

INFLUENCE OF LEAF REMOVAL ON BERRY THERMAL EFFICIENCY FOR ANTHOCYANIN ACCUMULATION

INFLUENCE DE L'EFFEUILLAGE SUR L'EFFICIENCE THERMIQUE DE L'ACCUMULATION DES ANTHOCYANES DANS LA BAIE

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Abstract

The aim of this work was to analyse the effects of leaf removal on Touriga Nacional berry temperature and consequent thermal efficiency for anthocyanins biosynthesis. The field experiment was located at Dão Wine Research Station, Nelas, Portugal in an adult vineyard planted with North-South oriented rows, with the red grape variety Touriga Nacional grafted on 110R rootstock. The vines were trained on a vertical shoot positioning, spur-pruned on a bilateral Royat cordon system and deficit irrigated (50% ET_c). The experimental design was a randomized complete block design with four replications of twelve vines per elemental plot, and the following two treatments: basal leaf removal (LR) and a control non-defoliated (ND). Berry temperature (T_b) was measured continuously during the second half (3rd to 19th September) of the 2009 ripening period using two-junction, fine-wires copper-constantan thermocouples manually inserted into the berries and connected to a data logger. A sample of clusters located in different canopy positions (exposed and internal; facing East and West) of 4 vines per treatment were used. To quantify the effect of T_b on anthocyanins biosynthesis, the berry hourly mean temperatures were converted into normal heat hours (NHH) and accumulated per day (NHH_d) and per monitoring period (NHH_c). For quantification of thermal requirements for anthocyanins synthesis and accumulation, a minimum of 10°C, a maximum of 35°C, and an optimum of 26°C were used. Meteorological variables were measured at an automatic weather station installed within the experimental plot. For all days of the monitoring period, daily average berry temperature (dT_b) of all monitored berries was lower in ND treatment than in LR, being the maximum differences between treatments registered on 11th September. The highest dT_b differences between treatments were registered on the clusters located at the west side of the canopy on 7th September while dT_b of the clusters located in the centre of the canopy was less affected by leaf removal. The control non-defoliated treatment (ND) presented a significantly higher NHH_c than that of LR being the higher differences presented by the clusters located in the west side. The lowest differences in NHH_c were obtained in the clusters located in the centre of the canopy. Our results show that the thermal efficiency for berry anthocyanins accumulation was significantly affected by leaf removal and that this effect was dependent of the meteorological conditions, time of the day and berry/cluster location into the vine canopy.

Keywords : Touriga Nacional, leaf removal, berry temperature, Normal Heat Hours, anthocyanins biosynthesis

Résumé

L'objectif de cette étude était d'analyser les effets de l'effeuillage sur la température des baies du cépage Touriga Nacional et l'efficacité thermique conséquente pour la biosynthèse des anthocyanes. La parcelle expérimentale est localisée au Centre d'Études Vitivinicoles de la Région du Dão, Nelas, Portugal dans un vignoble adulte planté avec des rangs orientés Nord-Sud, avec la variété 'Touriga Nacional' greffée sur 110R. Les vignes sont conduites en cordon bilatéral de Royat et soumis à un déficit d'irrigation (50%, ET_c). Le dispositif expérimental était organisé en blocs aléatoires complets avec quatre répétitions de douze plants et les deux traitements suivants: effeuillage basale (LR) et un témoin non défolié (ND). La température des baies (T_b) a été mesurée en continu au cours de la seconde moitié de la maturation (3 jusqu'au 19 Septembre) de l'année 2009 avec des thermocouples du type « copper-constantan » insérés manuellement dans les baies et reliés à un enregistreur de données. Les mesures ont été réalisées dans un échantillon de grappes situées dans différentes positions de la canopée (exposée et intérieure; face à l'Est et à l'Ouest) de 4 pieds par traitement. Pour quantifier l'effet de la T_b sur la biosynthèse des anthocyanes, les températures moyennes horaires de la baie ont été converties en heures de chaleur normalisées (NHH) et cumulées par jour (NHH_d) et par période d'enregistrement (NHH_c). Pour la quantification des exigences thermiques pour la synthèse et l'accumulation anthocyanes, on a considéré un minimum de 10 °C, un maximum 35 °C et 26 °C comme la température optimale. Les variables météorologiques ont été mesurées par une station automatique installée dans la parcelle expérimentale. Pour tous les jours de la période d'acquisition, la température moyenne journalière de toutes les baies (dT_b) était inférieure dans le traitement ND que dans de traitement LR. Les différences maximales entre les traitements ont été enregistrés le 11 Septembre. Les principales différences de dT_b entre les traitements ont été enregistrées sur les grappes situées sur le côté Ouest de la canopée le 7 Septembre tandis que la dT_b de grappes situées dans le centre de la canopée ont été moins affectées par l'effeuillage. Le traitement témoin (ND) a présenté un NHH_c significativement plus élevé que celui du traitement LR. Les différences les plus élevés sont présentées par les grappes situés sur le côté Ouest. Les différences du NHH_c les plus bas ont été obtenues dans les grappes situées au centre de la canopée. Ces résultats montrent que l'efficacité thermique pour l'accumulation des anthocyanes de la baie a été significativement affectée par l'effeuillage et que cet effet était dépendant des conditions météorologiques, de la période de la journée et de la localisation des baies/des grappes sur la canopée de la vigne.

Mots-clés : « Touriga Nacional », effeuillage, température de la baie, heures de chaleur normalisées, biosynthèse des anthocyanes

1. Introduction

Grape berry composition at harvest is the result of a complex balance from a large number of factors ranging from the environment to the variety. Environmental stresses, namely drought and its interplay with light and temperature, have been shown to play a major role on berry composition. Grape-derived secondary metabolites are the sources of wine characteristics including aroma, flavour, colour and taste, being the polyphenolic composition very important for red wine quality Adams (2006). The concentrations of those compounds are influenced by the site, cultivar, climate and management practices such as irrigation (Santos et al., 2003) or defoliation (Vasconcelos and Castagnoli, 2000). However, relationships between cluster sunlight exposure and secondary metabolites are still not fully understood. Cluster microclimate can influence the biochemical processes that determine berry ripening such as the activity of photoinducible enzymes responsible for the biosynthesis of polyphenols and ultimately define berry ripening and wine quality. Previous studies involving shading (Downey et al. 2004) or defoliation of the vines (Guidoni et al., 2008; Vasconcelos and Castagnoli, 2000) showed that, in general, well exposed clusters, when compared with shaded ones, presented higher total soluble solids, anthocyanins and phenolics, lower titratable acidity and lower juice pH and berry weight.

However little consideration for the effects of excessive cluster exposure to solar radiation was paid. Early studies on the effects of temperature suggest that a 15°C day temperature during ripening improved berry colour while the 35°C reduced the formation of anthocyanins (Kliewer and Torres, 1972). Yamanne et al. (2006) found that skin anthocyanins accumulation was significantly higher at 20°C than at 30°C. High temperatures (> 35° C) seem to inhibit anthocyanins synthesis but also to induce their degradation (Bergqvist et al. 2001, Spayd et al.; 2002). It is difficult to separate the effects of light from those of temperature on berry metabolism as many of the biochemical pathways are both light and temperature sensitive. It was shown that exposure to sunlight increased total skin anthocyanins regardless of temperature (Ristic et al., 2007, Spayd et al., 2002), but a decrease in skin anthocyanins was also observed in berries from west-exposed clusters (Spayd et al.; 2002). Basal leaf removal is one of the most frequently applied canopy management practices on dense canopies to promote incident light and air circulation in the cluster-zone. The effects of defoliation on berry colour and aroma are not always consistent, with some experiments showing positive effects (Guidoni et al, 2008, Poni et al 2006) and others finding no differences (Lee et al.; 2007, Vasconcelos and Castagnoli, 2000). Before deciding the level of leaf removal it is important to have a comprehensive understanding of the interaction of light with other environmental factors, namely temperature and water availability. The aim of this work was to analyse the effects of leaf removal on the red variety Touriga Nacional berry temperature and consequent thermal efficiency for anthocyanin biosynthesis.

2. Material and Methods

This work took place at the Dão Grape and Wine Research Station (CEVDão), Nelas, Portugal (Latitude 40° 31'N, Longitude 7° 51'W, Elevation 440 m) in a vineyard planted in 2000 with the red grape variety 'Touriga Nacional' grafted on 110 R rootstock. The vines were spaced 1.1 m within and 2.0 m between rows, trained on a vertical shoot positioning with a pair of movable wires, spur-pruned on a bilateral Royat Cordon system and deficit irrigated (50% ETc) according to Rodrigues et al. (2012). The experimental design was a randomized complete block design with four replications of 12 experimental vines per elemental plot, and the following two treatments: basal leaf removal (LR) and a control non-defoliated (ND). The soil is derived from granite, with a coarse texture, acid pH and a very good infiltration capacity. Meteorological variables were measured with an automatic weather station installed within the experimental plot. Berry temperature (T_b) was measured continuously, during the second half of 2009 ripening period (3th to 19th September), using two-junction, fine-wires copper-constantan thermocouples manually inserted into the berries and connected to a data logger. A sample of clusters located in different canopy positions (exposed and internal; facing East and West) of 4 vines per treatment were used. The effect of T_b on anthocyanins biosynthesis has been quantified following the approach of Rustioni et al. (2006). The berry hourly mean temperatures were converted into normal heat hours and cumulated per day (NHH_d) and per monitoring period (NHH_p). Rustioni et al. (2006) describe the procedure for computing NHH as based on a function that varies from 0 to 1 giving 0 for temperatures outside minimum and maximum ($T_{min} = 10\text{ }^{\circ}\text{C}$ and $T_{max} = 35\text{ }^{\circ}\text{C}$, respectively) and 1 for optimum temperatures ($T_{opt} = 26\text{ }^{\circ}\text{C}$). At harvest a sample of 100 berries per elemental plot was collected and the juice was analyzed for soluble solids, titratable acidity, pH and total phenols by Fourier Transform Infrared Spectroscopy (FTIR). Berry skin anthocyanins were determined by the method of discoloration by sodium bisulfite and total phenols by Folin-Ciocalteu index (Ribéreau-Gayon et al. 1972). Data were subjected to a T-student analysis of variance using the Statistical Package for the Social Sciences (SPSS for Windows, Standard Version Release 19.01).

3. Results and Discussion

Daily average berry temperature

During all the monitoring period, daily average berry temperature (dT_b) of all monitored berries was lower in ND treatment than in LR one, being the maximum differences between treatments registered on 11th September. The defoliated treatment showed a higher dT_b than the control being the highest differences registered on the clusters located at the west side of the canopy and the lowest ones on the clusters located in the centre of the canopy. This positive effect of sun exposition on berry temperature was also observed by Keller and Hrazdina (1998), Bergqvist et al. (2001), Spayd et al. (2002) and Guidoni et al. (2008) and was mainly due to the diurnal temperature differences as during the night the effect was opposite (but less intense) (Table 1).

Table 1. Effect of basal leaf removal and cluster position on daily average berry temperature measured during the second half of 2009 ripening period (September 3th to 19th). LR - basal leaf removal; ND- control non-defoliated.

Tableau 1. Effet de l'effeuillage et de la position des grappes sur la température moyenne journalière des baies au cours de la seconde moitié de la maturation (du 3 au 19 Septembre) de l'année 2009. Effeillage basal (LR) et témoin non défolié (ND).

Clusters position	Daily average temperature (°C)				Night (21h a 7h) average temperature (°C)				Diurnal (8h a 20h) average temperature (°C)			
	LR	ND	difference/sig		LR	ND	difference/sig		LR	ND	difference/sig	
East	23.70	23.30	0.40	<i>ns</i>	15.40	15.50	-0.10	<i>ns</i>	24.30	23.90	0.40	<i>ns</i>
Centre	23.40	23.20	0.20	<i>ns</i>	15.40	15.70	-0.30	<i>ns</i>	24.10	23.90	0.20	<i>ns</i>
West	23.90	23.40	0.50	<i>ns</i>	15.40	15.60	-0.20	<i>ns</i>	24.70	24.10	0.60	<i>ns</i>
All clusters	23.70	23.30	0.40	<i>ns</i>	15.40	15.60	-0.20	<i>ns</i>	24.40	23.90	0.50	<i>ns</i>

Normal heat hours

As compared to the control, the LR treatment presented a significantly lower accumulated normal heat hours during the monitoring period (NHH_c), on all the cluster locations, being the highest differences presented by the berries of the clusters located in the west side and the lowest ones in the berries of clusters located in the centre of the canopy (table 2). Similar results were also observed by Rustioni et al. (2011).

Table 2. Effect of defoliation and cluster position on accumulated normal heat hours during the monitoring period (September 3th to 19th). LR - basal leaf removal; ND- control non-defoliated.

Tableau 2. Effet de l'effeuillage et de la position des grappes sur l'accumulation d'heures de chaleur normalisées au cours de la seconde moitié de la maturation (3^{ème} jusqu'aux 19^{ème} Septembre) de l'année 2009. Effeillage basale (LR) témoin non défolié (ND).

Clusters position	NHHc (hours)			
	LR	ND	difference/sig	
East	193.61	203.97	-10.36	**
Centre	193.25	201.65	-8.40	**
West	185.28	196.36	-11.08	*
All clusters	190.72	200.66	-9.94	***

However, the effect of leaf removal vary on a daily basis depending on climate condition. On September 11th, a typical summer day at Dão region, the LR treatment presented a significantly lower NHH_d on all clusters location being the highest differences observed on the east side clusters and the lowest ones on the clusters located in the canopy centre. On September 19th, a partly cloudy day with lower air temperature, with the exception of the west side cluster of LR treatment, both treatments showed similar NHH_d on the remaining cluster positions.

Figure 1 shows that the diurnal pattern of the berry thermal efficiency for anthocyanins accumulation was also very dependent on the meteorological conditions. On September 11th (sunny and high air temperature: min =17°C; max = 33°C) the maximum efficiency took place during the morning (9:00 to 11:00) and at late afternoon (18:00 to 19:00) while at early afternoon, the thermal efficiency reached the lowest value in both treatments. This NHH decrease observed at the hottest part of the day in both treatments are explained by the increase of berry temperature above the optimum temperatures for anthocyanins accumulation. A similar behaviour was observed by Rustioni et al. (2011), on sun exposed clusters. On September 19th (cloudy day with mild air temperature: min =11°C; max = 23°C) the berry thermal efficiency increased during the morning, reached the maximum at early afternoon and then decreased during the rest of the afternoon.

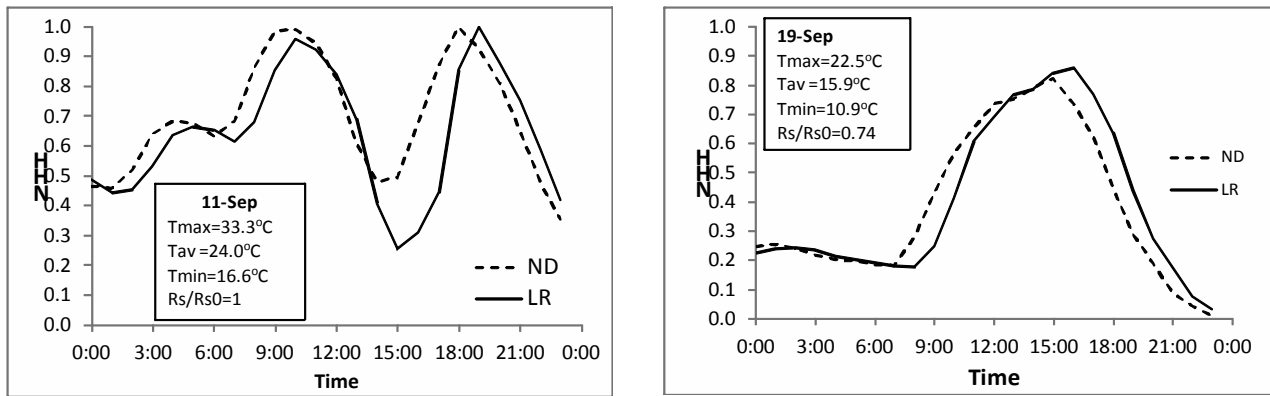


Figure 1. Effect of meteorological conditions (e.g. two contrasting days) on the diurnal pattern of the berry thermal efficiency for anthocyanins accumulation. LR - basal leaf removal; ND- control non-defoliated; Rs/Rs0-Relative shortwave radiation - ratio of the solar radiation (Rs) to the clear-sky solar radiation (Rs0) and NHH – normal heat hours.

Figure 1. Effet des conditions météorologiques sur l'évolution journalière de l'efficacité thermique des baies pour l'accumulation des anthocyanes. Effeuillage basal (LR); témoin non défolié (ND); Rs/Rs0 rapport du rayonnement solaire (Rs) et rayonnement solaire par ciel clair (Rs0) et NHH – heures de chaleur normalisées.

Berry composition at harvest

At harvest, the analysis of berry skin anthocyanins concentration of a composite sample of 100 berries collected from all the cluster positions show a significantly lower concentration on the LR treatment (932 mg/L) as compared to the control non-defoliated (1131 mg/L). These results are in line with the data on NHH presented above showing that in this terroir, the excessive cluster exposure on the defoliation treatment appears to have induced a negative effect on thermal efficiency for anthocyanins biosynthesis as also observed by Spayd et al. (2002).

4. Conclusion

Our results show that the thermal efficiency for berry anthocyanins accumulation was negatively affected by leaf removal and that this effect was dependent of the meteorological conditions, time of the day and berry/cluster location into the vine canopy. The defoliation, by its diurnal positive effect on berry temperature, induced a reduction on the NHH_c as compared to the control non-defoliated. This negative effect was recorded mostly on the exposed clusters located at both sides of the canopy (being higher on the west side) while the berries from the clusters located in the centre of the canopy showed almost no effect. These effects of leaf removal on NHH were much more pronounced in sunny and hot days (mainly during the hottest hours of the day) than on cloudy and mild temperature days.

These results show that, before deciding the level of leaf removal it is important to have a comprehensive understanding of the interaction of light and temperature on berry metabolism. Additional studies are needed to fully understand how canopy manipulations influence grape composition and wine sensory properties.

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