higher in treatments I and II compared to treatment III and the control in both cultivars (Table 2). It is in accordance with the findings of Poni et al. (2005), which have

Table 1
Yield, grape and berry structure as affected by the timing of defoliation in cvs. Cabernet Sauvignon and Prokupac (2008/2010)

Variety	Treatment	Yield, kg per vine	Cluster weight, g	Berry number per cluster	Berry weight, g	Cluster compactness (OIV code 204)	Skin to pulp ratio
Cabernet Sauvignon	I	0.78^{a}	77.3^{a}	86^{a}	1.14^{a}	5	0.165^{ns}
	II	0.99^{ab}	96.4^{b}	97^{a}	1.15^{a}	5	0.181
	III	1.23^{bc}	117.1^{c}	105^{ab}	1.32^{b}	5-7	0.175
	k	1.39^{c}	124.1^{c}	121^{b}	1.42^{b}	5-7	0.173
	Lsd$_{0.05}$	0.392386	17.3775	22.3934	0.168351		0.0289915
Prokupac	I	0.97^{a}	142.0^{a}	65^{a}	2.33^{a}	3	0.046^{ns}
	II	1.79^{b}	303.0^{b}	102^{b}	2.59^{ab}	3-5	0.056
	III	2.54^{c}	369.5^{bc}	146^{c}	2.63^{bc}	7	0.050
	k	2.25^{c}	417.3^{c}	149^{c}	2.84^{c}	7	0.044
	Lsd$_{0.05}$	0.43224	79.9711	19.2311	0.242041	-	0.0123611

Means separated by LSD multiple range test ($p \le 0.05$). Data followed by same letter in each column are not significantly different.

Table 2
Influence of defoliation time on the must and berry skin chemical composition in cv. Cabernet Sauvignon and Prokupac (2009/2010)

Variety	Treatment	Soluble solids, Brix %	Total acids, g l^{-1}	Total anthocyanins, mg g^{-1}FW	Total phenols, mg l^{-1} GAE
Cabernet Sauvignon	I	24.8^{a}	7.6^{a}	18.10^{a}	839.7^{a}
	II	23.9^{ab}	8.0^{b}	17.90^{a}	828.9^{a}
	III	23.1^{b}	8.2^{b}	17.46^{a}	802.5^{ab}
	k	23.3^{b}	8.2^{b}	16.84^{a}	702.7^{b}
	Lsd$_{0.05}$	0.753331	0.278655	1.86553	123.848
Prokupac	I	23.5^{a}	7.0^{a}	6.35^{a}	632.4^{a}
	II	23.3^{a}	7.0^{a}	5.69^{b}	573.1^{b}
	III	22.6^{b}	7.1^{a}	5.61^{b}	524.8^{c}
	k	22.2^{b}	7.3^{b}	5.69^{b}	518.13^{c}
	Lsd$_{0.05}$	0.612985	0.184976	0.65261	28.935

Means separated by LSD multiple range test ($p \le 0.05$). Data followed by same letter in each column are not significantly different shown that the removal of source leaves and increased sugar in berries is at odds with a classic source/sink relationship. However, authors suggests that an early defoliation treatment influences an increased development of lateral shoots, whose younger leaves have a higher photosynthetic activity during the ripening period. Similar results of the influence of an early leaf removal on the total leaf area compensation were reported by Tardaguila et al. (2008). Also, previous studies have shown that early partial removal of leaves from shoots directs assimilator transportation to clusters (Koblet et al. 1993).

Leaf removal decreased titratable acidity in treatment I in Prokupac, and in all defoliation treatments in Cabernet Sauvignon. It is in accordance with other studies, were greater fruit exposure resulted in a higher fruit temperature and a faster rate of malic acid metabolism (Lakso and Kliewer, 1975). In the present study, we did not investigate the quality of wines, and it is not clear if a reduction in acidity of 4.1% in treatment I on Prokupac, and 7.3% on Cabernet Sauvignon would have affected wine quality. We believe that this slight decrease of titratable acidity would not affect the balance and freshness of the wine. In support of this are studies of Hunter et al. (1991 and 1995), where meaningful increases in both wine composition and wine sensory data were found when vines were partially defoliated.

Chemical analysis of berry skins indicated that a significantly higher content of total anthocyanins was recorded only in treatment I, in Prokupac, and total phenols in treatments I and II in both cultivars (Table 2). Results indicated that early defoliation had more effect in Prokupac in both total anthocyanins and total phenolics. Prokupac is a high yielding cultivar with compact, large clusters and medium to large berries. Based on the results of cluster structure analysis (Table 1), we can say that the effect of early defoliation was more pronounced in Prokupac, in terms of producing looser clusters with smaller berries. This structure is favorable for light exposure of berries and an increased content of total anthocyanins. Kliewer and Smart (1989) stated that in raising the anthocyanin content under manual defoliation may have been achieved by improved cluster microclimate. Increase in the dry matter content, total anthocyanins and phenolics in berry skins due to early defoliation may be a consequence of several mechanism. Many investigations confirmed that the light exposure of clusters increases dry matter content, total anthocyanins and phenols, decrease in the total acids and pH and the malate content (Kliewer, 1970; Smart et al., 1985; Morrison and Noble, 1990; Dokoozlian and Kliewer, 1996; Chorti et al., 2010). Other studies similarly show that the effects of light on fruit composition are dependent on the degree of berry exposure and the resulting temperature (Haselgrove et al., 2000; Bergqvist et al., 2001). The authors concluded that increased but moderate exposure to sunlight improved grape and wine composition. The increased temperature accelerates the rate of metabolic processes in the plant, with subsequent acceleration in the development and metabolite accumulation (Downey et al., 2006). However, many metabolic processes in plants ceased or were markedly reduced at high temperatures. The excessive cluster light exposure, expecially in warm climates, can reduce skin anthocyanins and berry coloration (Downey et al., 2004). The high temperature limit for metabolic processes in grapevines is thought to be around 30^0C (Coombe, 1987).

The second factor involves source : sink ratio disorder between the leaf surface and clusters. The early partial removal of leaves from shoots directs assimilator transportation to clusteres (Koblet et al., 1993). Another study (Candolfi-Vasconcelos et al., 1994), reported that much more ^{14}C reserves were translocated to the fruit of defoliated vines compared to untreated control vines.

Conclusion

This examination confirmed a significant effect of leaf defoliation on the cluster structure and berry composition. Defoliation carried out during the period of pericap cell division and growth caused an irreversible decrease in berry size when compared with defoliation at later treatment (veraison) and the control in both cultivars. Early defoliation also decreased fruitset, resulting in looser grape clusters and less average weight. Therefore, the average yield within the early defoliation treatment was also lowered. All these effects are more pronounced in cv. Prokupac. Defoliation causes higher soluble solids content in must in both cultivars. Early defoliation (full bloom), increased the content of total phenols in comparison with control, while the total content of anthocyanins was not significant changed in Cabernet Sauvignon. In cv. Prokupac, erly defoliation increased content of total phenols (in the full bloom and fruit set treatments) and total anthocyanins (in full bloom treatment). The possibility that early defoliation can significantly affect the structure of the cluster, yields and chemical composition of grapes, especially the content of anthocyanins, is very important for canopy management of cv. Prokupac. From a practical perspective, early removal of basal leaves can replace the costly and time-consuming cluster thinning as tool of yield control, especially if it can be mechanized.

Acknowledgements

This paper was realized as a part of the project 31063 financed by the Ministry of Education and Science of the Republic of Serbia for the period 2011-2015.

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Received April, 2, 2012; accepted for printing December, 2, 2012.