

# Microtensiometers for detecting and shaping the response of grapevine to increasing water stress levels

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## Abstract

Plant-based sensors able to continuously measure the stem water potential could be useful for the early detection of water stress, having the advantage of overcoming the labour-intensive and destructive procedures needed to assess water potential with the traditional pressure chamber. The experiment aimed to test the efficacy of microtensiometers to characterize grapevines' water status response to the increasing limitation of soil water availability. To this purpose, four Florapulse microtensiometers were applied to 4 potted grapevines ('Pinot gris' on SO4) growing under controlled conditions mimicking typical summer days. Plants were constantly weighted to assess transpiration gravimetrically. Two consecutive cycles of water stress were imposed by withholding irrigation for 10 days. During this period, midday stem and pre-dawn water potential were periodically assessed using the traditional pressure chamber. The vines developed a different canopy surface, with leaf area ranging from 1.19 (pot 4) and 1.55 (pot 1 and 2) m<sup>2</sup> vine<sup>-1</sup>, and thus responded differently to the absence of irrigation. The first stress cycle caused the loss of 42, 25, and 49% of the leaves in pots 1, 2, and 3, respectively, while no visible symptoms were observed in pot 4. The two microtensiometers that provided a complete data set during the whole experiment clearly detected the diurnal course and the daily trend of plant water potential. The maximum and minimum daily values extracted from the continuous time series were used as a proxy of pre-dawn and midday stem water potential, respectively. They were consistently 0.2 MPa higher (on average) than stem water potential values obtained using the pressure chamber. Microtensiometer data were well correlated with daily transpiration. The same values, binned in classes of 0.1 MPa were used to characterize the water use strategy through the slope metric, evidencing a strict anisohydric behaviour of 'Pinot gris' even under severe water limiting conditions (<-1.0 MPa of midday stem water potential).

**Keywords:** Florapulse, water deficit, 'Pinot gris', transpiration, slope metric, stem water potential

## INTRODUCTION

Grapevine (*Vitis vinifera*) plays a central role in the agricultural economy and cultural heritage of many regions around the world (International Organization of Vine and Wine, 2019). Among the several risks associated with climate change, the changes in precipitation patterns can result in an increasing risk of water scarcity, with detrimental effects on grape yield and quality. The successful implementation of accurate indicators of vine water status, will help to improve irrigation water management strategies and maintain grapevine productivity under harsher condition (Benyahia et al., 2023; EIP-AGRI, 2016).

Water potential ( $\Psi$ ) gradients within the soil-plant-atmosphere continuum (SPAC) serve as the driving force for the absorption of soil water by the root system, the conveyance of water through the plant xylem to the leaves, and the release of water into the atmosphere via stomatal pores. By regulating its stomatal and hydraulic conductance, plants maintain their  $\Psi$  within a suitable range. Stem water potential ( $\Psi_{\text{STEM}}$ ) assessed at midday by the pressure chambers is less influenced by high temperatures and vapour pressure deficit (VPD) than  $\Psi_{\text{LEAF}}$ , allowing for detection of smaller differences among treatments. However, both



$\Psi_{\text{STEM}}$  and  $\Psi_{\text{LEAF}}$  are discontinuous and destructive measurements, and the ability to track the plant response to changing water availability rely on the frequency of sampling as well as on the number of measured leaves.

In the last decades, new plant-based methods have emerged as valuable tools to continuously monitor water stress and possibly schedule irrigation in fruit tree crops (Fernández, 2014). Recently, micro-tensiometers providing continuous measurements of  $\Psi_{\text{STEM}}$  were developed, paving the way for a new type of information and overcoming the labour-intensive procedures needed with the traditional pressure chamber (Gonzalez Nieto et al., 2023; Lakso et al., 2022). Besides their possible use for a direct and real-time assessment of tree-water status, they can help to improve the current understanding about the water use strategy of the different crops and cultivars under different levels of water stress. The possibility of extracting the daily maximum and minimum values from continuous time series of  $\Psi_{\text{STEM}}$  will help discriminate between iso- and anisohydric behaviour and their intermediate classes (Hochberg et al., 2018; Serrano et al., 2024), providing more data to characterize crop behaviour through recently developed metrics such as the so-called *slope* method (Martínez-Vilalta et al., 2014) or the *hydroscape* (Álvarez-Maldini et al., 2021; Meinzer et al., 2016).

The main goal of this work was to integrate continuously collected data from innovative micro-tensiometers to characterize grapevine transpiration dynamics based on plant water status. Additionally, we aimed to assess the water stress response strategy of cultivar 'Pinot gris' grafted on SO4 rootstock using the slope metric approach.

## MATERIAL AND METHODS

### Plant material and experimental setup

The trial was conducted in summer 2023 inside one growth chamber of the experimental greenhouse facility of UNIBZ on four adult vine trees ('Pinot gris' grafted on SO4 rootstock). The grapevines (15 years old) were previously explanted from the field condition and transplanted in February 2023 to 52-L pots filled with the same field soil (15% sand, 74% silt and 11% clay). Vines had a stem circumference at 50 cm height ranging from 11 to 13.5 cm. During the experiment, a plastic membrane covered the soil in the pots to minimize water evaporation, enabling a gravimetric estimate of vine transpiration. The experiment was carried out between July 25 and August 17, and consisted in two drought cycles, each lasting 10 days, which began the day after an irrigation event (referred to as DAI, day after irrigation, 0). At the end of each cycle, plants were subjected to a recovery irrigation that brought soil moisture back to field capacity. Optimal growing conditions were maintained within the growing chamber of the greenhouse in terms of photosynthetic active radiation, temperature and relative humidity (see details in the result section).

### Measurements of vine transpiration and water potential

Continuous measurement of pot weight was performed using load cells (Laumas Elettronica), with transpiration ( $T_{\text{GRAV}}$ ,  $L \text{ tree}^{-1} \text{ day}^{-1}$ ) obtained by subtracting the pot weights of two consecutive days and expressed as % of T at DAI 1 (Benyahia et al., 2023; Wenter et al., 2022). The soil water potential was measured at 30-min intervals with the capacitive moisture sensors Teros 21 (Meter group) installed at 15 cm depth in each pot. Plant water potential was measured using two different methods: a) microtensiometer sensors (FloraPulse) were installed on the stem of each of the four vines at approximately 45 cm above the grafting point, and connected to a CR1000 data logger (Campbell Scientific), programmed to record data every 5 min; b) midday stem water potential ( $\Psi_{\text{MIDDAY}}$ ) was measured at approximately 12:30 using a pump-up pressure chamber (PMS instrument company) on selected days: DAI 2, 7, during the first cycle, and DAI 3, 5, 7, and 10 during the second cycle. For each measuring time, three mature leaves were selected from each vine and placed in darkening plastic bags for approximately 90 min prior to measurement. Predawn stem water potential ( $\Psi_{\text{PD}}$ ) measurements were taken at the end of cycle 1 (DAI 10) and at the beginning of cycle 2 (DAI 3) measuring leaf water potential before 5:30 am

### The slope metric

We adopted the concepts proposed by Martínez-Vilalta et al. (2014), known as “slope metric” to describe the water use behaviour for the cultivar ‘Pinot gris’ at increasing water stress levels. The daily minimum and maximum values of stem water potential obtained from each FloraPulse were used as a proxy of midday and pre-dawn water potential, respectively, and binned at 0.10 MPa intervals. The slopes ( $\sigma$ ) of the linear fit considering the binned  $\Psi_{PD}$ - $\Psi_{MIDDAY}$  points of all intervals starting from highest values were calculated and then plotted against  $\Psi_{PD}$ . Finally, a moving average curve was used to illustrate the trend of the values. The objective of this metric is to represent the variation in behaviour trend along a continuum from anisohydry to isohydry as drought increases (Serrano et al., 2024), with extreme anisohydric behaviour associated to slope  $>1$  and partially isohydric behaviour when with slope between 0 and 1.

## RESULTS

### Environmental conditions

During the experiment, the temperature range was moderate, with maximum values slightly above 30°C and minimums slightly below 20°C, thus remaining within an optimal transpiration range for the plants. Vapor pressure deficit had comparable diurnal patterns with peaks ranging from 20 to 30 hPa (Figure 1). The photosynthetic active radiation was kept at optimal levels thanks to artificial lamps mimicking the diurnal pattern, with only minor differences among the daily cumulated values (daily PAR ranged from 17 to 19 mol m<sup>-2</sup> d<sup>-1</sup>).

The increasing drought stress and the subsequent recovery phases were visible by observing the soil water potential dynamic (Figure 1). Differences among the four vines occurred in the timing and intensity of the water stress, with soil water potential for vines 1, 2 and 3 decreasing faster than for the vine in pot 4. These differences were due to a different development of the vine canopy surface (1.56, 1.55, 1.33, 1.19 m<sup>2</sup> of leaf area in pots 1 to 4, respectively) and the consequent different transpiration rate. The severity of the stress caused the loss of 42, 25, and 49% of the leaves in vines of pot 1, 2, and 3 at the end of the first drought cycle with respect to the beginning, while any symptom on leaves was observed in pot 4.

### Stem and pre-dawn water potential

FloraPulse sensors values were detecting a proxy of  $\Psi_{PD}$  and  $\Psi_{MIDDAY}$  on daily basis in three and two out of four vines, for the first and second dry cycle respectively.  $\Psi_{PD}$  values ranged from approximately -0.01 to -1.00 MPa, while  $\Psi_{MIDDAY}$  were between -0.14 to -1.41 MPa (Figure 2). The same parameters measured at the same time with the traditional pressure chamber reported more negative data, with  $\Psi_{MIDDAY}$  reaching -1.67 MPa in the last day of stress (DAI 10). Unfortunately, technical problems affected the FloraPulse sensors installed in plant 1 and, partially, in plant 3. The signal provided was noisy, unable to measure either the expected diurnal pattern or the drop in  $\Psi_{STEM}$  associated with increasing water stress, and we did not consider them in further analysis. We attributed this malfunctioning to the presence of unhealthy wood in the trunk portion interested by sensor installation which was not allowing the maintenance of the tension state. Despite these issues, the two measured vines were representative of different degrees of water stress and allowed us to test the hypothesis of the study.

The diurnal pattern of the cumulated transpiration values assessed gravimetrically ( $T_{GRAV}$ ) and the daily values of  $\Psi_{PD}$  and  $\Psi_{MIDDAY}$  extracted from the microtensiometers installed in vines 2 and 4 during the two drought stress cycles are reported in Figure 3. These two plants were selected as they shown the most continuous and distinct behaviour with the vine in pot 2 exhibiting the greatest stress and vine in pot 4 the least. The joint data set was used to identify the best relationship between the  $T_{GRAV}$  and  $\Psi_{PD}$  or  $\Psi_{midday}$  (Figure 4). A significant exponential decay of  $T_{GRAV}$  at decreasing  $\Psi_{PD}$  values was found ( $R^2=0.78$ ), while  $\Psi_{MIDDAY}$  was linear related to  $T_{GRAV}$  ( $R^2=0.80$ ).

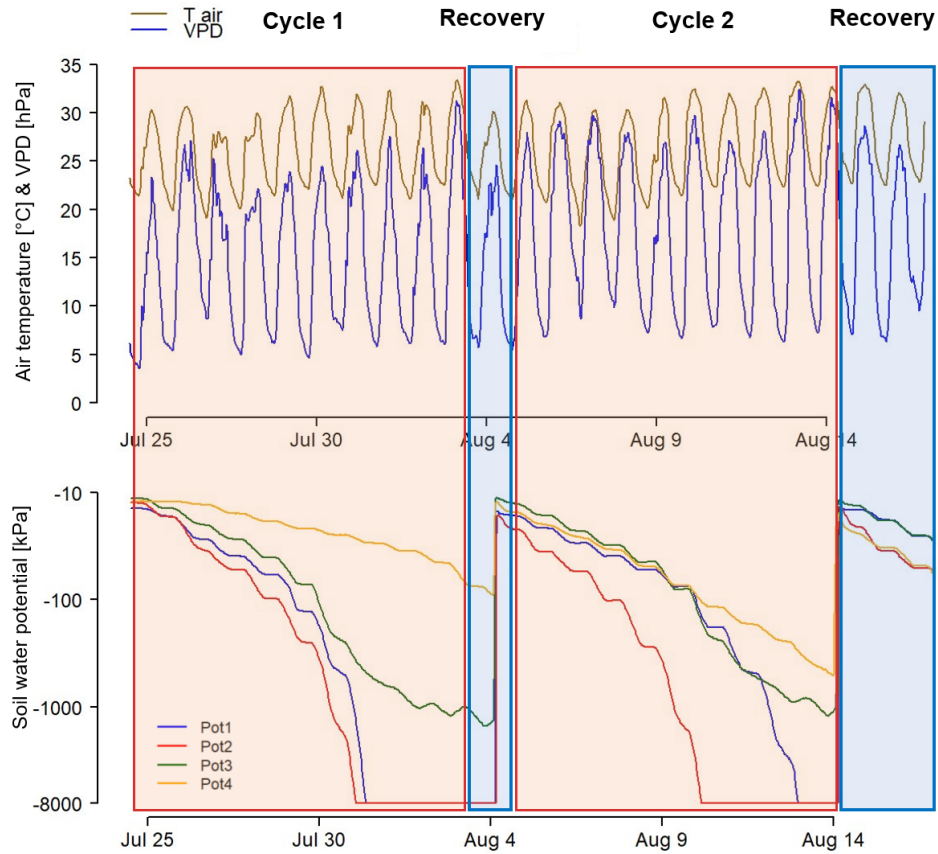


Figure 1. Continuous measurements of ambient air temperature ( $T_{air}$ , °C) and vapour pressure deficit (VPD, hPa) in the growth chamber (top) and trend of the soil water potential (SWP, kPa, logarithmic scale) during the two drying cycle of the experiment and relative recovery phase (bottom).

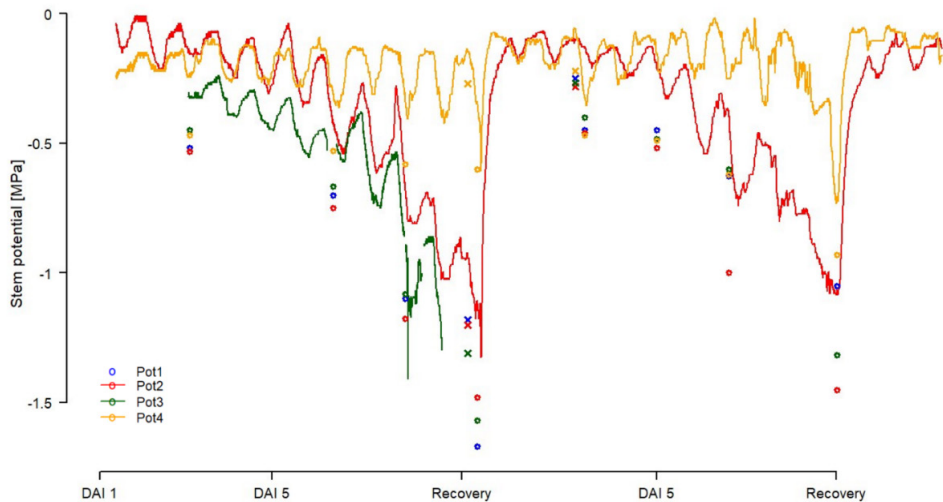


Figure 2. Evolution of stem water potential throughout two progressive drought cycles. The continuous measurements from Florapulse are represented with lines (data for pot 1 and the second cycle of pot 3 are not present due to instrument technical problems), “x” and “o” points represent the average  $\Psi_{PD}$  and  $\Psi_{MIDDAY}$  values, respectively, measured with the pressure chamber.

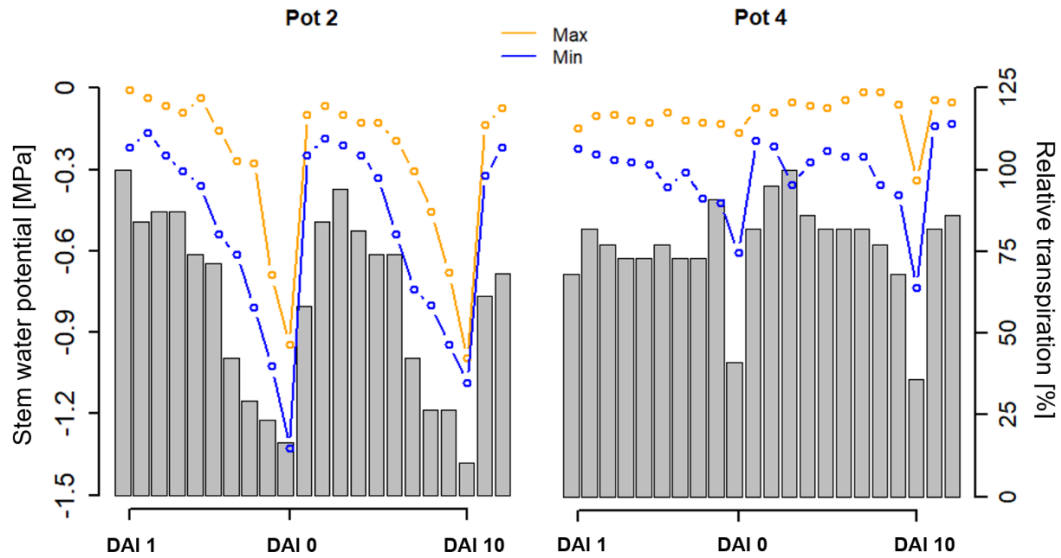


Figure 3. Trend of the relative daily transpiration ( $T_{\text{GRAV}}$ , %) and minimum and maximum stem water potential measured with microtensiometers during the two drought cycles in pots 2 and 4. Day after irrigation (DAI) 10 of the first cycle corresponds to DAI0 of the second cycle, since the recovery irrigation was carried out in the afternoon of the same day.

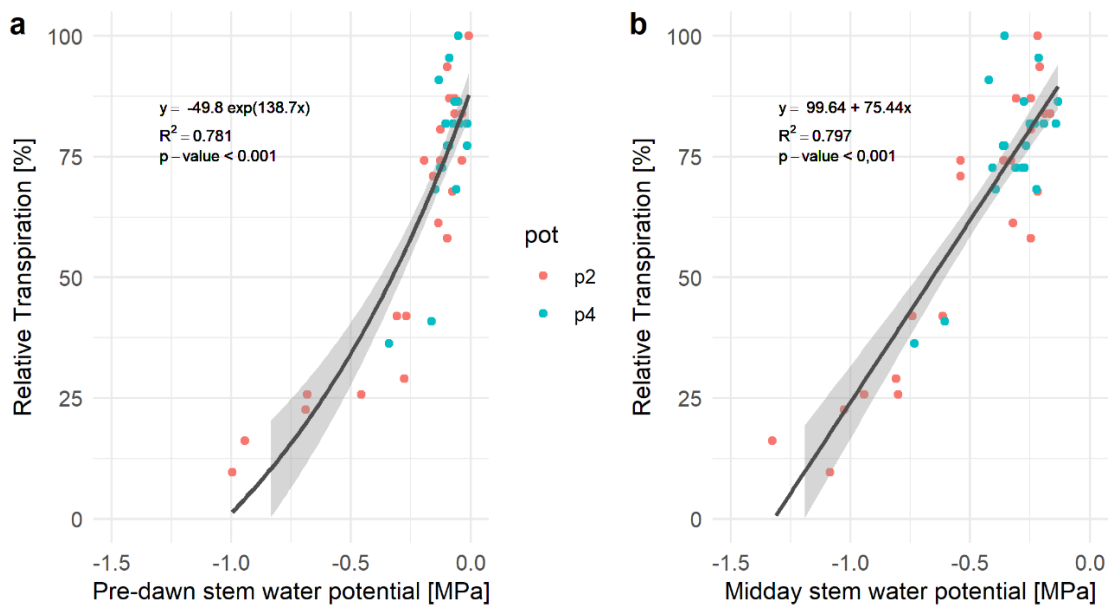


Figure 4. Correlation between the relative transpiration of the selected grapevines (pot 2 and pot 4) and the daily maximum (pre-dawn) and minimum (midday) stem water potential assessed via microtensiometers.

### The slope metric

Maximum and minimum daily values of the  $\Psi_{\text{STEM}}$  measured with the microtensiometers were assumed as proxy of  $\Psi_{\text{PD}}$  and  $\Psi_{\text{MIDDAY}}$ , respectively, binned at  $\Psi_{\text{PD}}$  intervals of 0.1 MPa (Figure 5A), were linearly correlated ( $\Psi_{\text{MIDDAY}} = 1.0364\Psi_{\text{PD}} - 0.2401$ ,  $R^2 = 0.985$ ,  $p\text{-value} < 0.001$ ). The change in the slope values of the  $\Psi_{\text{MIDDAY}}$  vs.  $\Psi_{\text{PD}}$  correlation at decreasing  $\Psi_{\text{PD}}$  values throughout soil drought is shown in Figure 5B. Following the interpretation proposed by

Serrano et al. (2024), the cultivar ‘Pinot gris’ grafted on SO4 exhibited strict anisohydric behaviour at any value of  $\Psi_{PD}$  up to severe stress ( $\sim -1$  MPa) thus showing a labile ability to control their stomata under water limiting conditions.

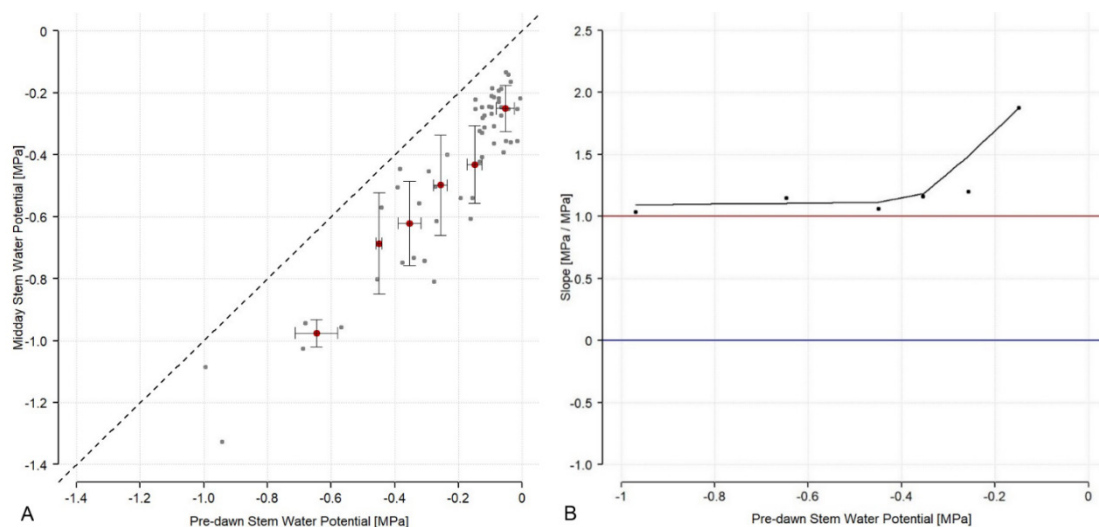


Figure 5. A) Correlation between pre-dawn and midday water potential of the row values (grey dots) and binned values at 0.10 MPa intervals (average  $\pm$  st.dev., red dots); B) change of slope  $\sigma$  values over the operating range of  $\Psi_{PD}$  ( $\Psi_{PD}$  class = 0.10 MPa), the figure shows the limits of a strict anisohydric and a strict isohydric behaviour, represented by the red and blue lines, respectively.

## DISCUSSION AND CONCLUSIONS

All the soil- and plant-based sensors used in the trial consistently showed increasing drought conditions in the period after the irrigation.  $\Psi_{SOIL}$  was the first parameters to be sharply reduced as a consequence of the decrease soil water availability in the pots (Figure 1) which, in turn, affected  $\Psi_{STEM}$  (Figure 2). Despite the trend being similar in all pots, soil water potential in pots 1 and 2 showed a faster decrease with respect to pot 4 due to the larger canopy surface area developed by the respective grapevine, causing higher transpiration rates. A correlation analysis with the soil water potential could not be performed because the latter reached the minimum value measured by the sensors after a short time. The use of soil-based sensors may be limited by their small volume of influence which are not offering a realistic representation of the water present along a gradient (Suter et al., 2019). This is possibly explaining the reason why the vines continued to transpire despite the low levels of soil water potential (Figure 3).

In both stress cycles, the cumulative  $T_{GRAV}$  reached an average value of approximately 10.5 L pot<sup>-1</sup> over 10 days. Under full watered conditions, the pots reached a maximum transpiration value of 1.3 L day<sup>-1</sup> on average. As soil water availability decreased, transpiration was adversely affected, decreasing to 0.5 L day<sup>-1</sup> at the end of the cycles (Figure 3). This decline was observed on average between DAI 7 and DAI 8, particularly in the first cycle. The highest stress was reached at the end of the first cycle, when  $\Psi_{STEM}$  was -1.67 MPa (Figure 2) and the lowest transpiration values were recorded. At the end of the second cycle,  $\Psi_{STEM}$  did not reach values lower than -1.5 MPa. This pattern can be explained by the slightly reduced transpiration rate measured in the second cycle after the partial leaf drop caused by the first stress cycle. The ‘Pinot gris’ vines maintained high transpiration rates until  $\Psi_{STEM}$  reached approximately -0.4 MPa (Figure 3).

FloraPulse micro-tensiometers responded well to changes in environmental conditions, and the data obtained were consistent with Scholander pressure chamber’s measurements (Figure 2), despite reporting values approximately 0.2 MPa higher. It is likely that the differences were due to the different height of measurement in the plant, with the more

negative values recorded at the leaf level being theoretically consistent with the water potential gradient occurring in the different plant compartments (roots, trunk, shoots and leaves). Our values are in line with those of Lakso et al. (2022), who already compared microtensiometers and pressure chambers in grapevine. Although issues about the duration of the sensors, the possibility of reinstalling them multiple times, and difficulties to carry out a proper installation in small sized and twisted trunks such as grapevines still persist, properly installed microtensiometers provided season-long diurnal data, opening new opportunities to use such detailed knowledge for early water stress detection and for modelling the continuous response of trees to decreasing levels of water availability. These results can be achieved by the application of several metrics such as the slope metric, the hydroscape and conductive surface (Serrano et al., 2024). Here we reported a first attempt of using microtensiometers to shape the response of 'Pinot gris' × SO4 to increasing water deficit according to the slope metric: being the  $\sigma$  always >1 we concluded that this combination of cultivar × rootstock exhibits a strict anisohydric behaviour, with no clear indication of changes in the behaviour at increasing level of water stress. Our results agree with those proposed by Faralli et al. (2021) for 'Pinot gris' and other cultivars such as 'Moscatel' and 'Riesling' (Serrano et al., 2024). Drawing from our experience, we believe that future studies would benefit from standardizing the installation height to obtain comparable absolute values of  $\Psi_{STEM}$  and increasing the number of sensors to better capture the potentially higher variability under field conditions.

## ACKNOWLEDGEMENTS

This study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

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