

Grapevine rootstocks differently affect physiological and molecular responses of the scion under water deficit condition

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SUPPLEMENTARY MATERIALS

Supplementary Materials and Methods

Plant material and experimental conditions

Two years old grafted (CS/M4, CS/101.14) and autografted (CS/CS) vines were used for the experimental trial. In particular, bench-grafting material was provided directly by commercial nursery production as bare-root vines. Bare-root vines were arranged in 3-liters pots in winter 2013.

Potted vines were grown for one entire season outdoor at the experimental farm of University of Milan. At winter time, before the start of the experimental season (at the end of winter 2014), the potted vines were transplanted, root were thinning out and trimmed to ca 10cm in length and vines were arranged in 3-L pots filled with a sand–peat mixture (7:3 in volume) and then moved into the greenhouse sited in Milan (Italy) equipped with supplementary light and a cooling system, with a 16 h light (~PPFD of 600 μmol of photons $\text{m}^{-2} \text{s}^{-1}$) and an 8-h dark photoperiod. Plants were fertilised monthly with 100 mL of solution containing 0.54 g KNO_3 , 0.084 g NH_4HPO_4 , 0.42 g MgSO_4 and 0.01 g of a microelement mixture (OligoGreen, GREEN Italia, Canale d'Alba, Italy). During this period, plants were managed by shoot thinning and lateral shoot removal to provide uniform material.

Sample pots were selected during soil preparation and were weighed before and after saturating the soil with water at 100% field capacity (Fc 1.0) to calculate the soil water content (g of water/g of dry soil in the pot) at Fc 1.0. Plants used for the experiment were randomly arranged in greenhouse benches. At the 5-separated-leaves stage (stage 15 according to the extended BBCH scale), the plants were thinned to two shoots and trained vertically on jute threads. This training method allow to lift the pots onsite, placing the scale underneath and carry out the weighing procedure twice a day

without handling and moving the pots from their position. Before to start the experiment, plants of each graft combinations were maintained in well-watered conditions (at 80% of soil Fc 1.0) for 5 days. In this time and for the duration of the experiment, plants were daily irrigated with municipal tap water (pH of 5.5). In order to mitigate the fluctuations in soil water content, all pots were weighted twice a day, at 8:00 AM and at 6:00 PM and the amount of water needed to maintain the desired soil field capacity was then added.

At the starting of the experiment, vines of different treatments were 1.20 m in height with approximately 25-28 leaves unfolded and resulted very homogeneous in size among and within treatments.

Half the plants of each graft combination were maintained in well-watered conditions (at 80% of soil Fc 1.0), while half the plants were subjected to water deficiency (WD). The water supply was progressively reduced to 30% of Fc 1.0, and water stress conditions were assessed when the average reading of the stress-exposed pots reached the required soil water content.

Supplementary figures

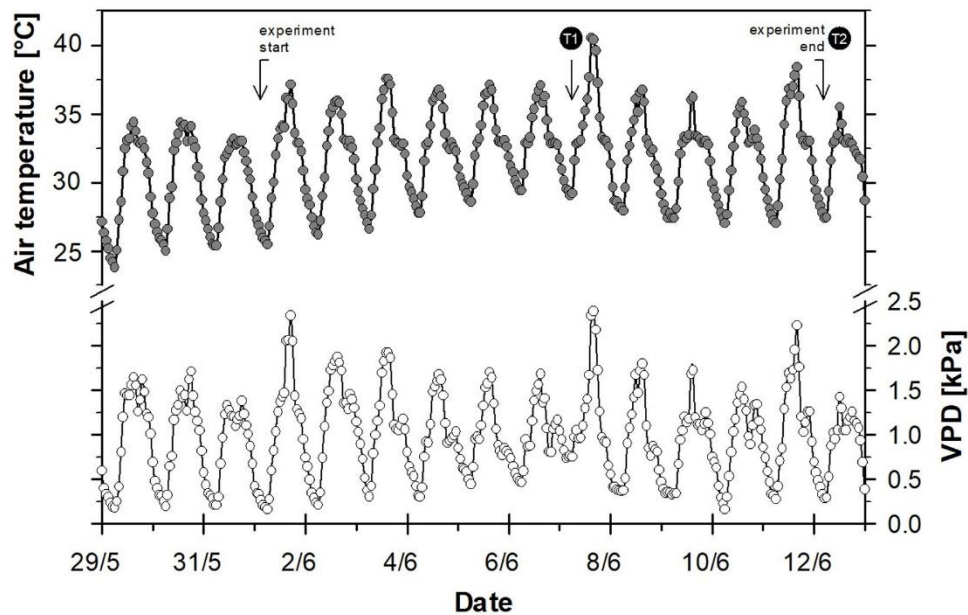


Figure 1S. Trend and daily range variation of air temperature (grey dotted line) and relative humidity (white dotted line) measured at the weather station within the experimental greenhouse at the University of Milan (Italy) during the water stress trial period. Air temperature and vapor pressure deficit (VPD) were measured within the greenhouse using a humidity and temperature probe (model HMP-45A, Vaisala, Vantaa, Finland) housed inside a solar radiation shield. A daily average of 26.8 ± 0.3 °C and 24.8 ± 0.4 °C was measured in the light and dark periods, respectively, while the mean daily VPD was 0.96 ± 0.1 kPa. T1 and T2 indicate the sampling dates, in which soil of stressed plants was at 50% and 30% of field capacity, respectively.

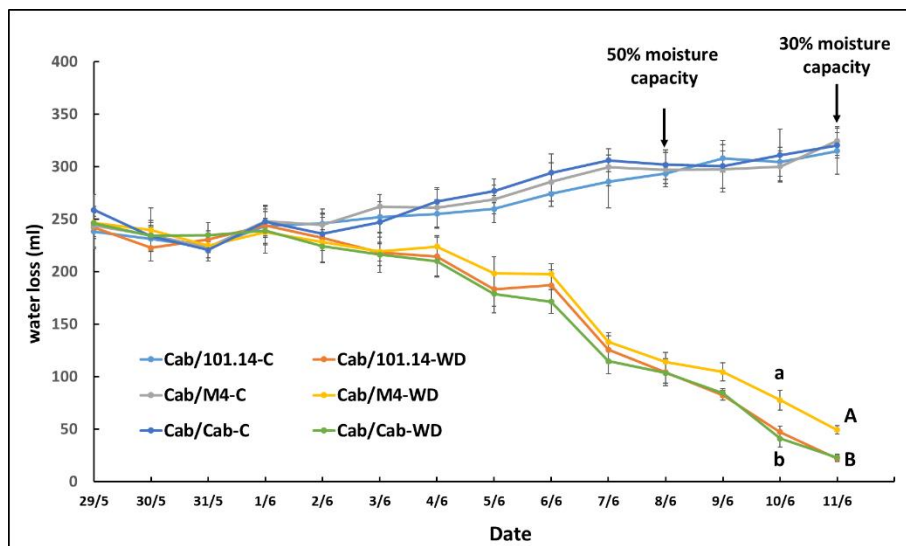


Figure 2S. Trend of daily water loss by plants. Water loss was measured weighing the plants before and after water addition. Measurements were carried out twice a day, at 8:00 AM and at 6:00 PM, and the amount of water needed to maintain the desired soil field capacity was then added. The values are means $\pm$ SE ($n=8$). Statistical significance was assessed by one-way ANOVA and Holm-Sidak method within each time and among WD conditions. Letters indicate significant differences ($p \leq 0.05$).