

Full Length Research Paper

# The effect of extreme water stress on leaf drying limits and possibilities of recovering in three grapevine (*Vitis vinifera* L.) cultivars

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This study was conducted in three grapevine cultivars (cv. Chardonnay, Merlot and Cabernet-Sauvignon) vines during the 2009 growing season in the ECOTRON of the campus of Montpellier SupAgro/France. The aim of this research was to analyze the effect of sudden and extreme water stress (EWS) to determine the limit of the leaf drying (depend on  $\text{leaf}$ ), possibilities of recovering, and its relationship with grapes composition. A randomized block design was used. There were three blocks with three replicates. In the experiment all plots consisted of totally 36 grapevines. During the entire experiment in ESW vines both  $\text{pd}$  and  $\text{md}$  values were close to each other by decreasing to about -2.1 MPa in all three varieties. After this level (-2.1 MPa) they both get close to each other until their equality in -3.7 MPa. This value was determined as the threshold of all leaf dryings in the vines. In EWS vines the lowest read  $\text{pd}$  and  $\text{md}$  was -4.6 MPa. In the next measurement  $\text{pd}$  and  $\text{md}$  were forced to -5.0 MPa, and water exit from leaf petiole was not observed in the same vines. After EWS treatment, berries became smaller and these lead to increase of Anthocyanin concentration, Folin-Ciocalteu index (FCI) and PTI values at harvest time. However as a result of EWS applications pH values increased to a level which had a negative effect on wine quality. There was a reduction in the values of 100 berry weight, berry volume, total soluble solids (TSS), sugar concentration, sugar content per berry, K and tartaric acid. The conclusion is that as a result of sudden EWS although all the leaves dried, vines did not die even they recovered by rewatering. However EWS had a negative effect on the berry quality.

**Key words:** Leaf water potential, extreme water stress, berry composition, recovering, leaf drying limits.

## INTRODUCTION

Water stress is one of the factors that may greatly influence grape and vine metabolism. If large portions of the soil become dry, the rate of shoot growth slows and the shoot tips gradually become more grayish green, like the mature leaves. As water stress continues, leaves appear wilted, particularly during mid-day heat. Under prolonged and severe stress, leaves curl, brown, and eventually drop. Vines that suffer severe water stress

begin to defoliate, exposing more of the berries that had been shaded by foliage.

Depending on the time and severity of water shortage, berries of stressed vines may not attain their full size. Water-stressed berries exposed to the sun can sunburn and shrivel. Water shortages also reduce the vine's ability to absorb nutrient from the soil. Symptoms of nutrient deficiencies are therefore more apparent during prolonged dry periods (Coggan, 2002; Selker and Baer, 2002).

Since development of the pressure chamber (Scholander et al., 1965), measurement of leaf water potential ( $\text{leaf}$ ) has been used as a tool to assess the water status of plants (Jones, 1990). Accordingly,  $\text{leaf}$  has been used to monitor the water relations of grapevine

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**Abbreviations:** EWS, Extreme water stress; TSS, total soluble solids; TPI, total phenol index; FCI, folin-ciocalteu index; WI, well irrigated.

(*Vitis vinifera* L.) (Smart and Coombe, 1983; Williams et al., 1994). It has been correlated with various aspects of grapevine physiology (Williams et al., 1994), vegetative growth (Schultz and Matthews, 1993), and reproductive growth and yield (Greenspan et al., 1996).

In vineyard leaf water potential is considered as the most practicable method for the control vine water status. Carbonneau (1998) and Deloire et al. (2004) use both pre-dawn ( $\psi_{pd}$ ) and mid-day ( $\psi_{md}$ ) leaf water potential as a criterion to evaluate vine water status at different developmental stages. Moreover Deloire et al. (2005) proposed different levels of  $\psi_{pd}$  for various vine styles. Leaf and vine response to water stress depend on both current situation and previous conditions.

By the time leaf wilting occurs, vines are severely stressed. The severity of water stress can affect vines either reversibly or irreversibly. Reversible effects are decreased in cell turgor pressure and shoot growth rate, and reduced in stomatal conductance, photosynthesis and berry size. As water stress intensifies, irreversible effects become apparent. These effects, in order of increasing water stress and severity include leaf chlorosis, defoliation, berry shriveling, vine death, irreversible reduction in berry size, decrease in fruit set, delay of sugar accumulation in fruit, reduce in fruit coloration, wood maturation, vine cold hardiness and bud fruitfulness (Coggan, 2002; Selker and Baer, 2002).

The effect of water stress on leaf photosynthesis has been extensively studied (Chaves, 1991; Lawlor, 1995). Inhibitions in plant growth rate, stomatal conductance and leaf photosynthesis as a result of soil drying are commonly observed (Davies and Zhang, 1991). Reduced photosynthesis of water stressed plants can be caused by stomatal closure, and/or altered pathway of photosynthetic process (Farquhar and Sharkey, 1982). In water-stressed grapevines, fully expanded sun-exposed leaves usually show large variation in photosynthesis both on a seasonal and a diurnal basis (Chaves et al., 1987; Escalona et al., 1999; Flexas et al., 1999). Severe water stress reduces photosynthesis and transpiration in all locations of the canopy except for most shaded leaves in the inner part. Photosynthetic radiation use efficiency strongly depends on both, pre-dawn leaf water potential and light-saturated stomatal conductance (Escalona et al., 2003).

Intensity of water stress is correlated to production levels. Environmental stress enhances grape quality, probably because it limits vine vigor. It further anticipates growth cessation and limits yield (Coipel et al., 2006). The water status of the grapevine can affect grape composition profoundly both directly or indirectly (Smart, 1974; Hidalgo, 1977) and in positive or negative way depending on the degree as well as the duration of water stress (Hardie, 1981) Berry solutes that are sensitive to vine water status include organic acids, sugars, anthocyanins and soluble phenolic compounds (De la Hera et al., 2005).

The manipulation of water limitation towards some

extreme values and short periods around veraison, allows to control berry size and to differentiate primary metabolites such as sugars from secondary metabolites such as polyphenols (Carbonneau and Bahar, 2009).

Much work was done concerning the effect of water stress on berry solutes and contradictory results were obtained. However the effect of sudden and extreme water stress (EWS) ( $-1.6\text{MPa}_{\text{leaf}} -5.0\text{MPa}$ ) on vine and berry metabolism was not studied before.

The aim of this research was to determine the effect of sudden and EWS and to determine the limit of the leaf drying (depend on  $\psi_{\text{leaf}}$ ), possibilities of recovering, and its relationship with grapes composition.

## MATERIALS AND METHODS

The experiment was conducted during the 2009 growing season on cv. Chardonnay, Merlot and Cabernet-Sauvignon grapevines (*Vitis vinifera* L.) grafted onto SO<sub>4</sub> in the Ecotron of the campus of Montpellier SupAgro/INRA, France.

A randomized block design was used. There were three blocks with 3 replications. The experiment consisted of 36 vines (18 stressed and 18 control vines). The eight-year-old potted grapevines were grown under natural conditions, had a volume of 70 L for individual vine. The containers were isolated from rainfall and the growing medium was a mixing of coarse sand and perlite and a controlled drainage. Vine spacing was 3.5 to 0.8 m and the vines were pruned as bilateral cordon on a Lyr. Six spurs with 2-3 node per vine were retained at pruning time for a shoot load of 10-12 shoots per vine.

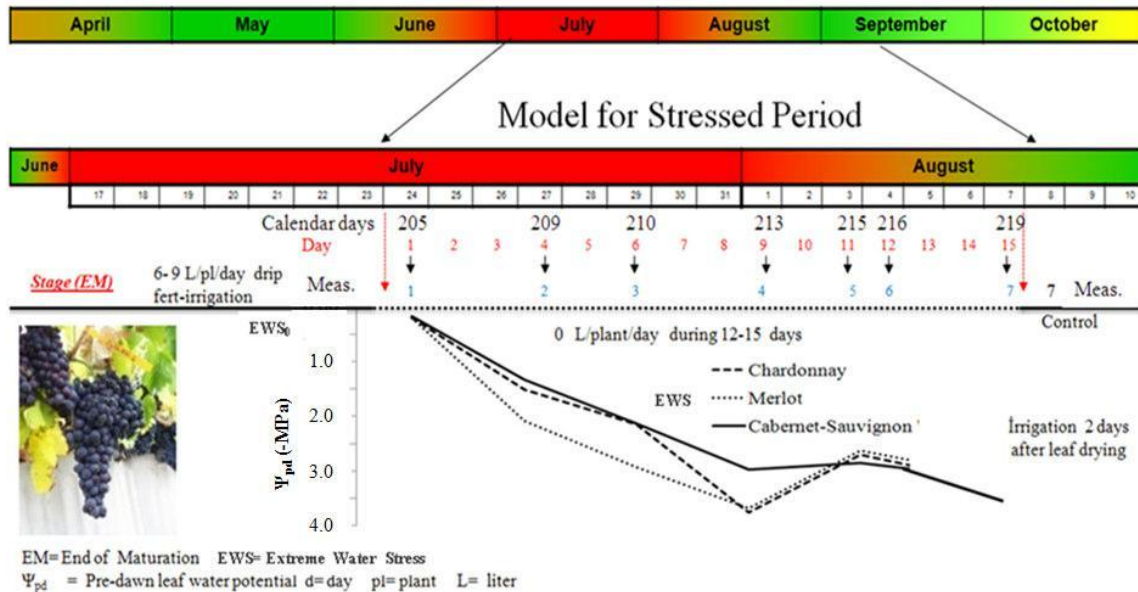
Rows were north-south oriented. Drip fert-irrigation was applied with two drip emitter for each plant. The calculated volumes of nutrient solution (between 6 to 9 L.day<sup>-1</sup>) for each day were applied regularly on each 6 h. Stressed period was started in 205th calendar day of growth under well irrigated (WI) conditions in the Ecotron at the end of maturation (EM) phenological stage (stage 36) (Eichhorn and Lorenz, 1977). Well irrigated [(WI)=(Control)] and EWS were established in relation to the reference of maximal transpiration, and monitored in function of the vine response measured as the predawn leaf water potential ( $\psi_{pd}$ ) (Carbonneau, 2001). Stressed vines were not irrigated during 12-15 days for each variety until all leaves get dry and fall. Vines were retained two more days not irrigated after all leaf drying. At the end of stressed period, potted vines were irrigated once on the saturation point. Besides that, cultivation practices were classical and examined possibilities of existence restoration after.

Harvest was done in early morning (7:00-9:00 AM) of September 7th (250th calendar day).

Net photosynthesis (A), stomatal conductance ( $g_s$ ) and transpiration (E) was measured with a portable infrared gas analyzer (Li-Cor 6200 Lincoln, NE, USA) at afternoon (13:00-15:00 PM). Measurements were taken for each vine, on mature, undamaged leaves that had grown fully exposed to the sun.

The  $\psi_{\text{leaf}}$  of each vine was determined with a Scholander pressure chamber (Scholander et al., 1965). Pre-dawn ( $\psi_{pd}$ ) and mid-day ( $\psi_{md}$ ) leaf water potential were measured 7 times on each of 1 to 3 consecutive days from beginning to the end of stressed period for each variety. Measurements were carried out until 9th day on freshly cut, healthy and fully expanded (mature) leaves from each vines for the stress levels.

While the leaves in the middle of shoot were drying, measurements in the upper mature leaves were done. After the first measurements, EWS vines water and mineral supply cut off until all leaves get dry and some fall. Water exit from petiole for both  $\psi_{pd}$



and  $\psi_{pd}$  leaf water potentials was determined as about -4.5 MPa. As to see whether there is a water exit from petiole, measurements were forced to -5.0 MPa. These modalities and their distribution over time were illustrated in Figure 1.

Harvest date was fixed on the basis of ripening dynamics of the berries related to sugar concentration ( $\text{g l}^{-1}$ ), titratable acidity ( $\text{g-tartaric acid l}^{-1}$ ) and pH. At harvest in early morning (07:00- 09:00 AM), 200 berries per treatment were sampled from different parts of various clusters and transported to the laboratory. Berry volume and chromatic characteristics were measured immediately after sampling by Dyostem apparatus (Sferis technology). After that, classical measurements were made on berries. Berries were weighed with an electronic balance and processed to determine 100-berry weight (g) and then juice extraction was analyzed for total soluble solid [(TSS), (Brix°)] were, pH and titratable acidity ( $\text{g-tartaric acid.l}^{-1}$ ). Total soluble solids (TSS) measured using refractometer equipped with a temperature control system (20°C). Juice pH was measured using a pH-meter. Titratable acidity was analyzed by pH-meter with a base to an end point of pH 7.0 (20°C), and results were expressed as a  $\text{g-tartaric acid.l}^{-1}$ . Besides that, tartaric acid ( $\text{g-tartaric acid l}^{-1}$ ), K ( $\text{g.l}^{-1}$ ), total phenol index (TPI), anthocyanins ( $\text{g.l}^{-1}$ ) and Folin- Ciocalteu Index (FCI) were analyzed also. Anthocyanin, TPI and total tannin index (Folin-Ciocalteu index) (FCI) were obtained by juice analyses using a diode array spectrophotometer.

Anthocyanins ( $\text{mg.l}^{-1}$ ) were analyzed as reported by Ribéreau-Gayon and Stonestreet (1965). The juice was centrifuged at 8000 rpm for 5 min at 15°C. Juices were diluted in a solution of HCl and Ethanol (98:2 v/v). Absorbance of the diluted juice samples was measured at 550 nm and the anthocyanin concentration was calculated with the formula:

$$\text{Anthocyanin (mg.l}^{-1}\text{)} = 15 \times A \times f$$

where A refers to the absorbance and f refers to the volume of the dilution.

TPI was quantified according to Ribéreau- Gayon (1970) by measuring the absorbance at 280 nm of must diluted 1:100 with distilled water, and the TPI was calculated with the formula:

$$\text{TPI} = A \times f$$

where A refers to the absorbance and f refers to the volume of the dilution.

Total tannin index (Folin-Ciocalteu Index) was analyzed according to Ribéreau-Gayon (1970) by measuring the absorbance at 750 nm, and the FCI was calculated with the formula:

$$\text{FCI} = A \times 100 \text{ for red varieties}$$

$$\text{FCI} = A \times 20 \text{ for white varieties}$$

where A refers to the absorbance.

Sugars ( $\text{g.l}^{-1}$ ) (depend on Brix°), and berry sugar loading or total quantity of sugar per berry ( $\text{mg.berry}^{-1}$ ) is estimated as:

$$\text{Sugar (mg.berry}^{-1}\text{)} = (1 \times 1.3^{-1}) \times [\text{sugar (g.l}^{-1}\text{)}] \times (1 \times 100^{-1}) \times [100 \text{ berries weight (g)}] \text{ (Carbonneau and Bahar, 2009).}$$

Potassium (K) ( $\text{g.l}^{-1}$ ) analysis were conducted by flame photometer and expressed as a  $\text{g.l}^{-1}$ .

Tartaric acid ( $\text{g.l}^{-1}$ ) were analysed according to Cemeroglu (2007).

MSTAT-C and Fishers protected least significant difference (LSD) tests were used to compare all treatments.

## RESULTS AND DISCUSSION

During the entire experiment in WI (Control) vines of Chardonnay  $\psi_{pd}$  (these units should be changed as not bold and italic characters) changed between -0.19 MPa and -0.26 MPa and  $\psi_{md}$  maintained between -0.82 MPa and -1.18 MPa.  $\psi_{pd}$  in WI vines of Merlot remained close to -0.28 MPa and was always -0.44 MPa and  $\psi_{md}$  changed between -0.85 and -1.4 MPa.  $\psi_{pd}$  and  $\psi_{md}$  in Control vines (WI) of Cabernet-Sauvignon remained - 0.2 MPa (av. - 0.18 MPa) and -1.32 MPa (av. -1.19 MPa)

respectively (data was not shown) (Figures 3, 4 and 5). Drought symptoms for extreme water stressed (EWS) vines were observed after 6th day for Merlot, 7th day for Chardonnay and 10th day for Cabernet-Sauvignon. The leaf drying and fall started from the base of shoots. For EWS vines in Merlot leaf water potential ( $\psi_{leaf}$ ) decreased more quickly than the other varieties. The decrease in  $\psi_{leaf}$  for EWS of Cabernet-Sauvignon was slower and lasted longer. According these results Cabernet-Sauvignon is more tolerant to sudden and severe water stress than Chardonnay and Merlot (Figures 3, 4 and 5). In Chardonnay the lowest read  $p_d$  in 9th day was -4.6 MPa. Although  $m_d$  measurement was forced to -5.0 MPa, water exit from leaf petiole was not observed in the same vines.

The average of the values for both  $p_d$  and  $m_d$  of Chardonnay in 9th day was -3.75 MPa and -3.65 MPa respectively. In Merlot the lowest values for  $p_d$  were -4.52 MPa and for  $m_d$  they were -4.42 MPa. The average values in the 9th day for  $p_d$  were -3.69 MPa while for  $m_d$  they were -3.86 MPa. In Cabernet-Sauvignon the highest  $p_d$  value was -4.37 MPa and for the same vine in -5.0 MPa ( $m_d$ ) any water exit in 15th day of experiment was not observed. The lowest  $m_d$  value was -5.03 MPa. The average of the values in 15th day for both  $p_d$  and  $m_d$  of Cabernet-Sauvignon were -3.45 MPa and -3.72 MPa respectively (data was not shown). Both  $p_d$  and  $m_d$  values were close to each other by decreasing to about -2.1 MPa in all three varieties. In Merlot this reduction was seen in 4th day, in Chardonnay and Cabernet-Sauvignon in 6th day. After this level (-2.1 MPa) they both get close to each other till their equality in -3.7 MPa. This value was determined as the threshold of all leaf dryings in the vines. In Chardonnay and Merlot all the leaves dried in 12th day as for Cabernet-Sauvignon the period was 15 days. Although all the leaves were dried and fallen because of the sudden, short-lasting (12-15 day) and severe water stress the vines did not die. The recuperation from the auxiliary bud at the top of shoots started about 7 to 10 days after irrigation of the vines.

Shoots from the auxiliary bud reached about 20 cm at the harvest time in all varieties (Figure 2).

Flexas et al. (1999), Lopes (1999) and Williams and Araujo (2002) obtained a linear relationship between A and  $p_d$  from -0.1 to -0.8 MPa. Pilar et al. (2007) obtained a constant slope within the range of their study (-0.2 to -0.8 MPa). In stressed vines a curvilinear relationship was found between  $p_d$  (from -0.1 to -4.6 MPa) and A,  $g_s$ , E because of the sudden and severe water stress as Zufferey et al. (2000). A,  $g_s$ , and E were found to decrease suddenly with sudden decrease in  $p_d$  (Figures 3, 4 and 6). The slope of the curve decreased when -0.5 MPa was reached. The 4th day values of  $p_d$  in all varieties were between -1.3 and -1.6 MPa. There was a sudden decrease in A,  $g_s$ , and E in the same day. A decreased from 8.48 to 2.37  $\mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Chardonnay, from 3.75 to 1.44  $\mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Merlot

and from 4.9 to 2.54  $\mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Cabernet-Sauvignon. Stomatal conductance ( $g_s$ ) decreased from 0.203 to 0.038  $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Chardonnay, from 0.124 to 0.034  $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Cabernet-Sauvignon and from 0.113 to 0.029  $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Merlot. E decreased from 10.02 to 1.00  $\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Chardonnay, from 7.03 to 0.86  $\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Cabernet-Sauvignon and from 6.49 to 0.77  $\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in Merlot. For A,  $g_s$  and E the minimum values were obtained in the 6th and 9th days. For all criterions the lowest values were measured in 6th day and after that the slope of curve nearly became horizontal.  $p_d$  values were below -2.1 MPa for both Chardonnay and Cabernet-Sauvignon. As for Merlot this value was about -3.0 MPa. According to these values A decreased below 0.8  $\mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ ,  $g_s$  below 0.0182  $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ , E below 0.92  $\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in all varieties. The 9th day  $p_d$  values for Cabernet-Sauvignon were about -3.0 MPa while for both Chardonnay and Merlot these values were about -3.7 MPa. In the same day A was about 1.2  $\mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ ,  $g_s$  was approximately 0.037  $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  and E below 1.45  $\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  in all varieties. In stressed vines there were not any measurements after the 9th day for A,  $g_s$ , and E because of extreme water stress and partial leaf drying (data was not shown). Also it was determined that transpiration was occurred when leaf value between 0 and -3.7 MPa. When  $\psi_{leaf}$  was between -3.7 and -4.6 MPa there was water in xylem but there was no transpiration from leaf while  $\psi_{leaf}$  was between -4.6 and -5.0 MPa there was no transpiration from leaf and no water in xylem.

Final berry size is an important factor which determines grape quality via the ratio skin area/juice volume (Champagnol, 1993); besides other factors, the grapevine water status strongly affects berry size. A significant differences were found in 100 berry weight ( $P>0.05$ ) and in berry volume ( $P>0.10$ ) at harvest, according to varieties and their stress levels (Figure 7). In WI (Control) vines 100 berry weight and berry volume is generally heavier and higher than stressed vines (EWS) respectively such as 28% Cabernet-Sauvignon, 21% Merlot and 14% Chardonnay. Ojeda et al. (2001) showed that water deficit modified the diameter and therefore the volume of berry which affected the of skin surface to juice content and as a consequence, the composition of must and wine. Final berry size is more influenced by water deficits of similar intensity between flowering and veraison than between veraison and maturity. During the ripening period the size of stressed berries recovers partially or totally, if water is available (Van Zyl, 1984; Naor et al., 1993; Poni et al., 1994; McCarty, 1997). When water deficits occurs from veraison through to harvest cause little reduction in berry size (Becker and Zimmermann, 1984). However in our study a berry shriveling in EWS was observed in all clusters in the 9th day. All the stressed berries recovered completely after the irrigation (data was not shown) (Figure 2). The size of stressed berries (EWS) became smaller than WI berries in the result of an extreme stress all the leaves dropped and occurred the



## Chardonnay



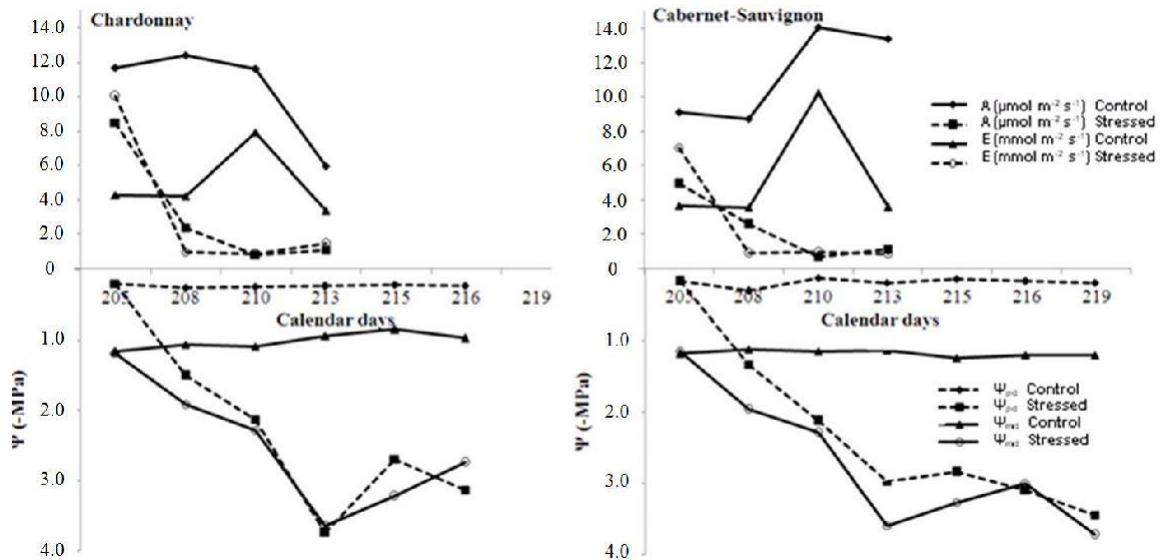
## Merlot



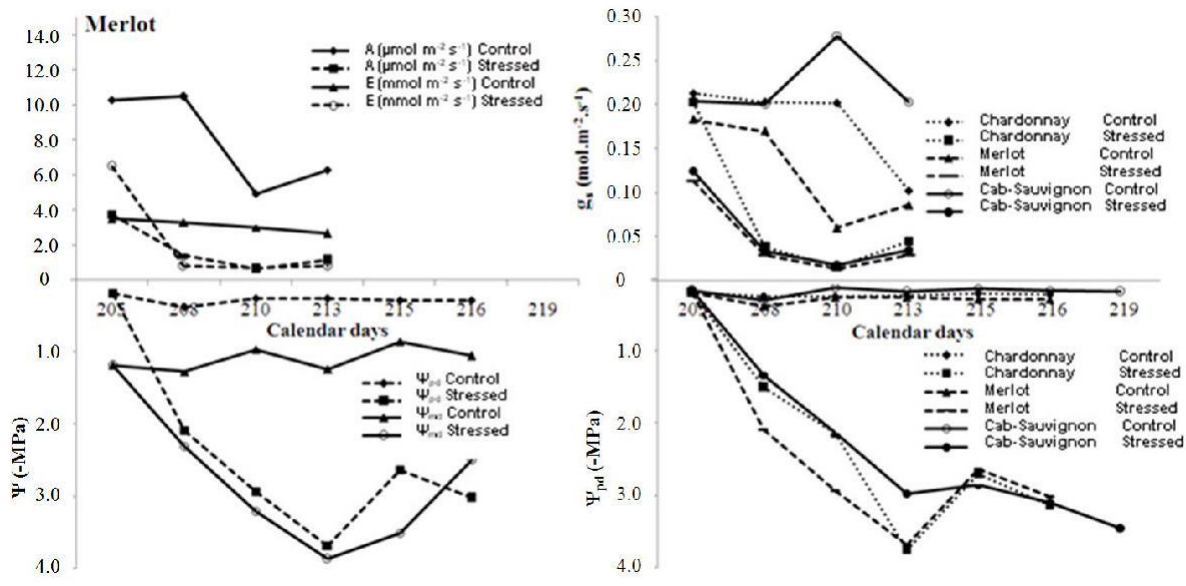
## Cabernet-Sauvignon



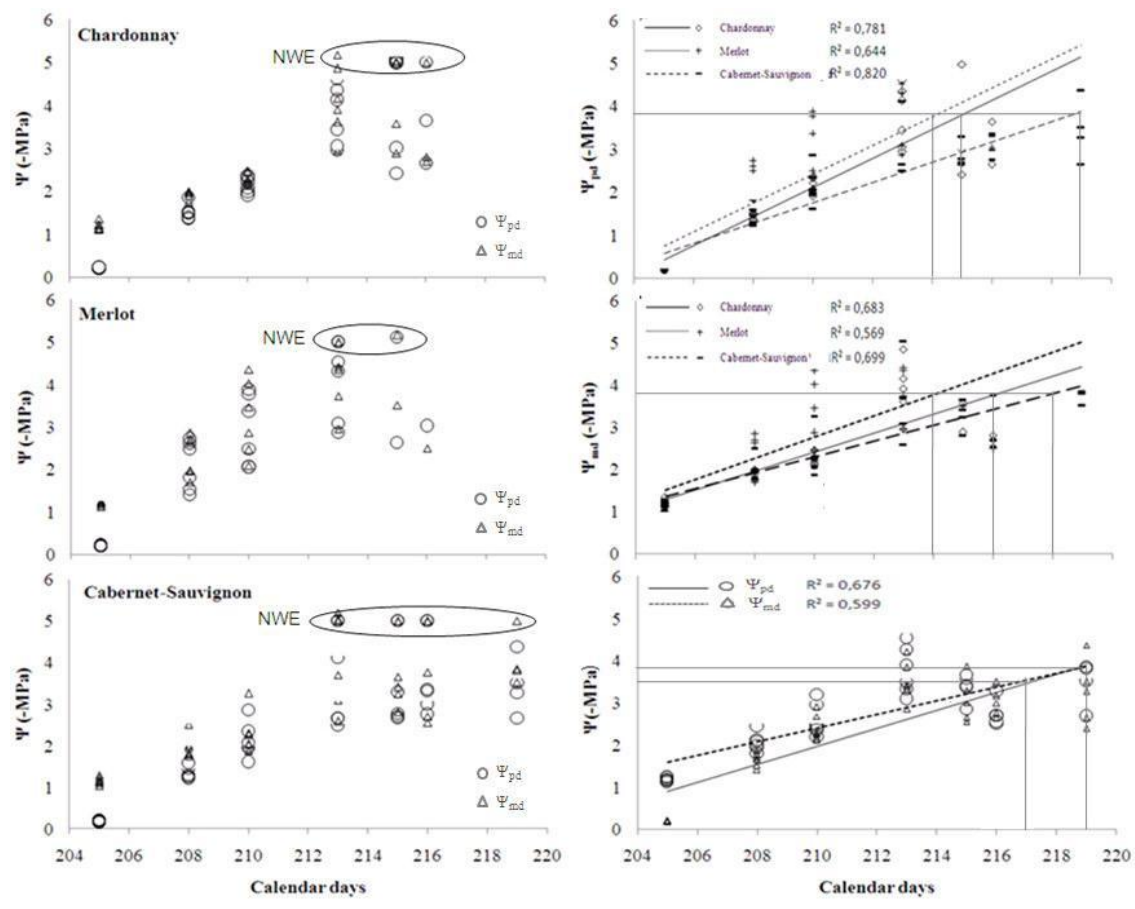
**Figure 2.** The effects of extreme  $p_d$  and  $m_d$  values on leaf drying, berry shriveling and recovering in three varieties.



**Figure 3.** The effects of extreme  $p_d$  and  $m_d$  values on A and E in varieties (From the figure we can present 3 assessments; (1) transpiration was occurred when  $\Psi_{leaf}$  value between 0 and -3.7 Mpa; (2) when  $\Psi_{leaf}$  was between -3.7 and -4.6 MPa there was water in xylem but there was no transpiration from leaf 3- when  $\Psi_{leaf}$  was between -4.6 and -5.0 MPa there was no transpiration from leaf and no water in xylem).

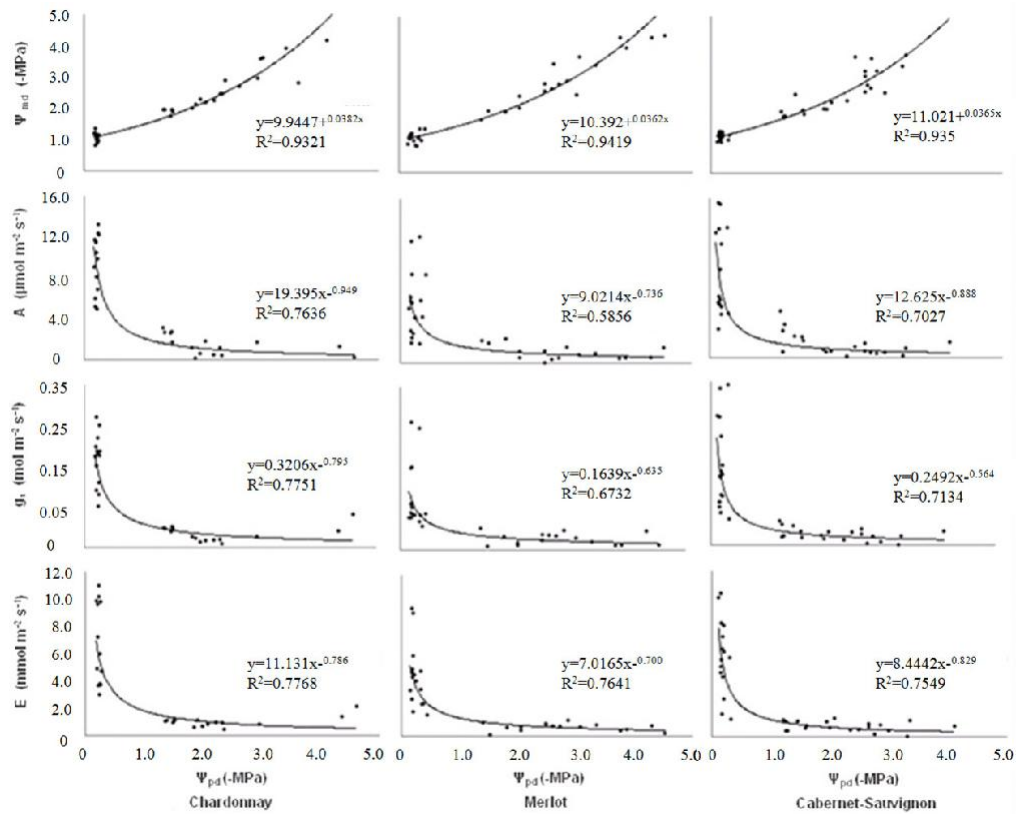


**Figure 4.** The effects of extreme  $\psi_{leaf}$  values on  $A$ ,  $g_s$  and  $E$  in varieties (From the figure we can present 3 assessments; (1) transpiration was occurred when  $\psi_{leaf}$  value between 0 and -3.7 Mpa; (2) when  $\psi_{leaf}$  was between -3.7 and -4.6 MPa there was water in xylem but there was no transpiration from leaf; (3) when  $\psi_{leaf}$  was between -4.6 and -5.0 MPa there was no transpiration from leaf and no water in xylem).

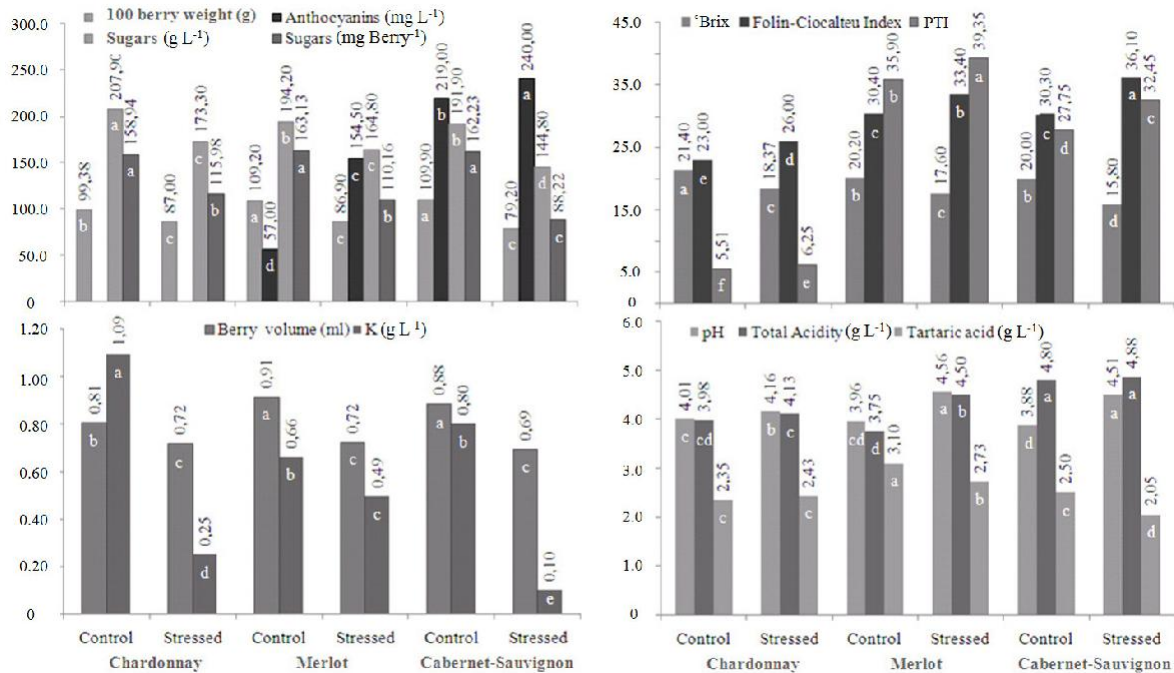


**Figure 5.** The effects of extreme  $\psi_{leaf}$  values on relationship between  $\psi_{pd}$  and  $\psi_{md}$  in three varieties (NWE = No water exit from leaf petiole).





**Figure 6.** The relationship between  $\Psi_{pd}$  and A,  $g_s$ , E,  $m_d$  in three varieties.



**Figure 7.** The final values of berry composition (100 berry weight:  $\text{LSD}(P=0.05)=8,710096$ , Anthocyanins  $\text{LSD}(P=0.05)=7,68455$ , Sugars concentration  $\text{LSD}(P=0.05)=9,056959$  Sugar per berry  $\text{LSD}(P=0.01)=8,48316$ , TSS (Brix)  $\text{LSD}(P=0.05)=0,811565$ , Folin-Ciocalteu Index  $\text{LSD}(P=0.01)=1,16877$ , PTI  $\text{LSD}(P=0.01)=0,6232009$ , Berry volume  $\text{LSD}(P=0.10)=0,066181711$ , K  $\text{LSD}(P=0.01)=0,1417342$ , pH  $\text{LSD}(P=0.01)=0,1157255$ , Total acidity  $\text{LSD}(P=0.01)=0,2714003$ , Tartaric acid  $\text{LSD}(P=0.01)=0,1829781$ ).

absence of both photosynthesis (A) and transpiration (E) and with the auxiliary buds burst after the irrigation there was a competition in consumption of carbohydrates between the new shoots and berries. 100 berry weight and berry volume changed between 86.9 and 109.9 g and between 0.69 and 0.91 ml respectively according to varieties and water stress applications.

As Girona et al. (2009) indicated, independently of phenological stage final dry matter accumulation is negatively affected by water stress. In EWS vines generally TSS (Brix°), sugar concentration ( $\text{g.l}^{-1}$ ) and the sugar content per berry ( $\text{mg.berry}^{-1}$ ) was lower than in WI vines. Sugar loading in the berry depends on environmental conditions and the grape variety associated with efficiently the vine uses its water and carbon supplies (Wang et al., 2003; Carbonneau, 1996). Significant differences were obtained according to varieties and stress levels interactions [TSS ( $P = 0.05$ ), sugar concentration of juice ( $P = 0.05$ ) and the sugar content per berry ( $P = 0.01$ )]. In all varieties TSS was over 20.00Brix° in WI vines while in EWS vines it was between 15.8 and 18.37Brix°. The highest sugar concentrations were determined in WI vines and they were between 191.9 and 207.9  $\text{g.l}^{-1}$  while in EWS vines these values were between 144.8 and 173.3  $\text{g.l}^{-1}$ . Similar results were obtained from the sugar content per berry ( $\text{mg.berry}^{-1}$ ), but their values showed a difference according to the berry size. While the highest sugar concentration in WI Chardonnay vines was 207.9  $\text{g.l}^{-1}$ , the sugar amount in the berry was lower (158.94  $\text{mg.berry}^{-1}$ ) than the other varieties (Figure 7). Deloire et al. (2001) showed that there was a relationship between the berry volume and the sugar loading.

The active berry sugar loading depends mainly on the vine water status and photosynthesis. Severe water stress tends to decrease vigor but also the sugar and acid content since photosynthetic activity may be compromised. As it was determined in our study, the sugar content also may diminish due to competition between vegetative growth and fruit development (De la Hera et al., 2005).

Decline in shoot growth, berry size, cluster weight, yield, trunk growth, cluster number, and berry titratable acidity correspond with a decline in  $\text{leaf}$  (Shellie, 2006). There were obtained significant differences ( $P = 0.01$ ) in pH, total acidity ( $\text{g-tartaric acid.l}^{-1}$ ), tartaric acid ( $\text{g.l}^{-1}$ ) and K ( $\text{g.l}^{-1}$ ) content in juice according to varieties and stress levels interactions (Figure 7). In EWS vines pH and total acidity was higher than WI vines, especially pH increased a lot and according to varieties reached the values between 4.16 and 4.56. As for WI vines these values were between 3.88 and 4.00. In EWS vines total acidity was higher than WI vines. The highest acidity level was determined in EWS vines of Cabernet-Sauvignon as 4.88  $\text{g-tartaric acid.l}^{-1}$ , while the lowest one was 3.75  $\text{g-tartaric acid.l}^{-1}$  in WI vines of Merlot. The tartaric acid ( $\text{g.l}^{-1}$ ) and K ( $\text{g.l}^{-1}$ ) levels in EWS vines were generally lower than in WI vines. During the course of berry development, potassium

may play different roles depending on developmental stage. After veraison, grape berries continue to enlarge, but presumably the cell expansion during this phase of development is driven by the increase in sugar in the cell vacuole and potassium may play a secondary role in the accumulation of sugars (Davies et al., 2006). Grape berry potassium accumulation is important because elevated levels of berry potassium can have a negative effect on wine quality by increasing berry and wine pH (Gawel et al., 2000). It is assumed that in EWS vines the falling of all leaves during the stressed period decreased tartaric acid ( $\text{g.l}^{-1}$ ) and K ( $\text{g.l}^{-1}$ ) levels, increased pH values and inhibited the decrease of total acidity. K content of juice was between 0.1 and 0.49  $\text{g.l}^{-1}$  in EWS vines. In grape berries, potassium is the most abundant cation ( $-0.9 \text{g.l}^{-1}$ ) where it contributes to charge balance and may be involved in sugar transport (Lang, 1983; Blouin and Cruège, 2003).

The effect of water stress on the metabolism of anthocyanins and phenols depend on the degree of water stress, the point in time at which it is applied and its duration (Deloire et al., 2004). According to varieties and stress levels interactions significant differences were obtained in Anthocyanin concentration ( $P = 0.05$ ), FCI ( $P = 0.01$ ) and PTI ( $P = 0.01$ ) values. Generally there was an increase in all three criterions in EWS vines.

Increased fruit exposure to sunlight generally improves berry composition and wine quality (Carbonneau, 1995). Anthocyanin concentrations in EWS vines of Cabernet-Sauvignon and Merlot were 240.00 and 154.5  $\text{mg.l}^{-1}$ , while in WI vines these values for Cabernet-Sauvignon and Merlot were 219.00 and 57.00  $\text{mg.l}^{-1}$  respectively. For FCI as like in Anthocyanin concentration in ESW vines has higher values. The highest two index values in ESW were determined for Cabernet-Sauvignon (36.10) and for Merlot (33.40). Merlot has the highest PTI values in both WI (35.90) and EWS (39.35). Chardonnay has at least 6 times lower values than Merlot.

## Conclusion

The present study showed the importance of sudden and extreme water stress (EWS) on the vines and berry composition of three wine cultivars. The obtained results showed that both  $p_d$  and  $m_d$  values were close to each other by decreasing to about -2.1 MPa in all three varieties.

After this level (-2.1 MPa) they both got close to each other until their equality in -3.7 MPa. This average value was determined as the threshold of all leaf dryings in the vines. Also it was determined that transpiration was occurred when  $\text{leaf}$  value between 0 and -3.7MPa. When  $\text{leaf}$  was between -3.7 and -4.6 MPa there was water in xylem but there was no transpiration from leaf while  $\text{leaf}$  was between -4.6 and - 5.0 MPa there was no transpiration from leaf and no water in xylem. Although all the leaves were dried and fallen because of the sudden and EWS



the vines did not die and recovered. The vines recovering started from the auxiliary bud at the top of shoots after rewatering of the vines.

The results also indicate that after EWS treatment because all the leaves were fallen the clusters were fully exposed to sun light, berries became smaller and these lead to increase of Anthocyanin concentration, FCI and PTI values at harvest time. Similar to this there was an increase in pH and total acidity values. However as a result of EWS applications pH values increased to a level which had a negative effect on wine quality.

On the contrary there was a reduction in the values of 100 berry weight, berry volume, TSS, sugar concentration, sugar content per berry, K and tartaric acid.

The conclusion is that as a result of sudden EWS although all the leaves dried, vines did not die even they recovered by rewatering. However EWS had a negative effect on the berry quality. These results must be reinforced under different conditions and extended to the study of longer extreme stress.

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