

VARIATION OF STRUCTURAL BUNCH AND BERRIES INDICATORS AND PHYSICO-CHEMICAL WINE PROPERTIES OF CABERNET SAUVIGNON CV. UNDER INFLUENCE OF DEFOLIATION AND HARVEST TIME IN AGROECOLOGICAL CONDITIONS OF CENTRAL SERBIA

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ABSTRACT

Defoliation and different harvest time can affect structural composition of bunch and berries. Research was carried out at Cabernet Sauvignon variety in vineyard of King Peter I Karadjordjevic-Royal Winery, Serbia. Defoliation included treatments with 4 and 8 removed leaves and control. The harvest was done at full maturity and 15 days after. Structural cluster indicators in time of full maturity had higher value in treatment with 8 removed leaves. At late harvest the highest values was founded in treatment with 4 removed leaves (epidermis 15.2%, mesocarp 73.8%, seeds 10.9%). By physico-chemical analysis of wines was founded the highest alcohol content in treatment with 8 removed leaves (12.13% v/v). In control was recorded the highest levels of total extract and extract without sugar. Total acid content was higher in treatment with 8 removed leaves. The highest level of total polyphenol level (1.584 g/l) was noted in treatment with 4 removed leaves. In treatment with 8 removed leaves was recorded the highest level of total anthocyanins (0.166 g/l). In term of late harvest most of examined parameters had higher values. In the second experimental year parameters of physico-chemical wine analysis had significantly higher values compared to both harvest terms.

INTRODUCTION

Defoliation is ampelotechnical technique which is used to improve grape quality. In conditions of Serbia is mostly applying 20-30 days before grape harvesting (*veraison*). It is conducted by leaves removing from clusters zone which overshadow clusters. Defoliation as ampelotechnical technique has multiple advantages: improves brightness at clusters area, improves aeration which improves microclimate of vines, maturation process of clusters is better, better is berry skin coloration, and are present less degree of gray mold and damage which it creates by this deases [1].

Effect of defoliation dependent of intensity- number of removed lives and application time or growth stages when is performed. Based on application time (growth stages when is performed) can be carried out before flowering or at fruit set (early defoliation) or at *veraison*. Early defoliation, ie. removing bigger part of leaf mass, causing photosynthetically shock which stoped influx of necessary quantity of newly formed asimilatives in inflorescence. As a result is forming less number of berry, grapes are struggly worse, berries are smaller with reduction of risk of occurrence of gray mold, also epidermis participate in bigger ration to mesocarp [2-3].

Some authors [4] report that defoliation during berry growth and veraison is not shown a significant effect on total growth of leaf mass and an increasing of photosynthetic activity, which is not affect yield (berries mass, cluster mass and yield per vine).

On the other hand, some authors [5] indicates that removal of several basal leaves from shoot in flowering time, can change total assimilation surface and photosynthetic activity of remaining leaves on shoot which is changed parameters of yield and qualitative grape parameters (content of accumulated sugars and acids), berry coloring and aromatic complex.

When defoliation carrying out to achieve positive effects it is necessary to pay attention to phenological stage and number removed leaves. When is removing 15-25% leaves at 20-30 days before grape harvesting positive effects are highly noticeable and are then is removing the oldest leaves that have reduced photosynthetic activity, while the younger leaves remain on main shoot, and leaves of laterals which are the most photosynthetic active [6].

Mechanical composition of grape and berry represents ampelographic and technological characteristic of of each variety. Structural indicators of bunches and berries, represented through participation of individual elements such as percent of epidermis and mesocarp in berries, hard residue which participate in cluster or berries. This elements significantly affect technological properties of cluster and eventually physical and chemical properties of wine. Defoliation can be significantly change ratio of epidermis and mesocarp, which influences concentration of anthocyanins and other flavonoid compounds essential for wine quality [7].

MATERIALS AND METHODS

Research was carried out in vineyard of King Peter I Karadjordjevic-Royal Winery at Oplenac-Topola municipality at Cabernet Sauvignon cv. Vineyard is an altitude of 250 m and covers area of 3.7 ha. Row spacing is 2.7 m and 1.0 m spacing between vines in row. It is geographically positioned at GPS coordinates N 44° 14 '4" and E 20° 41' 15". Training system is characterized with height of 90 cm on which Guyot pruning is used. All experimental vines were uniformly pruned where one arc was left with eight buds and spur with two buds.

Defoliation was carried out at *veraison* and included treatments with 4 and 8 removed leaves and control vines on which was not applied defoliation. The harvest was done at full maturity and 15 days after. For the purposes of ampelografic analysis was harvested average sample of ten bunches at full maturity and 15 days after.

After clusters harvesting and separation an average sample of 100 berries are separated epidermis and seeds and were determined values for: participation of skins, seeds and mesocarp in berries, % of hard residue structure of cluster and cluster indicator.

For all treatments was prepared wine which was analyzed. Analysis was conducted in laboratory of Faculty of Agriculture, University of Belgrade Department of Viticulture.

By physico-chemical analysis were determined values for the following parameters:

- a) specific wine gravity-D_{120/20} by densitometry method (DMA)
- b) alcohol content (% v/v) NIR spectrometric method (Alcolyzer)
- v) total extract (g/l) by densitometry method (DMA-Alcolyzer)
- g) extract without sugar (g/l)
- d) content of reducing sugars (g/l) Bertrand method

- f) total acids as tartaric (g/l) volumetry method
- e) content of malic acid
- f) content of citric acid
- i) contents of lactic acid-Reflectoquant-RQflex® 10
- i) volatile acids such as acetic (g/l) volumetry method
- j) total SO₂ (mg/l) volumetry method
- k) free SO₂ (mg/l) volumetry method
- l) total polyphenols (g/l) by spectrophotometry-Folin Ciocalteu method
- m) total anthocyanins (g/l) by spectrophotometry-Riberao Gayon method
- n) intensity wine color by spectrophotometer
- nj) color wine tone by spectrophotometry
- o) Fe content (mg/kg) AA spectrophotometry
- p) content of Cu (mg/kg) AA spectrophotometry

For two year data analysis software IBM SPSS Statistics 2.0, Chicago, IL, USA was used.

RESEARCH RESULTS

In paper were present values for: participation of epidermis, seeds and mesocarp in berry, structure of hard residue of cluster and cluster indicator. During two-year experiment higher participation of epidermis in berry is observed in later harvest while at full maturity recorded a smaller value. In the first year of experiment at later harvest the largest percent of berry epidermis was distinguished in treatment with four-removed leaves (15.18%), while in other two treatment varied from 13.76-13.87%. In full maturity were noticeable smaller variation among treatments with slightly larger participation in control (9.40%).

In the second year is also recorded higher participation of berry epidermis in a later harvest with less variation by treatments. In full maturity was wider variation whereby the highest percent emphasizes treatment with four removed leaves (9.98%) and with lower value control (8.47%) and treatment with eight removed leaves (9.22%).

Using of trofactorial ANOVA in the first year is determined influence of main effects: cultivar, harvest time and interaction-variety*harvest time, while in the second year, is determined statistically significant effect of main effects: variety and harvest time (table 1).

A higher percent of seeds per berry was noted in treatment with four removed leaves (10.98%), while lower percent was recorded in control (10.33%) and treatment with four removed leaves (9.99%). The largest percent at full maturity were recorded for treatment with eight removed leaves (7.41%), while other two treatment has been with decreased values (7.09 and 6.53%).

In the second year, was observed same variation trend. Higher values were recorded at full maturity in treatment with four and eight removed leaves (6.08 and 6.14%). In period of later harvest was recorded an increase in participation of berry seeds with a larger number of removed leaves.

Using of trofactorial ANOVA in the first year is determined influence of main effects: cultivar and harvest time. Also, there was influence of interaction-variety*harvest time. In the second year was is determined statistically significant effect of main factors: variety, harvest time and treatment and is interaction-variety*harvest time and variety*treatment (table 1).

Cabernet sauvignon in both years had higher participation of mesocarp in berry at full maturity, while later harvest mesocarp participation decreased. In the first year at

full maturity in treatment with four removed leaves indicated a slight increase of mesocarp participation compared to other two treatments. Same treatment in later harvest recorded decrease compared to other two treatments. In the second year, higher values were observed in control (87.14%) and treatment with eight removed leaves (86,05%) and at a later harvest variation by treatments was minimal.

By statistical data processing for the first year of research were founded significant impact of variety, harvest time and interaction-variety*harvest time. In the second year there was impact of variety, treatment and interaction-variety*harvest time.

Some authors report [8] that in cluster grape stem participates with 3-4.5%, epidermis with 8-11%, seeds with 2-4,5% and mesocarp with 75-80%. While some report [9] that participation of epidermis, seeds and mesocarp in berry is in correlation with berries size. With berries growing it is growing percentage of epidermis, seeds and mesocarp in berry. In berry in average mesocarp contributing with 80%, epidermis with 15% and seeds with 5%. Using irrigation, fertilization (particularly potassium fertilizer) or defoliation come up disturbances of epidermis:mesocarp ratio and percent of seed in berry having a substantially constant value.

Table 1.

Structural composition of bunch and berries-statistical analyzes

Year	Source of variation	% of epidermis in berries	% of seed in berries	% of mesocarp in berries	Hard residue	Structure cluster indicator
		<i>p values</i>				
I	Variety	0,000	0,000	0,000	0,000	0,000
	Harvesting time	0,000	0,000	0,000	0,000	0,000
	Treatment	0,582	0,812	0,703	0,033	0,021
	Variety *	0,000	0,000	0,000	0,027	0,374
	Harvesting time					
	Variety * Treatment	0,345	0,901	0,779	0,571	0,366
	Harvesting time *Tretman	0,189	0,252	0,242	0,043	0,015
	Variety * Harvesting time * Treatment	0,641	0,317	0,405	0,010	0,010
II	Variety	0,000	0,001	0,000	0,000	0,000
	Harvesting time	0,000	0,000	0,041	0,000	0,714
	Treatment	0,228	0,000	0,024	0,013	0,001
	Variety *	0,345	0,000	0,004	0,879	0,353
	Harvesting time					
	Variety * Treatment	0,796	0,056	0,435	0,320	0,802
	Harvesting time *Tretman	0,818	0,066	0,479	0,805	0,917
	Variety * Harvesting time * Treatment	0,321	0,307	0,843	0,334	0,511

Significance $p=0,05$

In the first research year was higher participation of hard residue in later harvest in treatment with four removed leaf. In control and treatment with eight removed leaves was recorded less percent of hard residue. At full maturity in all treatment hard residue had increasing values with larger number of removed leaves.

In the second year of at full maturity is noted same trend of variation as in later harvest in the previous year. Statistical analysis showed that participation of hard residue was influenced by variety, treatment and harvest time. Effects of other factors has not been founded.

Cabernet Sauvignon in both years had more value for structure indicators of cluster which is recorded at full maturity. During the first year, in both harvest terms (full maturity and later harvesting dates) was recorded reducing of value of cluster structure indicator. With increase of number removed leaves value for structure indicator cluster was decline.

During the second year, a significant decreasing was observed in treatment with four removed leaf while control and treatment with eight removed leaves recorded higher values. Same variation trend for structure cluster indicator was noted in both terms of harvest.

By statistical data processing in ANOVA in the first year was determined influence of variety and harvest time while in the second year was founded effect of variety and treatment. Results of statistical data processing are shown in table 1.

Examination of differences between clones ISV FV 5 and ISV FV 6 in terms of uvometric characteristics variation of bunches and berries, came to following results [10]. Higher sugar and acid content in grape juice was found for clone ISV FV 5 (20.14% and 8.08 g/l) relative to clone ISV FV 6 (18.81% and 7.56 g/l). Research of uvometric characteristics of berries and bunches were founded higher cluster stem weight (4.6 g) and berries number (145) for clone ISV FV 6, whereas clone ISV FV 5 had higher berries mass (154 g). Higher percent of cluster stem (4.09%), percent of epidermis in cluster (8.32%) and percent of hard residue (17.55%) was found for clone ISV FV 6. Clone ISV FV 5 had a greater percentage of berries in cluster (96.87%), percent of seeds in cluster (5.38%) and percentage of mesocarp in cluster (83.26%).

On defoliation most varieties respond differently. Muscat Hamburg reacted very positively to partial defoliation from beginning of clusters developing because in the shade created by older leaves form straggly clusters. On the other hand the Cardinal variety form berries whose coloration is the best expressed if they are formed in the shade, not in direct sunlight. In areas where is a level of insolation increase is not necessary to complete defoliation due to fear of cluster necrosis or changes in relation epidermis:mesocarp [11].

Application of partial defoliation in two terms: after flowering and at veraison, results showed that increased content of anthocyanins, phenolic compounds and sugar have a greater impact defoliation during veraison. It was also found that content of coloring matters and sugar are directly connected, or that berries with a higher sugar content were more colored and had a higher proportion of epidermis [12]. A similar beneficial effect as well as influence on load asimilativa, shoots ripening and prepare for winter period was established by other authors [13] at Rkaciteli [14], Cabernet Sauvignon variety.

For Pinot noir [15] it was founded that change of shade conditions by reduction of leaves did not lead significant changes in mechanical composition of cluster, berries mass or other cluster uvometric characteristics, but there has been a change in accumulation of flavonoid compounds. Concentration of flavonoids in shaded bunches

during veraison was 5.5 times lower compared to clusters that were exposed directly to sun, and at full maturity, content of flavonoid compounds are shaded clusters was 8 times lower compared to non-shaded clusters. Content of flavan-3-ol in seeds and berry skin expressed as catechin and epicatechin, was same in veraison and at full maturity, wherein at sunny clusters concentration of epicatechin was higher compared to clusters formed directly at sun. Total content of flavan-3-ol in shaded grapes varied and amounted 0.74 mg/g, while in berry skin flavan-3-ols were present with 1.20 mg/g. Content of anthocyanins in berry skin was at sunny bunches 32% lower compared to non-shaded clusters. Shaded clusters contained low concentrations of delphinidin-3-glucoside, cyanidin-3-glucoside and petunidin-3-glucoside but it is recorded high concentration of peonidin-3-glucoside. Content of flavan-3-ol, according to some authors [16] is much lower in the skin compared to seed where content of flavan-3-ol ranges from 1-24 mg/g.

By physico-chemical analysis of wine were founded that in the first year of research the highest alcohol content was recorded in treatment with 8 removed leaves (12.13% v/v), while a slightly lower content were recorded in control (12.06% v/v) and the lowest were in treatment with 4 removed leaves (11.91% v/v). In control were observed maximum value of total extract and extract with sugar, while treatment with 8 and at list 4 removed leaves. Content of reducing sugars was both treatments of defoliation with same values (1.56 g/l). Values related to total acid content, expressed as tartaric, malic, citric and lactic acid, were the highest in control treatment, then in treatment with 8 removed leave and at the end treatment with 4 removed leaves, as shown in table 2. The highest level of volatile acid expressed by acetic acid was the highest in treatment with 4 removed leaves (0.66 g/l). The same trend was recorded for value of total and free SO₂. The highest level of total polyphenols was determined for the wine from treatment with 4 removed leaves (1.584 g/l). In treatment with 8 removed leaves was recorded the highest level of anthocyanins in the wine (0.166 g/l), then in control (0.126 g/l) and in treatment with 4 removed leaves (0.109 g/l). As most intensively colored rated wine from treatment with 8 removed leaves (1.301), followed by treatment with 4 removed leaves (1.170).

In term of later harvest most of examined parameters of physical and chemical analysis had higher value compared to full maturity. Alcohol content has increased compared to full maturity with the highest content recorded in control (13.33% v/v). Level of extract without sugar and total extract was recorded at a higher level in both defoliation treatment compared to control. Level of acids (total, malic, citric and lactic) were almost same in all treatments with less variation. The highest value of volatile acid were recorded in treatment with 4 removed leaves. Level of total anthocyanins decreased in period of later harvest. Wine from same treatment had the best color while wine color from the treatment with the 4 and 8 have removed leaves.

In the second year parameters of physico-chemical wine analysis had significantly higher values compared to both terms from previous harvest year. In period of full maturity were recorded lower alcohol content in relation to wine from later harvest. During full maturity with the highest alcohol content was characterized wine from treatment with four removed leaf (15.66% v/v), while the lowest content was recorded in control (14.89% v/v). Content of total extract and sugar-free extract had the highest level in treatment with 4 removed leaves, then in treatment with 8 removed leaves and ultimately in control. The highest level of malic acid, citric acid, lactic acid and volatile acid expressed as acetic acid, was recorded in wine from treatment with 8 removed leaves, then in control and at the end in wine from treatment with 4 removed leaves.

Total and free SO₂ were determined in the highest concentration in treatment with 8 removed leaves, then in treatment with 4 removed leaves at end in control.

Same variation trend was recorded for total anthocyanins content. The largest intensity and the most characteristic color was determined for wine from treatment with 4 removed leaves.

Table 2.
Physico-chemical analysis of Cabernet Sauvignon wine for first research

Harvesting time	Full maturity			Late harvest		
Treatment	Control	4 removed leaves	8 removed leaves	Control	4 removed leaves	8 removed leaves
Date of harvest	17.10.	17.10.	17.10.	1.11.	1.11.	1.11.
Specific wine gravity	0.99253	0.99315	0.9928	0.99633	0.99397	0.99407
Alcohol content (% v/v)	14.89	15.66	15.06	15.41	15.68	16.72
Total extract (g/l)	30.18	33.99	31.37	41.52	36.16	39.33
Sugar-free extract (g/l)	28.98	32.03	29.41	31.16	34.68	35.53
Content of reducing sugars (g/l)	2.2	2.96	2.96	11.36	2.48	4.8
Total acid as tartaric acid (g/l)	7.2	7.1	7.4	6.5	6.2	6.5
Content of malic acid (g/l)	6.43	6.34	6.60	5.80	5.54	5.80
Content of citric acid (g/l)	6.14	6.06	6.31	5.54	5.29	5.54
Content of lactic acid (g/l)	8.64	8.52	8.88	7.8	7.44	7.8
Volatile acids as acetic acid (g/l)	0.45	0.66	0.69	0.57	0.54	0.63
Total SO ₂ content (mg/l)	42.24	56.32	67.84	56.32	34.56	30.72
Free SO ₂ content (mg/l)	12.8	12.8	17.92	16.64	16.64	19.2
Total polyphenols (g/l)	1.914	2.442	2.7	2.136	2.28	2.91
Fe (mg/kg)	1.7	1.9	1.7	2.5	1.4	1.8
Cu (mg/kg)	0.1	0.1	0.1	0	0	0.1
Anthocyanins (g/l)	0.319	0.326	0.343	0.434	0.284	0.326
Color intensity	1.256	1.4	1.354	1.437	1.368	1.49
Color nuance	0.694	0.875	0.812	0.926	0.833	0.993

In later harvest, there was a trend of increasing of value of alcohol content, with the highest contents of removed leaves. In treatment with 8 sheets was 16.72% v/v. Unlike to full maturity in a later harvest the highest content of total extract was recorded in control, while sugar-free extract maximum value was in treatment with 8 removed leaves.

Acid content (total, malic, lactic, citric and volatile) recorded same variation trend wherein the higher value was recorded in treatment with 8 removed sheets, while a significant increase was recorded in control.

Total SO₂ content was the highest in control compared to the highest level of free SO₂ which was detected in the highest content in treatment with 8 removed leaves. In period of later harvesting was recorded a reduction of total anthocyanins in wine where defoliation was performed, while the higher values was determined in control.

Wine from treatment with 8 removed leaves had a better color they are followed by wine from the control, and finally the lowest values for this parameters are determined for wine from treatment with 4 removed leaves. The results are shown in table 3.

With different harvesting period is affects content of aromatic components such as content of higher alcohols, lactones, organic acids, and esters of [17,18,21].

Except yield control, uvometric composition of berries and bunches through lower percentage berries, defoliation can affect sugar accumulation in the berries, a higher concentration of these compounds, stability of the anthocyanin content in the wine or other physical-chemical parameter of wine [14,19].

In addition to these treatments on uvological or mechanical bunches and berries properties and physico-chemical parameters of wine can be influenced by using different doses of a potassium fertilizer.

Some authors [10,22,23] with treatment of 50, 100 and 150 kg K₂O/ha to determine impact on uvometric and technological properties of Sauvignon blanc.

The greatest variations were noted in control (for cluster mass and %c of luster stems, % of berries) and in variants with 150 kg K₂O/ha (% of berry in grapes, berries structure and cluster structural indicators).

Sugar content was significantly lower in variant with 150 kg K₂O/ha relative to other treatments, while the acid content had opposite trend.

The highest alcohol content in wine (12.7% v/v), pH (3.97), free SO₂ (8.96 mg/l) and potassium (798.0 mg/l) were observed in treatment with 100 kg K₂O/ha, while the highest value of ash content (6.5 g), specific gravity (0.9934) and alkalinity (16.5) recorded in the variant with 50 kg K₂O/ha.

CONCLUSIONS

After research can be carried out following conclusions:

For most of examined parameters of cluster and the berries mechanical composition the greater variations were observed between two harvesting terms wherein during later harvest was recorded less variation. Through experiment treatments (control, 4 and 8 removed basal leaves) had decreasing values for most of parameters which was recorded with increased number of removed leaves. Structural indicators were higher in the period of later harvest in control and treatment with 8 removed leaves.

By physico-chemical analysis of wine most parameters had more values of the treatment with 8 removed leaves, followed by treatment of 4 removed leaves. In period of later harvest wines during both years were recorded higher values of physico-chemical parameters.

Table 3.

Physico-chemical analysis of Cabernet Sauvignon wine for second research

Harvesting time	Full maturity			Late harvest		
Treatment	Control	4 removed leaves	8 removed leaves	Control	4 removed leaves	8 removed leaves
Date of harvest	17.10.	17.10.	17.10.	1.11.	1.11.	1.11.
Specific wine gravity	0.99253	0.99315	0.9928	0.99633	0.99397	0.99407
Alcohol content (% v/v)	14.89	15.66	15.06	15.41	15.68	16.72
Total extract (g/l)	30.18	33.99	31.37	41.52	36.16	39.33
Sugar-free extract (g/l)	28.98	32.03	29.41	31.16	34.68	35.53
Content of reducing sugars (g/l)	2.2	2.96	2.96	11.36	2.48	4.8
Total acid as tartaric acid (g/l)	7.2	7.1	7.4	6.5	6.2	6.5
Content of malic acid (g/l)	6.43	6.34	6.60	5.80	5.54	5.80
Content of citric acid (g/l)	6.14	6.06	6.31	5.54	5.29	5.54
Content of lactic acid (g/l)	8.64	8.52	8.88	7.8	7.44	7.8
Volatile acids as acetic acid (g/l)	0.45	0.66	0.69	0.57	0.54	0.63
Total SO ₂ content (mg/l)	42.24	56.32	67.84	56.32	34.56	30.72
Free SO ₂ content (mg/l)	12.8	12.8	17.92	16.64	16.64	19.2
Total polyphenols (g/l)	1.914	2.442	2.7	2.136	2.28	2.91
Fe (mg/kg)	1.7	1.9	1.7	2.5	1.4	1.8
Cu (mg/kg)	0.1	0.1	0.1	0	0	0.1
Anthocyanins (g/l)	0.319	0.326	0.343	0.434	0.284	0.326
Color intensity	1.256	1.4	1.354	1.437	1.368	1.49
Color nuance	0.694	0.875	0.812	0.926	0.833	0.993

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