

## ADJUSTMENTS OF WATER USE EFFICIENCY BY STOMATAL REGULATION DURING DROUGHT AND RECOVERY OF VERONA PROVINCE GRAPE VARIETIES GRAFTED ON TWO DIFFERENT *VITIS* HYBRID ROOTSTOCKS

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### ABSTRACT

Drought is considered to be the predominant factor both for determining the geographic distribution of vegetation and for restricting crop yields in agriculture. Furthermore, water stress is a limiting factor for a wide range of plant physiological processes and can have a profound effect on plant metabolism and development. Drought stress can decrease the sensitivity of photosynthesis to subsequent water deficits and similarly reduce the sensitivity of stomata to low atmospheric vapor pressure deficit during the dry season. Grapevine cultivars are known to differ in their drought adaptation mechanisms, but there is little knowledge on how many of them behave during a drought event and after recovering. The aim of this study is to analyze how stomatal conductance is regulated under water stress and recovery, as well as how water stress affects adjustments of water use efficiency in cultivar Corvina, Corvinone and Rondinella grafted on Kober 5BB and 140 Ruggeri rootstocks. The experiment was conducted on 4-year old vines, grown in an experimental field of Valpolicella in Verona province. The effects of water deficit and recovery after rewatering were evaluated by using thermal imaging, a potential tool for estimating plant temperature, which can be used as an indicator of stomatal closure and water deficit stress. The thermal indices were compared with measured stomatal conductance. Results of mid-morning and at noon measurements showed significant difference between cultivars for both stomatal conductance and canopy water stress index. An apparent difference between the cultivars was the highest speed of the recovery noted for Corvinone compared to Corvina and Rondinella.

### KEY-WORDS

Stomatal conductance, photosynthesis, water stress, recovery, grapevines.

### INTRODUCTION

Drought stress has profound effects on plant physiology such as photosynthesis; stomatal conductance and transpiration depend on the rapidity severity and duration of the drought event (Vadell and Medrano, 1992). Grapevine (*Vitis vinifera* L.) has developed various physiological and morphological mechanisms in order to sustain growth and productivity under water-limited conditions. One of the first vine responses to drought is the reduction of leaf stomatal conductance associated with an optimization of water use efficiency. In fact, this response depends on some factor such as the rootstock. Grapevine rootstock has important effect on gas exchange of field-grown grapevines due to differences in the relative capacity of rootstocks to extract and provide scions with water. Rootstocks have been reported to affect the efficiency of water transport to the shoots via conductivity constrains imposed by the anatomy of xylem vessels (de Herralde et al., 2006).

A technique to measure plant water stress should values provide non destructive, rapid, and reliable estimates of plant water status. Therefore, thermal imagery provides an ideal approach for the collection of the large number of individual leaf temperatures that are

necessary for methods based on temperature frequency distributions. Thermal imaging also allows leaves to be distinguished from the background.

Despite these recent significant improvements in the hard- and software used in thermal imaging, there is a current lack of knowledge in linking remotely measured canopy temperature and crop water stress index (CWSI) to true ground-based measures of crop water stress, such as leaf conductance and water potential in the leaf or stem. However, knowledge of these relationships is required in order to translate thermal imagery data accurately into water-stress estimates, which can then serve as irrigation decision support tools (Moller et al., 2007).

In the present study, the potential of using thermal images for the on-field estimation of crop water status of grapevine under different rootstocks and different soil types was investigated. The specific aims were to compare thermal based CWSI estimates with plant water status parameters, to compare the effect of soil types, to compare the effect of the rootstocks.

## MATERIALS AND METHODS

**1. Plant material and experimental conditions:** The field experiments were carried out at Negrar municipality of Verona province, Italy during the summer of 2009. Measurements were made on grapevines (*Vitis vinifera*) with cv. Corvinone cl. 8, Corvina cl. 48 and Rondinella cl. 76 grafted on Kober 5BB and 140 Ruggeri grapevine rootstocks planted in 2004 in a East-West direction with a row and vine spacing of 2.0 m and 1.0 m, respectively. The vineyard was established on two soil textures, loamy and clay soil. The region has a climate with less than 80 mm of summer rainfall. The average midday temperature and air humidity during the summer months were 31°C and 45%, respectively.

**2. Thermal imaging:** Infrared thermography pictures were obtained from the canopies of each cultivar using an infrared camera (Therma CAM<sup>TM</sup> FLIR systems, UK) with a built 24° lens. Reference leaves were used to avoid inclusion of non-leaf material in the picture analysis such as trunks, clusters, soil, sky, etc. For this purpose two mature representative leaves were chosen and painted 1 minute before imaging, on both abaxial and adaxial sides. One leaf was sprayed with water and the second leaf was covered with Vaseline to prevent transpiration, to obtain the 'wet' temperature ( $T_{wet}$ ) and 'dry' ( $T_{dry}$ ) respectively. Infrared thermography pictures were obtained at predawn and midday during stress water conditions and after rewatering. Imaging was taken on the sun side. Analysis of thermal images was performed using the ThermaCAM Reporter Software.

**Calculations:** thermal indices: Based on the temperature differences between the experimental plant, the 'dry plant', and the 'wet plant', the following thermal indices were calculated and used as a comparison with the measured stomatal conductance (Jones, 1999). An index analogous to Idso's (1982) crop water stress index (CWSI) was also calculated.

$$CWSI = (T_{leaf} - T_{wet}) / (T_{dry} - T_{wet}) \quad (1)$$

$$IG = (T_{dry} - T_{leaf}) / (T_{leaf} - T_{wet}) \quad (2)$$

Where  $T_{leaf}$ ,  $T_{dry}$ , and  $T_{wet}$  are the mean temperatures of the leaf area of the experimental plant, the dry reference plant, and the wet reference plant, respectively.

**3. Gas exchange measurements:** Photosynthesis, transpiration, stomatal conductance, evaporation, intercellular CO<sub>2</sub> concentration and leaf temperature were recorded at predawn and midday using the LCI-301 portable gas exchange system (ADC BioScientific Ltd, Hoddesdon, UK). Measurements were taken on three fully expanded, recently matured, sun-lit leaves per cultivar. Gas exchanges were measured with an open system. Gas exchange measurements were conducted at predawn and midday during stress water conditions and

after rewatering in order to obtain an accurate indication of grapevine response to environmental conditions (Medrano et al., 2003).

**4. Plant water status:** Leaf water potential was measured using a pressure chamber, according to Choné et al. (2001), One leaf per plant was obtained from the third and fourth fully expanded leaf from the tip with three plant replicates for each treatment. After cutting, the leaf was immediately enclosed in a plastic bag and the determination of the leaf water potential was started as described by Meron et al. (1987). These measurements were carried at the pre-dawn and midday during water stress and rewatering periods.

**5. Statistical analysis:** A four-factor (cultivar, rootstock and soil texture) analysis of variance (ANOVA) was used to test the main effect and interactions on leaf water status, gas exchange and transpiration and crop water stress index, using STATISTICA software (version7).

## RESULTS AND DISCUSSION

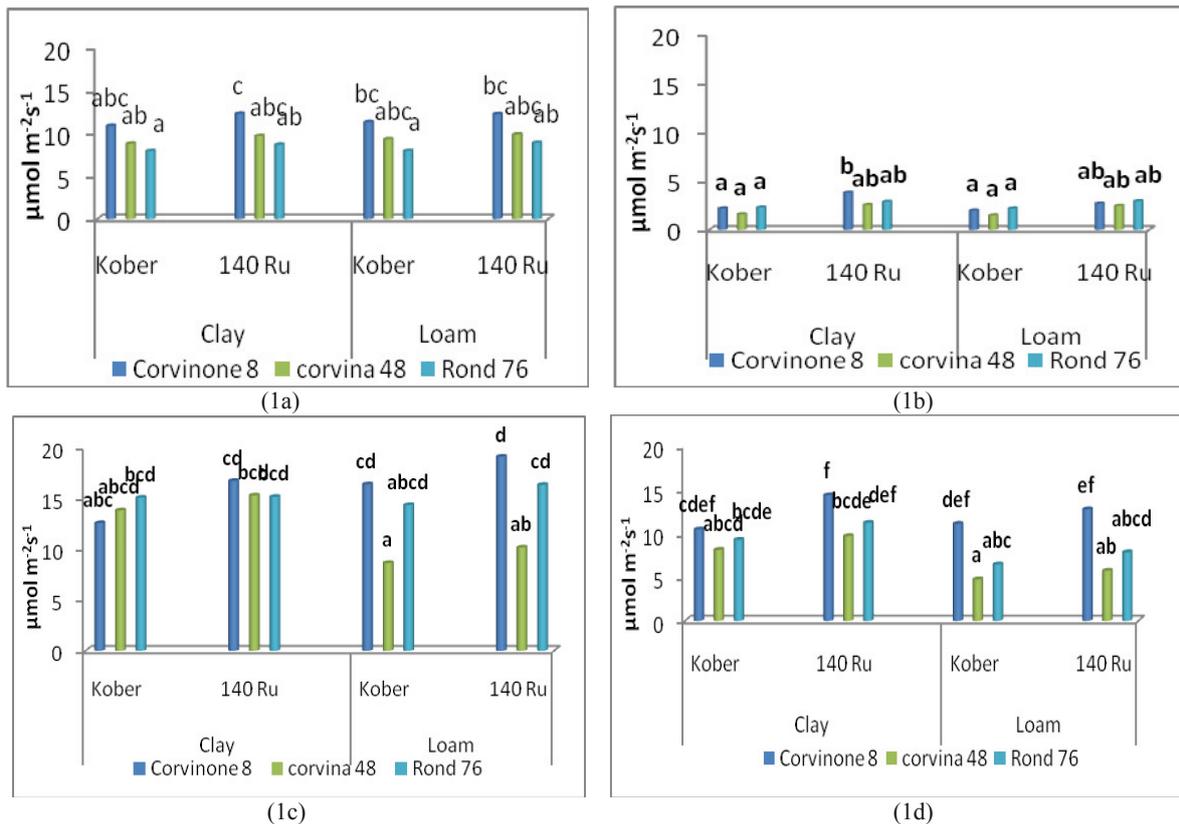
### 1. Photosynthesis

Differences between cultivars, rootstocks and soil textures concerning the effect of water deficit on plant photosynthesis has been discussed (Fig.1a and 1b). No significant differences were recorded at midday between Rondinella cl. 76 and Corvina cl. 48 cultivars and between Corvina cl. 48 and Corvinone cl. 8 cultivars. In contrast, Corvinone cl. 8 grafted on 140 Ru. rootstock and planted in clay soil was highly significant comparing to Rondinella cl. 76 cultivar grafted on Kober 5BB rootstock and cultivated in both loam and clay soil. Although, photosynthesis of Corvinone cl. 8 cultivar tend to be higher followed by Corvina cl. 48 which followed by Rondinella cl. 76 for all the treatment. Regarding the rootstocks, photosynthesis recorded by the 140 Ru. was higher comparing to the Kober 5BB for all the cultivar and under the two soil types. The same result was described by Paranychianakis et al., (2004) which demonstrate that under water stress conditions, grapevine rootstocks have different response from the point of view of stomatal conductance and photosynthesis. In fact, grapevine plants cultivated in clay soil show high photosynthesis values comparing to the others planted in loam soil for both predawn and midday measurement.

In the other hand, photosynthesis measured at midday decrease comparing to which measured at predawn due to increase of temperature and stomatal closure. The decrease of photosynthesis between predawn and midday was not similar for all the treatment. Fig. 2 of midday showed that Corvina cl. 48 cultivar was affected more by water deficit. In contrast, the photosynthesis decrease rate of Rondinella cl. 76 was lower comparing to the other cultivars and it maintain the highest photosynthesis value for all the treatment except for the plants grafted on 140 Ru. rootstock and cultivated in clay soil, where the Corvinone cl. 8 cultivar showed the highest photosynthesis value and it was highly significant comparing to cultivar grafted on Kober 5BB rootstock and planted in both loam and clay soil.

The intensity and effect of rewatering on the different treatment differs according to plant state. In fact, at predawn the cultivar which was more stressed (Rondinella cl. 76) responds better under water recovery condition comparing to the other (Fig.1c and 1d). Previous research with different species agrees with our results reporting that photosynthesis rate after rewatering is lower than in plants that have low affected by water deficit (Anyia and Herzog, 2004). Parra Quezada et al. (1999) explain this behaviour as a result of the lasting effect of the chemical signalling from the previous water deficit or due a damage in photosynthetic metabolism (Lauriano et al., 2004). The highest photosynthesis value was recorded usually for Corvinone cl. 8 except for the cultivar grafted on Kober 5BB and planted in the clay soil and Corvinone cl. 8 grafted on 140 Ru and planted in clay soil was highly significant difference comparing the most of treatments. In contrast at morning Corvinone cl. 8 was

highly significant comparing to the others treatments mainly comparing Corvina cl. 48 cultivars grafted on 140 Ru. and Kober 5BB and planted in loamy soil. Same as other parameters, photosynthesis in clay soil was higher comparing to the loam soil, cultivars grafted on 140 Ru. rootstock recorded high photosynthesis during drought and recovery conditions and during predawn and midday measurement. Corvinone cl. 8 cultivar show higher photosynthesis values for most the treatment. Therefore, it can be stated grapevine genotypes differ in their ability to acclimate to water deficits, as it has been reported earlier for different species (Lauriano et al., 2004).



**Fig. 1.** Effect of rootstocks, cultivars and soil texture on photosynthesis activity under water deficit condition at predawn (1a) and midday (1b) and rewatering at predawn (1c) and midday (1d).

## 2. Stomatal conductance

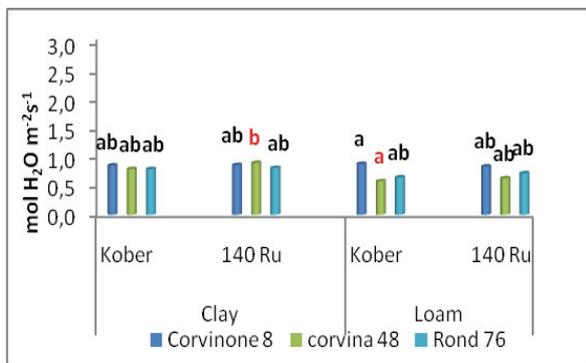
Stomatal conductance can be used as integrative parameter for determination of the degree of water deficit. In fact, the response of stomatal conductance parameter measured during the stress period is summarized in the Fig. 2a and 2b for both predawn and midday, respectively. For both, grapevine plants planted in the clay soil resist more to the water stress during predawn and midday; this result is due to the clay soil characteristic to water retention capacity. Soil water content explained nearly 60% of the reduction in gas exchange in summer.

According to rootstock point of view, 140 Ru. resist more than the Kober 5BB rootstock to water stress either in the clay and in the loam soil. In fact, the stomatal conductance of Corvinone cl. 8 cultivar tend to be the highest followed by Rondinella cl. 76 and Corvina cl. 48 cultivars for loam and clay soil and for the Kober 5BB and 140 Ru. rootstocks, the same for the predawn and midday measurement but the significant difference between Corvina cl. 48 grafted on Kober 5BB rootstock and planted in the loam soil and Corvinone cl. 8 grafted

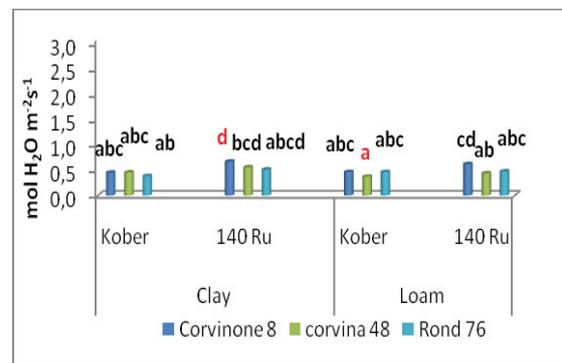
on Kober 5BB rootstock and planted in the loam soil and Corvina cl. 48 cultivar grafted on 140 Ru. rootstock and planted in the clay soil. At midday, the more important significant difference was recorded between Corvina cl. 48 grafted on Kober 5BB rootstock and planted in the loam soil and the and Corvinone cl. 8 grafted on 140 Ru. and planted in the clay soil. Similar result was reported by Williams et al. (1994), which indicate the importance of the relationship between soil water content and midday stomatal conductance of Thompson seedless grapevine.

Otherwise, the difference between predawn and midday stomatal conductance is remarkable difference for the predawn value, this difference is due to the increase of temperature which cause the increase of leaf temperature and the closure of the stomate as response for the temperature increase, which can be used as indicator of the leaf stomatal conductance and plant water stress (Idso et al., 1981, Jones 1999).

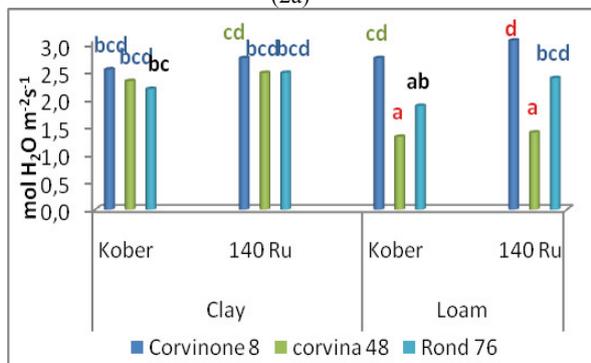
Under recovery conditions (Fig. 2c and 2d), stomatal conductance return to increase comparing to stress period for all cultivars and under different soil types, for both predawn and midday measurement. But during the recovery period the stomatal conductance value distribution differs to the stress period such as Corvinone cl. 8 cultivar grafted on 140 Ru. in the loam soil become the highest with highly significant difference comparing to Corvina cl. 48 grafted on both 140 Ru. and Kober 5BB and planted in the loam soil, which has the lowest values during the predawn measure uptake. But during the midday like which recorded at stress period highly significant difference was recorded between Corvina cl. 48 grafted on Kober 5BB and planted in loam soil and Corvinone cl. 8 grafted on 140 Ru. and planted in clay soil which determines the important effect of clay soil and 140 Ru. rootstock where the most measured values of stomatal conductance were recorded. Other differences respect to our results and those of Williams et al. (1994) may be due to differences in the time of day, when measurements were taken, different stomatal behaviour among cultivars under drought conditions (Schultz, 2000).



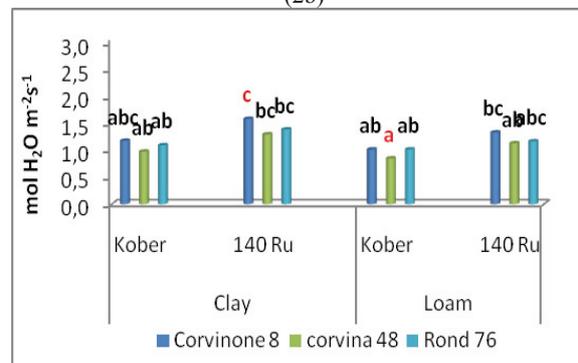
(2a)



(2b)



(2c)



(2d)

**Fig. 2.** Effect of rootstocks, cultivars and soil texture on Stomatal conductance activity under water deficit condition at predawn (2a) and midday (2b) and rewatering at predawn (2c) and midday (2d)

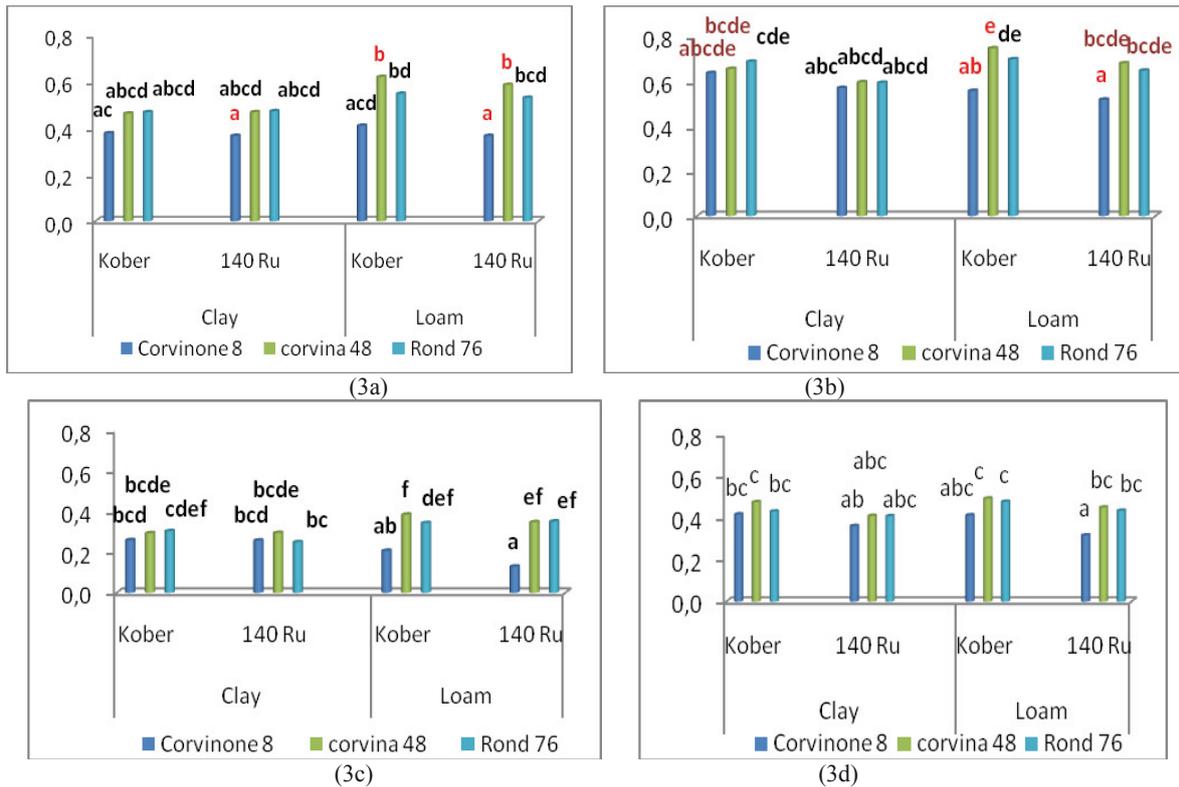
### 3. Crop Water Stress Index (CWSI)

Fig. 3a and 3b includes the values obtained from infrared camera during stress period at predawn and midday respectively, and the CWSI is calculated using Formula (1). There was no important difference among all the treatments except Corvinone cl. 8 grafted on 140 Ru. rootstock and planted in clay soil and the same cultivar grafted on the same rootstock, but planted in the loam soil show a lower CWSI which demonstrate the important effect of the Corvinone cl. 8 cultivar and 140 Ru. rootstock to resist to water stress. In contrast, Corvina cl. 48 cultivar grafted on 140 Ru. and Kober 5BB rootstocks and planted in the loam soil show the higher values of CWSI at morning during the stress period with significant difference comparing to the cultivars cited before. CWSI values of Rondinella cl. 76 cultivar grafted on both Kober 5BB and 140 Ru. Rootstocks planted in loam soil tend to be higher comparing to the other cultivated in clay soil, which demonstrate the effect of soil texture to resist to water stress. Variability in temperatures between plants in the same management treatment has been noted to increase with stress, for example, Gardner et al. (1981), probably as a result of variation in soil properties.

At midday CWSI values were increased for all the treatment and keeping the same shape of histograms distribution as morning measurement. This increase of CWSI is due to decrease of quantity of water in the leaves and the increase of leaf temperatures. The more important significant difference which recorded at midday was between Corvinone cl. 8 cultivar grafted on 140 Ru. rootstock and planted in the loam soil comparing with Corvina cl. 48 cultivar grafted on Kober 5BB and cultivated in the same soil texture, which was highly significant comparing the first one (Fig. 3b). Similar result was recorded by Jones et al., (2002) which reported that the difference between CWSI on the tested grapevine cultivar were between 6,7% and 16%. This variation demonstrate the important effect of the soil (clay soil) and the rootstock (140 Ru.) associated with cultivar (Corvinone cl. 8 followed by Rondinella cl. 76) to resist to water stress.

Rewatering after stress condition has been reached, result in the recovery of CWSI and intensity was not the same for all the cultivars for both predawn and midday measurement. Such as CWSI value of Rondinella cl. 76 cultivar grafted on 140 Ru. rootstock and planted in the clay soil recorded important decrease comparing to both the same cultivars and the cultivars the same for Corvinone cl. 8 grafted on 140 Ru. rootstock and planted in the loam soil (Fig. 3c). In fact, at midday Rondinella cl. 76 cultivar grafted on Kober 5BB rootstock has low CWSI value comparing to the other cultivars grafted on the same rootstock and planted in the same soil texture (Fig. 3d), which demonstrate of the importance of the intensity of Rondinella cl. 76 variety to response to rewatering.

The same as stress condition the lowest CWSI values was recorded on the Corvinone cl. 8 cultivar grafted on 140 Ru. with significant difference comparing to Corvina cl. 48 grafted on Kober 5BB and both of cultivar were planted in the loam soil for both predawn and midday CWSI measurement. The CWSI varied considerably within the same treatment, and even between two canopies of the same treatment imaged in quick succession (Grant et al., 2007). For the other treatment no important difference was recorded between each other but the most cultivars planted in the clay soil has low CWSI values comparing to the other planted in the loam soil and the cultivars grafted on 140 Ru. rootstock has low comparing to Kober 5BB.



**Fig. 3.** Effect of rootstocks, cultivars and soil texture on Crop Water Stress Index under water deficit condition at predawn (3a) and midday (3b) and rewatering at predawn (3c) and midday (3d).

## CONCLUSION

The present study shows that leaf water potential can be well estimated by combining temperature values derived from thermal images with the study of gas exchange under water stress and recovery conditions. Thus, the presented methodology has proved to be useful in order to contribute to evaluate the behavior of different cultivars and rootstocks to drought and recovery, and where irrigation is required according to soil characteristics.

Corvinone cl. 8 variety grafted on 140 Ru. rootstock and planted in clay soil shows highly photosynthesis comparing to Rondinella cl. 76 and Corvina cultivars grafted on Kober 5BB rootstock and cultivated in both loam and clay soil.

Also the stomatal conductance of Corvinone cl. 8 cultivar tend to be the highest followed by Rondinella cl. 76 and Corvina cl. 48 cultivars for loam and clay soil and for the Kober 5BB and 140 Ru. rootstocks.

In contrast, Corvina cl. 48 cultivar grafted on 140 Ru. and Kober 5BB rootstocks and planted in the loam soil show the higher values of CWSI at morning during the stress period with significant difference comparing to the Corvinone and Rondinella cultivars .

An apparent difference between the cultivars was the highest speed of the recovery noted for Corvinone variety compared to Corvina and Rondinella cultivars.

The 140 Ru rootstock shows a higher resistance to drought of Kober 5BB, but the speed of recovery of the initial conditions of rehydration it is very similar between the two and maybe with a slight prevalence of K 5BB.

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