

Intra-vineyard dynamic observations of grapevine root growth and canopy performance under extreme microclimate conditions

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Abstract: Climate change affects soil characteristics, with soil temperature being notably impacted. While the effects of elevated or extreme temperatures on the grapevine canopy are well documented, there is less understanding of their direct influence on fine-root turnover and their indirect effects on root growth mediated by the soil microbiome. This study aimed to evaluate grapevine fine-root turnover, root distribution in the rhizosphere, and associated leaf ecophysiological responses. The investigation was conducted in two nearby vineyards in Central Italy on the cultivar Aleatico (*Vitis vinifera* L.), comparing two different root systems: the rootstock 420A (*V. berlandieri* × *riparia*) for grafted vines, and own-rooted (ungrafted) vines. Fine-root growth was monitored using prototype minirhizotrons (MRs) installed at a 35° angle from vertical into the rooting zone in the inter-row area (20 cm from the trunk, down to 60 cm depth). The MRs were equipped with RGB cameras to capture sequential images of the root zone. These images were analyzed with ImageJ® software to quantify cumulative fine-root production over the growing season. Canopy functionality was assessed by measuring leaf chlorophyll content, stomatal conductance, and photochemical efficiency using the JIP-test. The results revealed distinct fine-root dynamics between the two root systems. Grafted vines exhibited greater sensitivity to elevated soil temperatures, as indicated by reduced root turnover and decreased canopy performance (lower photochemical efficiency and earlier leaf senescence). The use of MRs proved to be an effective tool for advancing knowledge of grapevine root–shoot interactions, which are essential for developing adaptation strategies to climate change.

Keywords: adaptation strategies, climate emergency, mycorrhizas, non-destructive testing, fine-root turnover, grapevine rootstocks.

1. Introduction

Climate change is a pressing global concern that has far-reaching implications for various agricultural productive systems, including viticulture (Altieri and Nicholls, 2017). The detrimental effects of climate change on *Vitis vinifera* L. have been widely studied (Biasi et al., 2019; Hatfield et al., 2011; Hannah et al., 2013; Olesen and Bindi, 2002; Van Leeuwen et al., 2019). In addition to warmer temperatures and radiative stresses, extreme drought events are expected to occur more often due to alteration of rainfall regimes (IPCC, 2014). These severe abiotic events affect the resilience of the terroirs, by altering the environmental suitability for grapevines cultivation, crop yield and quality, resulting in a decline in viticultural productivity (Olesen et al., 2011).

The occurrence of anomalous temperatures represents one of the main climate risks, as proved by

the increased frequency of late and early frost (Cirrincione et al., 2024). At the same time, anomalous high temperatures are increasing in frequency (particularly during ripening) and this is impacting negatively grape composition at harvest.

Vitis vinifera L. may adapt to environmental changes by exhibiting shifts in the phenology (Biasi et al., 2019; Van Leeuwen et al., 2019), a phenomenon known as phenotypic plasticity (Nicotra et al., 2011). Gaining insight into these plastic responses is essential for predicting and managing climate change's impacts on native species and crops.

One aspect that requires further investigation is the impact of the climate alterations on soil temperature and consequently on root growth (Costa et al., 2023; De Rességuier et al., 2023), as it can lead to profound consequences for crop yields, carbon allocation (Brunori et al., 2016), nutrient availability, soil microbiome activity and diversity, and overall ecosystem functioning, although grapevine generally well-adapts to arid and semi-arid climates (Luchaire, 2025). Drought stress combined with high temperatures occurring during flower differentiation could result in pollen sterility or ovary abortion, and in decreases in water consumption by plants, photosynthesis, and crop biomass (Biasi et al., 2021; Brunori et al., 2022). Drought can also lead to salinity stress that, in turn, reduces soil water potential, root growth, and altered assimilate partition (Mahmud et al., 2018). In summary, warmer soils cause alteration in above and below-ground biomass functionality triggering numerous changes in the whole plant to adapt to this climatic variance. To sustainably adapt viticultural production to drought, the planting of rootstock genotypes adapted to a changing climate is a promising means, yet there is a lack of knowledge on space-time root system development and its interaction with the environment and the management practices. A better understanding of the soil-plant-atmosphere relations under climate change implies the study of the root system growth and development. Many approaches, methods, and instruments have been adopted for studying and modelling root growth, ranging from advanced imaging techniques to field studies. Each approach offers unique insights into the complexities of root growth and development and their interaction with environmental factors. Early methods adopted were disruptive and included techniques like excavating the entire root system or a portion of it; the monolith method involves removing a block of soil that contains the roots (Taylor, 1986). Core methods require extracting cylindrical core samples from the soil profile and rinsing the soil off the roots (Brunori et al., 2016). Alternatively, the trench profile method necessitates digging a wide wall in the soil profile, cutting off the roots that extend from this wall, washing and scraping the soil wall, and then placing a square grid on the clean soil surface to facilitate the identification and counting of roots (Böhm, 1979).

The first method of studying root distributions in viticulture has been the profile wall method (Böhm, 1979) which helps to identify that almost 95% of fine roots are located in the upper 60 cm of soil in a one-meter square area within the berm of the vine row. Already, Bates (1937) proposed that roots could be observed through the walls of clear cylinders inserted into the soil profile. Since then, non-destructive methods have been adopted with several modifications focused on grapevine (Freeman and Smart, 1976), trees, and tea plants (Fordham, 1972). Recent advancements in digital image sequence processing have improved the measurement of spatial growth profiles with high temporal resolution, allowing the examination of the root's total elongation and the distribution of the elongation rate along its growth axis (Lecompte et al., 2003). Nowadays, minirhizotrons optimized image acquisition, adding multispectral root images (Rahman et al., 2020), enhanced grapevine root observation in the field (Linsenmeier et al., 2010), improving knowledge on grapevine root system development under different nutrition conditions (de Souza Kulmann et al., 2022), or application of plant growth-promoting rhizobacteria (Ma et al., 2024).

Understanding the spatial distribution of root growth is becoming increasingly important for studying long-term changes in overall growth and for examining how environmental stimuli influence both root growth and canopy physiological responses. Selecting rootstock genotypes that can adapt vines to drought conditions, emphasising the need for a better understanding of root system development and its interactions with environmental factors (Fichtl et al., 2023).

Vitis vinifera-based production systems are predominantly monospecific-crop systems, a condition that can amplify the effects of climatic changes. Adaptation strategies may imply increase biodiversity, complementary root growth promoting strategies, optimized nutrient use, therefore reducing abiotic stress especially in difficult soils, including those with nutrient deficiencies, aridity and waterlogging (Zhang et al., 2024).

This research aimed to firstly explore how different root genotypes, such as the rootstock 420A (*V. berlandieri* × *riparia*) and own-roots (*Vitis vinifera* L.), respond to thermal air and soil traits throughout the growing season, affecting the fine-root turnover and crown physiological behavior. The relationships among root growth, soil physical and biological fertility (AMF root colonization), responsible for the complex soil-plant-atmosphere interaction, are discussed. By better understanding these relationships, it is possible to effectively address the challenges posed by climate change and develop strategies to sustainably manage cropping systems in the face of evolving environmental conditions.

2. Materials and Methods

2.1. Plant materials and growth conditions

The investigation was carried out in a traditional grape-wine growing area (De Santis et al., 2017) located on the hillsides of the Bolsena lake basin (Latium region, central Italy), where grapevine cultivation is based mainly on local landraces, highly adapted to the native environment. The climate is Mediterranean with a mean annual rainfall of 806 ± 230 mm (De Santis et al., 2017), mainly concentrated in autumn (November to December). The drought events are concentrated in the summer (June to August), during which the mean distribution of rainfall does not exceed two to four days per month. The study was carried out in 2021 and 2022 on two nearby vineyards within the Protected Denomination of Origin (PDO) Colli Etruschi Viterbesi and of the autochthonous cv ‘Aleatico N.’ (hereafter AL) grafted (rootstock 420A *V. berlandieri* × *riparia*; hereafter AL-420A), and own-rooted (hereafter AL-ORV) (Figure 1) (AL-ORV $42^{\circ}38'42.52''$ N - $11^{\circ}51'51.43''$ E, altitude of 323 m a.s.l; AL-420A $42^{\circ}38'41.75''$ N - $11^{\circ}51'58.45''$ E, same altitude). At the experimental site, the volcanic soils, i.e., ignimbrite, characterized by heterogeneous textures (ashes and lapilli) and poorly consistent, allow grapevine to be cultivated without the use of the rootstock (ungrafted vines) (Biasi et al., 2023; Catalani et al., 2024). Vines’ training system was the “alberello”, with a spacing of 1×1.5 m, corresponding to a planting density of 10.000 vine ha^{-1} . The tested vineyards were managed according to organic farming principles (Regulation (EU) 2018/848) using regenerative soil management techniques (Canali et al., 2015). Roller crimpers was employed to mechanically roll over cover crops or weeds, crimping the stems and flattening them against the ground. Both the grafted and ungrafted vineyards proved to be colonized by the AMFs, although belonging to different species (Biasi et al., 2023; Catalani et al., 2024).

2.2. Prototypal minirhizotrons characteristics, data acquisition and processing

Fine-root growth was observed through six prototypal minirhizotrons (MRs) installed on March 2022 and placed about at 35° off vertically into the rooting zone, at a distance of 0.20 m from the vine trunk, down to a depth of 0.6 m. MRs were made of plexiglass, and had a parallelepiped shape and a rectangular base (0.20×0.10 m) (Rajurkar et al. 2022).

The developed MRs integrate advanced vision systems to enhance root growth monitoring. The developed prototype employs a transparent polycarbonate parallelepiped structure designed for underground deployment at a maximum depth of 60 cm. This structure protects the electronic components while maintaining visibility for imaging purposes.

The imaging system is based on four fixed Arducam NOIR 8 MP Sony IMX219 cameras, optimally positioned to cover the entire observation range without the need for mechanical movement. These cameras were selected for their high resolution (8 megapixels) and capability to operate efficiently in low-light conditions. The camera’s resolution allows for a spatial resolution of approximately 0.2 mm/pixel,

ensuring the detection of fine root structures with remarkable precision. To address illumination challenges in subterranean environments, low-consumption red LED lights were strategically positioned to reduce glare and shadows, improving image clarity.

The decision to adopt four fixed cameras instead of a single mobile camera system introduces several significant advantages that enhance the system's overall efficiency and reliability. One of the primary benefits lies in the improved durability and stability of the system. By eliminating the need for motors and moving components, the risk of mechanical failure, misalignment, or wear overtime is substantially reduced. This design choice ensures stable long-term operation, particularly in demanding field conditions. The adoption of fixed cameras also simplifies the system's overall design. Without the need for motors, linear guides, and position control mechanisms, the system becomes less complex, resulting in improved robustness and reduced maintenance requirements. In addition to these benefits, the fixed camera setup improves energy efficiency. Since no additional power is required for mechanical movement, the system consumes less energy, a critical advantage for off-grid installations powered by solar panels. This reduction in power demand extends the system's operational autonomy and ensures continuous data acquisition even in remote or resource-limited environments. Lastly, the use of fixed cameras reduces the need for frequent recalibration. As each camera remains stationary, the risk of misalignment or positional errors is minimized, ensuring consistent image quality and measurement accuracy throughout the monitoring period.

A Raspberry Pi board controls image acquisition and data transmission, enabling automated data collection with minimal human intervention. The system also includes an Arduino-based timing module that activates and deactivates the Raspberry Pi to optimize energy consumption.

A key innovation in this system is its off-grid functionality. The system is powered by rechargeable lithium batteries, which are charged via photovoltaic panels. This energy-independent setup ensures continuous data collection in remote or resource-limited environments.

The acquisition protocol has been defined to acquire one image every hour (Figure 2) on three representative vines per vineyard typology (grafted vs ungrafted). Images were collected from March to October 2022, and a total of n. 1092 images were manually processed by ImageJ® software to assess fine root growth rate. In particular, the best diurnal pictures for each couple of days have been used to set the scale in ImageJ® software and the best nocturnal pictures of the same days were used for root identification and root length measures. To determine if the roots were dead or alive, multiple images taken of the same position over time were analyzed. The analysis involved the assessment for any new rootlet of the length and the retainment of the white versus black/brown, color, that indicated they were dead. During this process, we manually recorded the lengths of living roots and calculated the elongation and branching of fine roots, along with the daily average growth rate of root development (Figure 2).

2.3. Air and soil temperature acquisition and analysis

A meteorological station (part of the regional network of meteorological stations - https://siar.arsial.it/?-page_id=99) located on site (UTM33N North: 4730688; UTM33N East: 24460042°38'55'') was used to measure weather variables (air temperature and humidity, rainfall, with hourly frequency). Collected data were used to decode the Bagnouls and Gaussen climate diagrams - to visualize the aridity period - and to calculate the main viticultural bioclimatic indices (Table 1) such as: the growing season temperature (GST) and extreme temperature events as hot days (HDs) and extreme hot days (EHDs), provided by the number of days with daily maximum air temperature (T_{MAX}) above 30 °C and 35 °C, respectively, and dry days (DDs), as number of days with rainfall below 1 mm.

During the 2022 season soil rhizosphere temperature was continuously monitored (every hour) at a distance of 0.20 m from the vine trunk base and a depth of 0.25-0.30 m using a combined Arduino– and Raspberry Pi–based data logging system (sensor for temperature model DS18B20). The same system (sensor for temperature model DS18B20) was used to assess the bunch (0.50-0.60 m above ground) and intra canopy (1.0 m above ground) temperature (Figure 2).

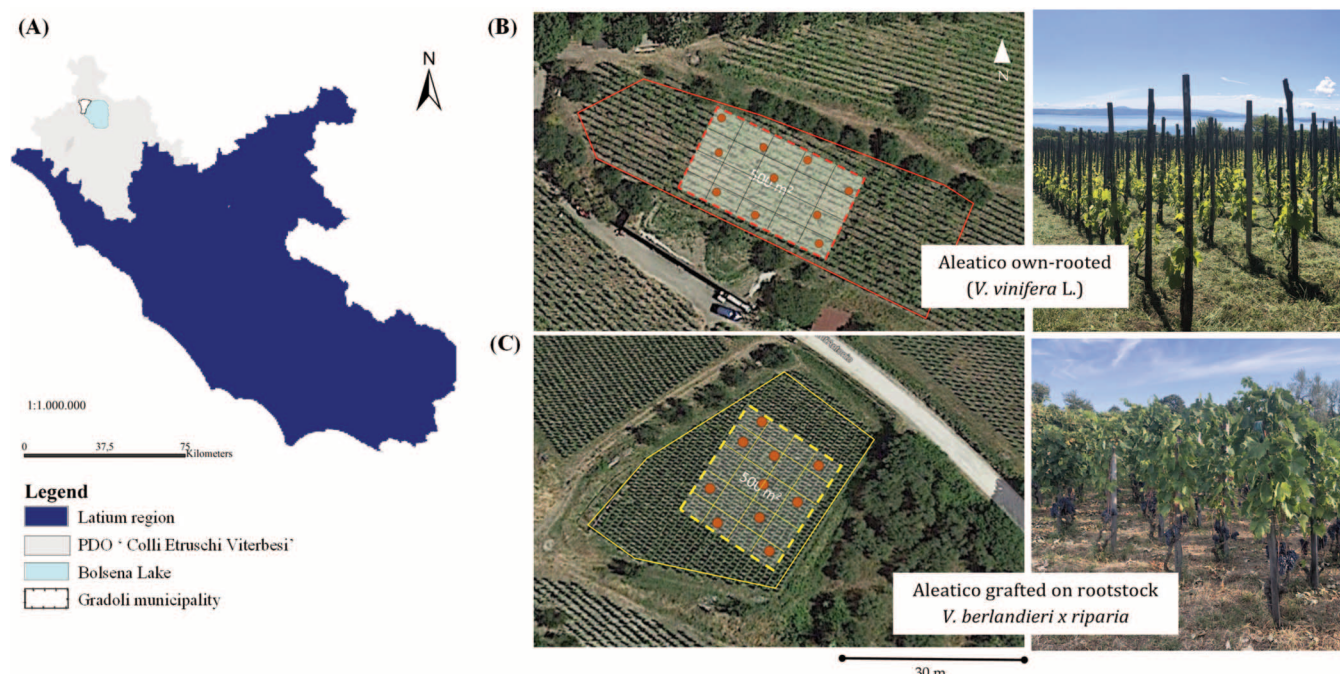


Figure 1. The grapevine growing area (A) Protected Denomination of Origin (PDO) Colli Etruschi Viterbesi where the two tested vineyards are placed: cv Aleatico grafted on rootstock (AL-420A) (B) and own-rooted (AL-ORV) (C). Distribution of prototype for soil and canopy temperature monitoring (T_{SOIL} – at 0.25-0.30 m of depth and a distance of 0.20 m from the vine); (T_{CANOPY} for the bunches and assimilating belt respectively at 0.50-0.60m and 1.0 above ground) (C - B), red circles.

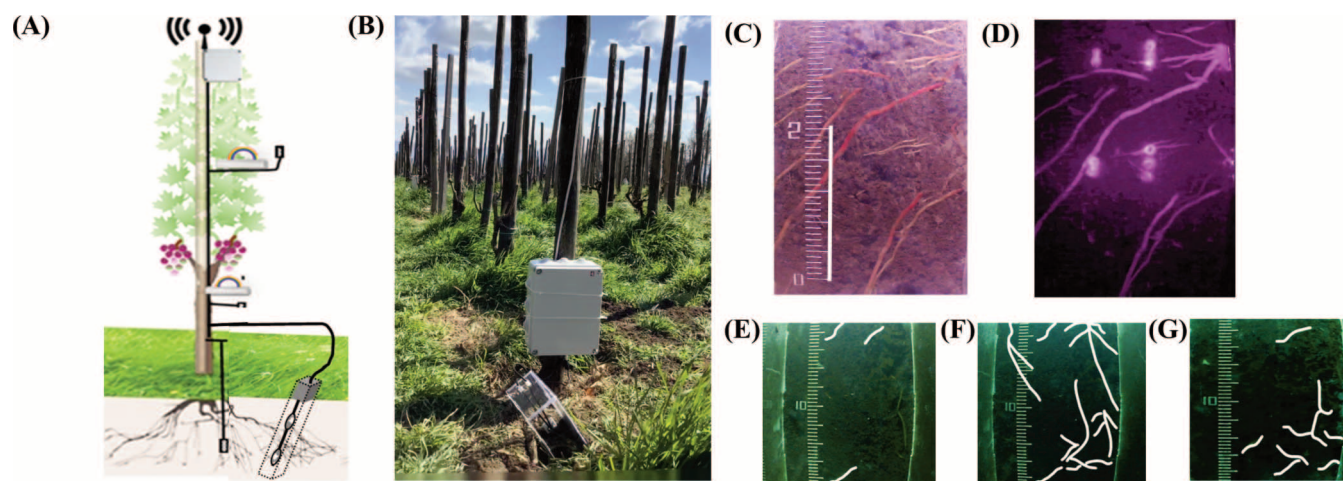


Figure 2. (A) Scheme of one of the six prototypal minirhizotrons (MRs) placed 35° off vertical at a distance of 0.20 m from the vine trunk, down to a depth of 0.6m (B). Prototype images: diurnal pictures (C) were used for the scale setting in the ImageJ® software and the nocturnal pictures (D) were used for the detection of white young rootlets. (E-G), image used for root length manual measurement.

2.4. Vine physiological measurements

Physiological assessments consisted in the measure of leaf chlorophyll content, stomatal conductance, and photochemical efficiency through the JIP-test. Data were collected during two consecutive seasons (2021 and 2022) at key phenological stages of grapevines according to the BBCH classifications (Olesen et al., 2011), specifically: full flowering (BBCH065), berries touching (BBCH079), and berry ripening (BBCH089). Physiological assessments were performed on ten vines from each vineyard,

using three fully expanded, undamaged sunlit leaves - the second leaf above the last cluster on each main shoot - between 10:30 a.m. and 12:30 p.m. (Brunori et al., 2022).

Table 1. Summary of bio-climatic indices calculated for seasons 2021-2022.

Bioclimatic indices	Abbreviation	Definitions	Unit	Reference
Growing Season Temperature	GST	T_{MEAN} (1 April - 31 October)	°C	Jones et al., 2005
Hot days /year	HDs	$T_{\text{MAX}} > 30^{\circ}\text{C}$	n	EEA, 2021
Extreme hot days /year	EHDs	$T_{\text{MAX}} > 35^{\circ}\text{C}$	n	
Dry days /year	DDs	$P_{\text{day}} < 1\text{mm}$	n	

Stress and stress adaptation of plant can be observed by monitoring the performance of the photosynthetic apparatus, especially PSII, because even though Chlorophyll a (CHL a) fluorescence represents only a small portion of the energy dissipated by the photosynthetic system, it has been proven to be a valuable non-invasive tool for investigating its structure and function (Strasser and Tsimilli-Michael, 2001). Photosystem II (PSII) activity and electron transport flux were determined by OJIP test performed on fully matured leaves (the same leaves used for CHL measurements) that were dark adapted for 20 min by wrapping aluminium foil and then exposed to saturating light pulse of $3000 \mu\text{mol m}^{-2} \text{s}^{-1}$ light intensity using hand-held device FluorPen FP-100 (Photon System International, Czech Republic). Various variable fluorescence (V_j , V_i) and other JIP-test parameters were registered following Strasser et al. (2001): the specific energy fluxes (per reaction centre) for absorption (ABS/RC), trapping (TR_0/RC), dissipation (DI_0/RC) and electron transport (ET_0/RC); the maximum quantum yield of primary photochemistry ($\phi_{\text{Po}} = \text{TR}_0/\text{ABS}$), the efficiency ($\Psi_0 = \text{ET}_0/\text{TR}_0$) with which a trapped exciton can move an electron into the electron transport chain, and the quantum yield of electron transport ($\phi_{\text{E0}} = \text{ET}_0/\text{ABS}$). ϕ_{ABS} was determined to assess the functionality of both PSI and II and produce quantitative information of the plant performance, especially under stress conditions (Bussotti et al., 2010; Bussotti et al., 2020).

The same leaves were used to quantify the chlorophyll leaf content (CHL) ($\mu\text{mol} \cdot \text{m}^{-2}$) as a leaf-senescence indicator using a hand-held MC- 100 Chlorophyll Concentration Meter (Apogee Instruments, Inc., Logan, UT, USA) and the stomatal conductance (gs) ($\text{mmol H}_2\text{O} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) using a leaf porometer (SC-1, Decagon Devices, Pullman, WA, USA) according to Brunori et al., 2022.

2.5. Statistical Analysis

The data obtained were statistically analysed using one-way ANOVA with the statistical computer package XLSTAT (Addinsoft, 2021). In addition, physiological response of vine in terms of stomatal conductance (gs) and leaf chlorophyll content, reported in Biasi et al. (2023) was used for statistical correlations (Pearson coefficient) performed with the same statistical packages.

3. Results

3.1. Abiotic stressors quantification

The two consecutive seasons 2021 and 2022 were characterized by dry periods occurring between end of May and August (Figure 3), in particular the dry period lasted until October for the 2021 season. The climate of the area is mainly characterised by periods of aridity (May–August) and periods of intense rainfall (October–March), data not shown.

The decrement in the amount of precipitation during the summer results in a warming of the ground surface, with soil temperature at 0.1 m of depth that exceeds 20°C , in particular this occurred during 2022 from June to September. In general, changes in soil temperatures followed changes in air temperatures (values in bold in Table2), and were also driven by low precipitation (e.g July and August 2021,

and June, July and August 2022 - Table 2). Based on these observations, both seasons were classified as “dry summers” according to DDs (210 and 187, respectively for 2021 and 2022), although the season 2021 was marked by severe-extreme drought (Figure 3).

Table 2. Abiotic stress conditions in the grape-wine growing area of the Bolsena Lake (Latium region, central Italy) for the seasons 2021 and 2022 (T_{MIN} - monthly average minimum temperature; T_{MEAN} - monthly average temperature; T_{MAX} - monthly average maximum temperature, RH - monthly average relative humidity; E_t0 - monthly average evapotranspiration rate; R - monthly cumulative rainfalls; T_{SOIL} - monthly average surface soil temperature 0.1 m depth. Values attributable to the onset of limiting abiotic conditions are shown in bold.

	T_{MIN} (°C)	T_{MEAN} (°C)	T_{MAX} (°C)	RH (%)	E_t0 (mm)	R (mm)	T_{SOIL} (°C)
2021							
Jan	-0.1	4.1	8.5	89	14.5	208.6	6.53
Feb	2.4	7.5	13.5	84	29.23	88	7.55
Mar	1.1	7.3	14.6	72	59.13	21	7.72
Apr	3.8	10	17	71	56.76	71.8	10.07
May	7.9	14.9	22.8	75	109.94	42.4	14.05
Jun	13.2	21.4	30.8	68	143.23	14.8	18.02
Jul	15.6	23.7	32.9	61	154.23	8.2	20.75
Aug	15.8	23.5	33.1	63	136.17	17.4	21.81
Sep	13	20.2	29.1	67	93.56	12.6	19.66
Oct	8.3	13	19.5	77	49.49	37.8	15.43
Nov	6.8	10.3	15	88	23.48	171.2	12.77
Dec	2.2	6.5	11.2	84	18.36	161	7.99
2022							
Jan	0.4	4.73	10.21	80.68	22.22	40.8	6.57
Feb	1.71	6.96	13.41	76.96	31.4	60.6	6.93
Mar	0.84	7.16	14.57	61.59	63.45	56.2	7.43
Apr	4.38	10.83	18.25	70.94	85.74	78.6	10.99
May	10.52	17.98	26.58	67.53	125.3	9	15.33
Jun	14.78	23.49	33.04	57.96	157.72	3.2	20.09
Jul	16.47	25.93	35.85	55.02	179.01	27.6	22.74
Aug	16.33	23.62	32.57	64.99	135.9	115.8	22.46
Sep	12.68	18.5	25.84	78	79.74	223.2	20.25
Oct	10.33	16.02	23.7	82.99	50.92	9.2	17.16
Nov	5.34	10.28	15.57	84.69	25.61	209.2	13.23
Dec	5.55	8.89	12.8	92.87	13.7	218.8	10.53

The total number of hot days (HDs) – together with an high number of DDs (50) – concurred to classify this area at “High-Risk Stress”. On the other hand, extremely hot days (EHDs) exceeded 18 days and 24 days, respectively for the 2021 and 2022 season. These values are consistent with climate projections under the RCP8.5 scenario, a high-emission pathway characterized by increasing radiative forcing and a higher frequency and severity of extreme temperature events, thereby representing the worst-case climate change scenario (Figure 3).

Intra-vineyard temperature, recorded by datalogger in the rhizosphere (0.20 m from the trunks of the vine, and 0.25–0.30 m of depth) and in the canopy (1.0 m from the ground) for the season 2022 in the two tested vineyards (AL-ORV and AL-420A) are shown in Figure 4.

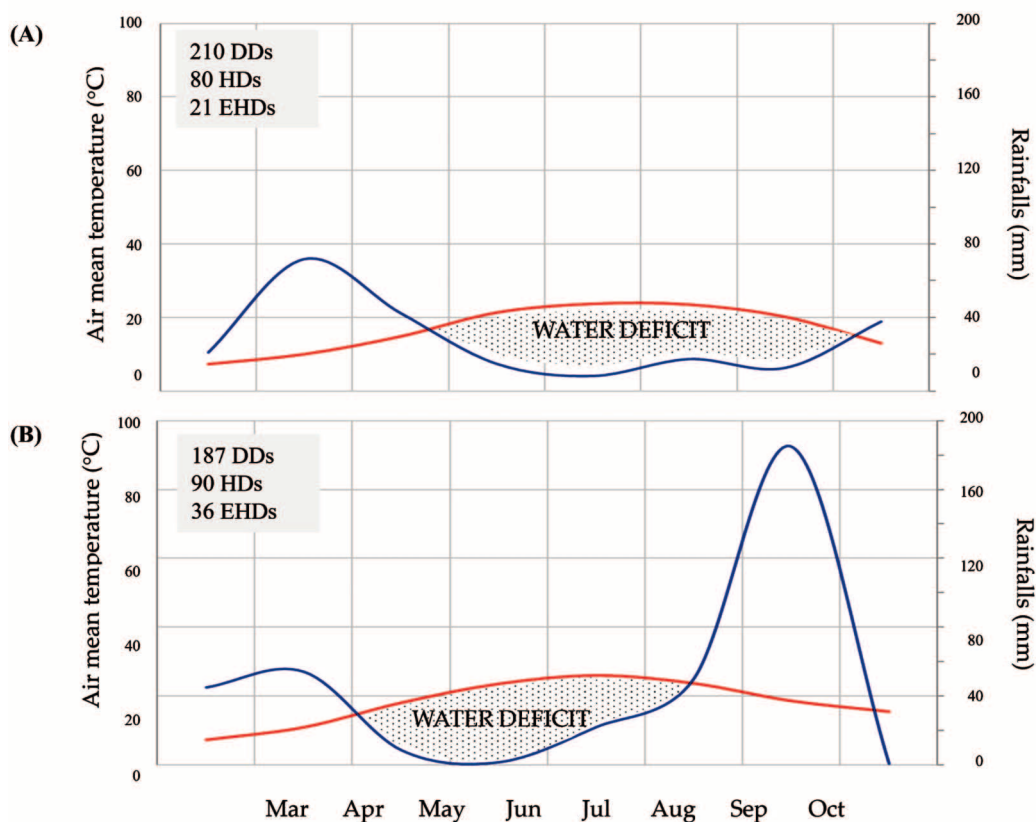


Figure 3. Climate diagrams of Bagnouls and Gausson of the growing season 2021 (A), and 2022 (B) representing monthly mean temperature (red line), monthly precipitation (blue line) and the water deficit. In the gray box the quantification of dry days (DDs), and the hot and extreme hot days (HDs, EHDs, respectively) are reported.

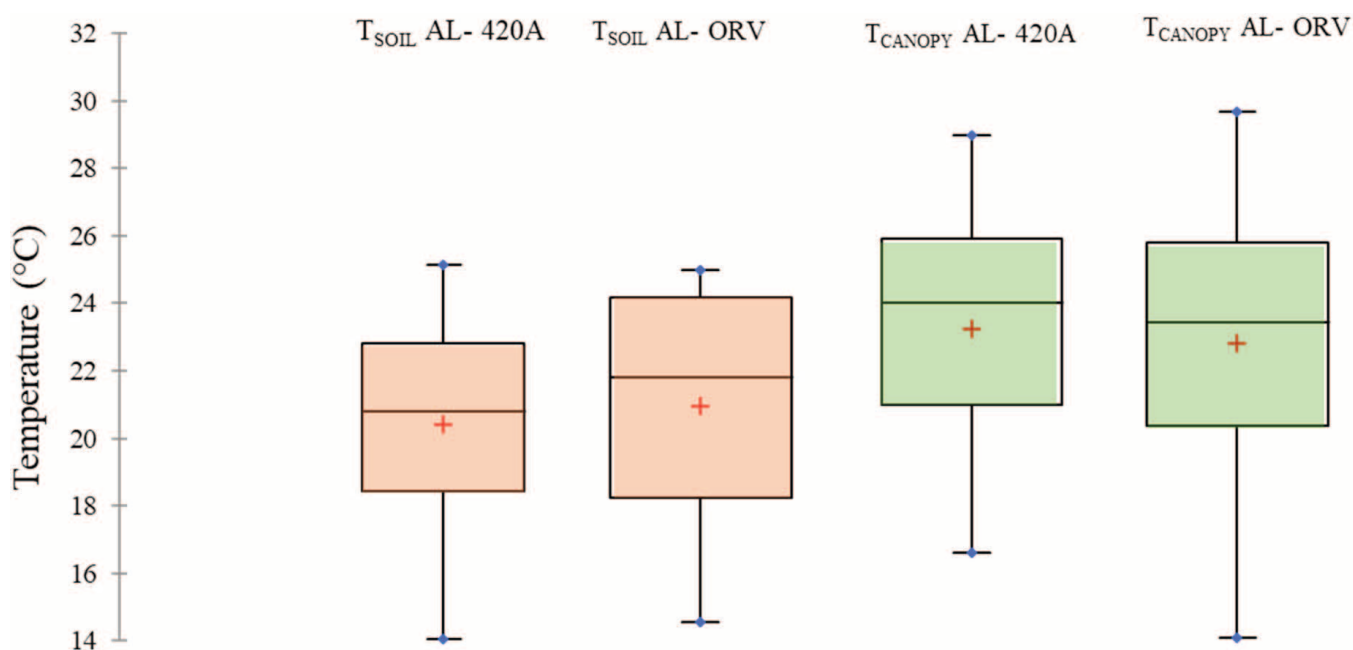


Figure 4. Intra-vineyard thermal availability in the two tested vineyards (AL-420A and AL-ORV) for the season 2022: soil temperature (T_{SOIL} – at 0.25-0.30 m of depth and a distance of 0.20 m from the vine trunk) and canopy temperatures (mean of T_{CANOPY} , respectively at 0.50-0.60 m and 1.0 above ground).

In both vineyards, during the warmer and drier season (2022), rhizosphere mean temperatures ranged between 14.1 – 25.2 °C, with the maximum soil temperature reaching 29.1°C at the BBCH 079 fruit set stage (June 2022). Intra-canopy temperature ranged between 15.2 – 30.3 °C, with the maximum temperature reaching 41.7 °C at BBCH 079 (June 2022). The mean temperature for soil and canopy during the season from April to October were 20.6 and 24.8 °C, respectively.

3.2. Fine root growth

Fine root growth was monitored during the 2022 season on grafted (AL-420A) and non-grafted vines (AL-ORV). Trends of the daily root growth rate for the two tested depths, 0-0.30 m and 0.30-0.60 m, are shown in Figures 5A and B. Data indicate that the rootstock root system had different rooting depth distributions and growth peaks, in respect to the cultivar root system (own-rooted vines). In the topsoil layer (0-0.30 cm depth) own-rooted vines (AL-ORV) started rooting in May, during the flowering time, presenting rooting peaks in late summer (August – September) and immediately after harvest time, in October. The most important root growth occurred during the early spring (Table 3) from May to June in the deeper soil layer (0.30 – 0.60 cm). On the other hand, fine-root growth in grafted vines (AL-420A) began several weeks later than in AL-ORV, shifting root growth in the period from June to August under more limiting climatic conditions (Figures 3 and 4; Table 3), when maximum air temperatures exceeded 30 °C, evapotranspiration was high (>120 mm), and soil temperatures reached their highest values (25–35 °C).

The most relevant root elongation was shown in autumn period and for the greater depth (Table 3).

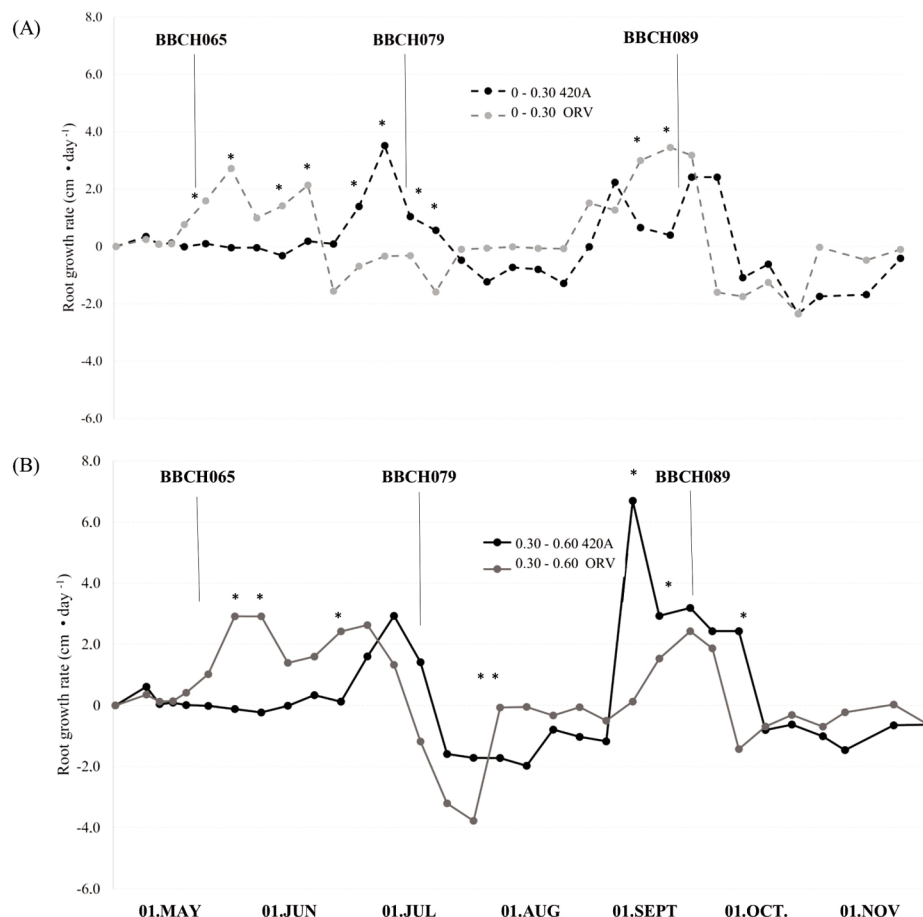


Figure 5. Root daily growth rate (cm day⁻¹) for grafted vine (*V. berlandieri* × *riparia*, AL-420A) and non-grafted vines (own-rooted vine, AL-ORV) at (A) 0-0.30 m, and (B) 0.30-0.60 m depth. Asterisk (*) showed statistical significance (p-value < 0.05) of the differences between treatments (AL-420A vs. AL-ORV).

Table 3. Relative root growth rate (cm day^{-1}) for the rootstock 420A and own roots (*Vitis vinifera* L.) in the two tested vineyards (AL-420A and AL-ORV, respectively) for the season 2022. Cumulative root growth is shown for the three main periods of vine growth stages and occurrence of abiotic stressors: May-June, July-August, September-October.

Vineyards	Period	Relative root growth rate (cm day^{-1})	
		Depth (0-30) cm	Depth (30-60) cm
AL-ORV	MAY-JUN	7.43	17.25
AL-ORV	JUL-AUG	-0.72	-9.16
AL-ORV	SEP-OCT	3.91	2.60
AL_420A	MAY-JUN	5.39	5.37
AL_420A	JUL-AUG	-2.95	-8.58
AL_420A	SEP-OCT	2.31	13.77

3.3. Vine physiological behaviour

The parameters of PSII light absorption, transmission and conversion are shown in Figure 6, and in Table S1. The JIP chlorophyll *a* fluorescence transient curves (log time scale) in mature leaves of two tested vineyard (AL-420A and AL-ORV) at ripening stage (BBCH 089) is shown in Figure 7. AL-420A showed for 2021 a lower value of ϕ_{E0} —the quantum yield of electron transport—, ET_0/RC —light quantum flux per active RC electron transfer—, and performance index (ϕ_{ABS}) for energy conservation from photons absorbed by PSII until the reduction of intersystem electron acceptors. ϕ_{P0} —the maximum quantum yield of primary photochemistry of a dark-adapted leaf—and TR_0/RC didn't show a wide variations between treatments. In the season 2022, photochemical indices such as ϕ_{P0} , Ψ_0 , and ϕ_{E0} exhibited a similar trend in the two tested vineyards during berry ripening, while ϕ_{ABS} showed significant variability between treatments.

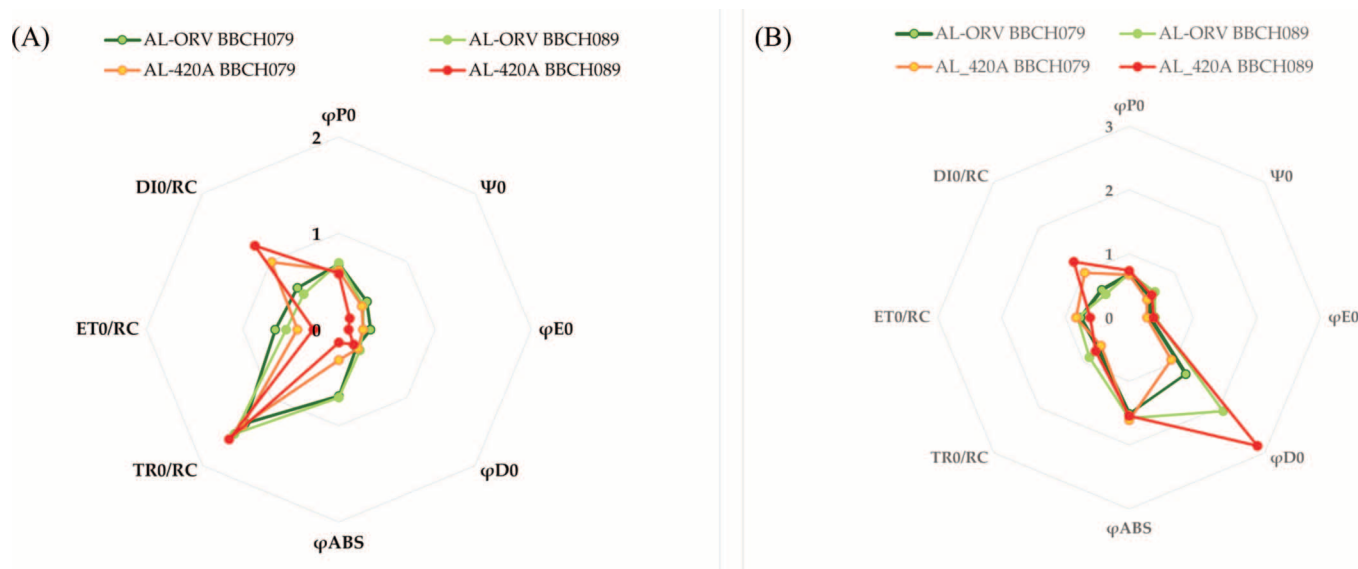


Figure 6. JIP-test parameters for the two seasons (A) 2021 and (B) 2022 for the mature leaves of the two tested vineyards AL-ORV and AL-420A during key phenological stages: BBCH079- majority of berries touching and BBCH089- berry ripening. [ϕ_{P0} – the maximum quantum yield of primary photochemistry of a dark-adapted leaf showed significant differences among leaves of two tested vineyards; Ψ_0 – the probability that a photon trapped by the PSII reaction centre enters the electron transport chain; ϕ_{E0} – the quantum yield of electron transport, and ϕ_{ABS} – the performance index for energy conservation from photons absorbed by PSII until the reduction of intersystem electron acceptors; TR_0/RC - trapping, DI_0/RC - dissipation and ET_0/RC - electron transport for reaction centre (RC)].

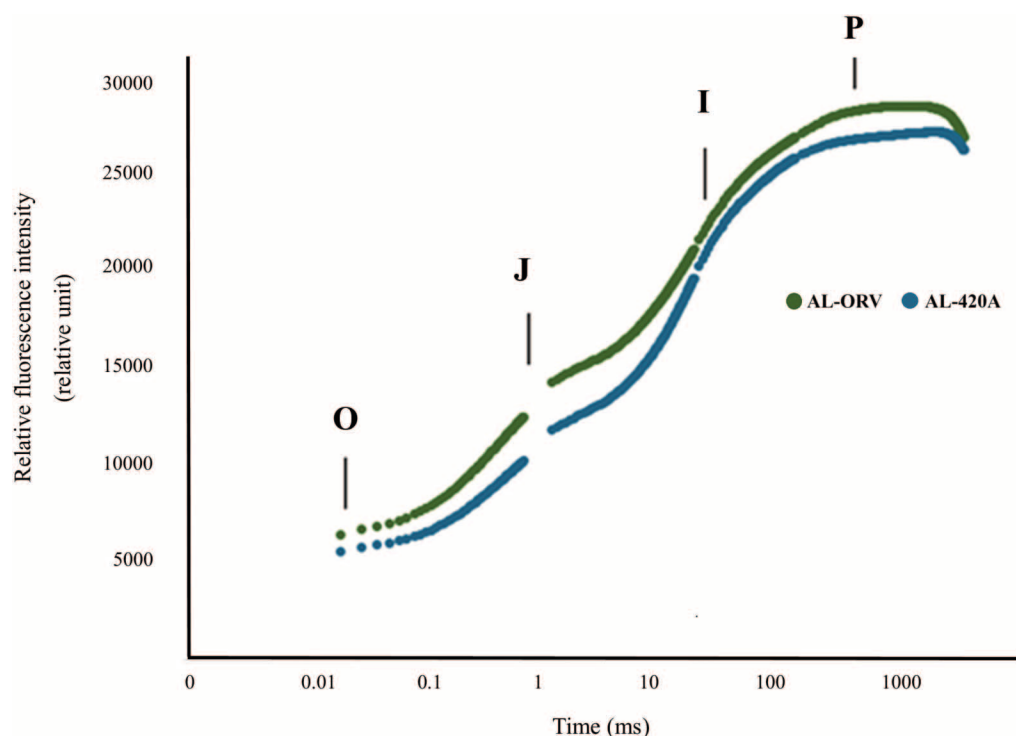


Figure 7. JIP transients curves for the mature leaves (mean values) of the two tested vineyards AL-420A and AL-ORV for the season 2022 at phenological stage BBCH 089: berry ripening.

Figure 7 displays the original JIP transients of the leaves (mean values) of the two tested vineyards (AL-420A and AL-ORV) at BBCH 089 for the season 2022, plotted on a logarithmic time scale. The OJIP curves exhibit the characteristic rise kinetics of chlorophyll *a* fluorescence. The fluorescence intensity rises from its minimum (F_0 , or O step) to its maximum (F_m , or P step), passing through two intermediate phases: J at around 2 ms and I at approximately 30 ms, in response to a saturating light flash. Notable differences in the difference in the JIP curves, in particular AL-ORV showed an increase in the OJ and JI Phases and after in the final part with Pstep greater than AL-420A.

Statistical correlation for the tested parameters showed a significant correlation between CHL and ϕ_{ABS} and ET_0/RC for the season 2021 and among g_s and JIP-test parameters for the season 2022 (Table S2).

For the 2022 growing season, correlation and PCA were run including all physiological parameters, root growth at different depths (ROOT 0-30, ROOT 30-60) and soil and canopy mean and maximum temperature (T_{MAX} SOIL, T_{MEAN} SOIL, T_{MEAN} CANOPY, T_{MAX} CANOPY). Correlation matrix (Pearson Coefficient - Table S2) showed a significant correlation among the root growth in the topsoil layer (0-0.30m) and photochemical parameters with value of Pearson's coefficient of: 0.579 for ϕ_{p0} – the maximum quantum yield of primary photochemistry, 0.776 for trapping flux per PSII RC (TR_0/RC), -0.532 for dissipation flux per RC (DI_0/RC) and -0.719 for stomatal conductance (g_s). TR_0/RC showed a positive correlation (+0.553) also with root growth at a deeper soil level (0.30-0.60 m). Among abiotic parameters, T_{MEAN} CANOPY showed significant and negative correlation with root growth at all depths, -0.800 and -0.884 Person's coefficient for topsoil root growth and deeper soil, respectively. T_{MAX} CANOPY was correlated with T_{MEAN} SOIL (Person's coefficient +0.959) and all photochemical parameters (Supplementary Table 2). Soil temperature (T_{MEAN} SOIL) was also correlated with them, in particular DI_0/RC (0.617) and ϕ_{p0} (-0.728) and ϕ_{ABS} (-0.926).

Mean data for all abiotic, physiological and photochemical parameters have been standardized and used to run a multivariate analysis (PCA) (Figure 8).

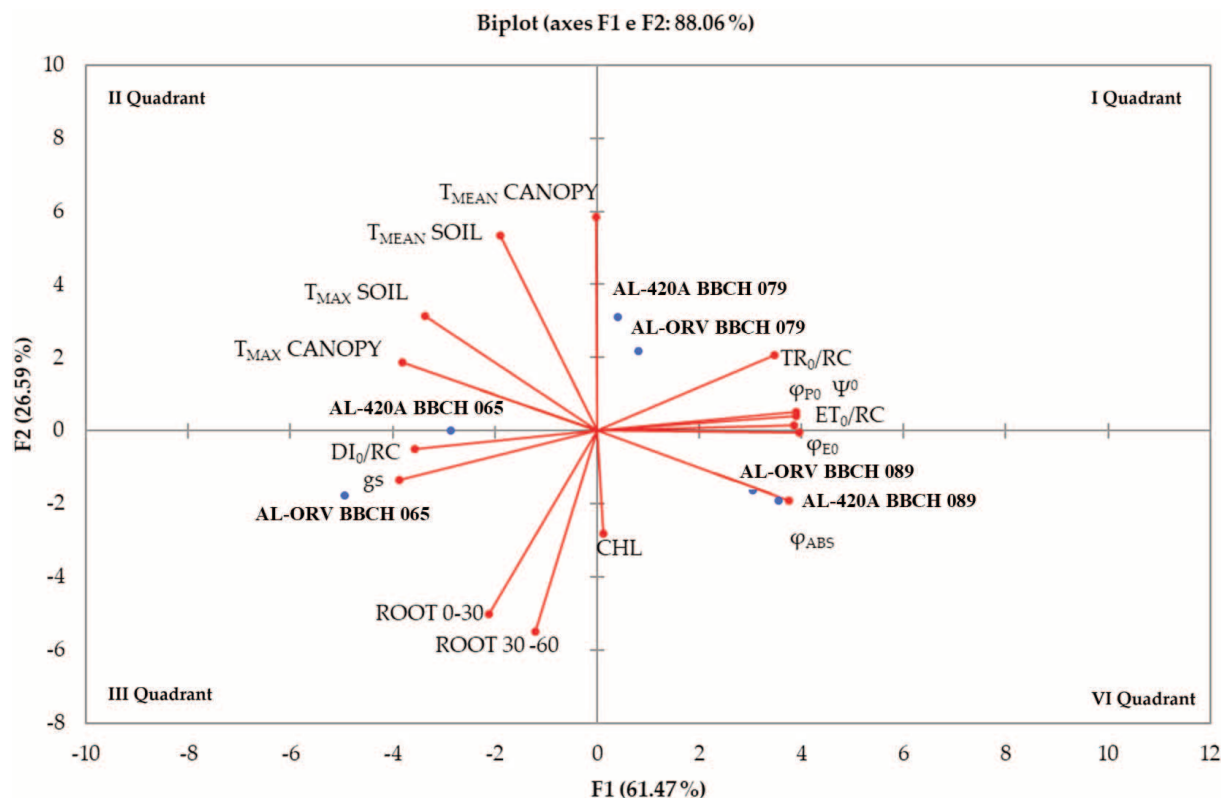


Figure 8. Multivariate analysis (PCA) obtained among microclimate data (mean and maximum soil – T_{MEAN} SOIL and T_{MAX} SOIL and canopy temperature T_{MEAN} CANOPY and T_{MAX} CANOPY during the growing season 2022, vine physiological measurements conductance (gs), mature leaves chlorophyll content (CHL), fine root growth (ROOT 0-30 and ROOT 30-60 respectively at t 0-30 and 30-60 cm soil depth), photosystem II (PSII) activity in terms of: ϕ_{P0} - the maximum quantum yield of primary photochemistry ; Ψ_0 - the probability that a photon trapped by the PSII reaction centre enters the electron transport chain; ϕ_{E0} - the quantum yield of electron transport, and ϕ_{ABS} - the performance index for energy conservation from photons absorbed by PSII until the reduction of intersystem electron acceptors; TR0 /RC - trapping , DI0/RC - dissipation and ET0/RC - electron transport for reaction centre (RC) for leaves of vine of the two tested vineyards: AL-420A (*cv* Aleatico on rootstock) and AL-ORV (*cv* Aleatico on own roots) during three phenological stages: BBCH065 - flowering, BBCH079 - berries touching, BBCH 089 - berries ripening.

The first two principal components (F1 and F2) explained the 88.06% of the total variance (Figure S1). Parameters such as ϕ_{P0} , Ψ_0 , ϕ_{E0} , ϕ_{ABS} , TR0/RC, ET0/RC, DI0/RC, gs, T_{MAX} SOIL, and T_{MAX} CANOPY, contributed mainly to F1, while ROOT 0-30 and ROOT 30-60, T_{MEAN} SOIL and T_{MEAN} CANOPY to F2, as shown by the correlation coefficients reported in Table S3. They indicate how much each variable contributes to a particular principal component. Biplot of PCA suggested that the main factor affecting the relationships between different OJIP-test parameters was the environmental conditions of tested vineyards (AL-ORV and AL-420A). A temporal gradient (phenological phases) is shown on the principal component F1 from left to right, which allows for the identification of the phenological phases of berry ripening (BBCH 079 - berry touching and BBCH 089 - berry ripening) in quadrants I and II of the biplot. All abiotic stressors measured related to temperature of soil and canopy were located in the second quadrant of the PCA biplot, and this indicate that they were responsible for the variation seen between AL-420A and AL-ORV during the seasons 2022.

DI0 and gs appear to be responsible for observed differences at BBCH 065 – Flowering; while TR0/RC, ET0/RC, ϕ_{P0} , Ψ_0 , and ϕ_{E0} appeared to be responsible for observed differences at BBCH 079 -

berry touch complete and 089 - berry ripening. Chlorophyll content (CHL) and root growth were negatively affected by mean soil and canopy temperature ($T_{\text{MEAN CANOPY}}$ and $T_{\text{MEAN SOIL}}$ - quadrant II), while the maximum temperature was negatively correlated with total photochemical efficiency of leaves (ϕ_{ABS}).

4. Discussion

Like other specialized and intensive crop productions, viticulture represents a highly vulnerable soil-plant-atmosphere system, which poorly withstands pests (Biasi et al., 2019) and extreme weather conditions (Brunori et al., 2022; Moresi et al., 2025), or events like wildfires (Brunori et al., 2020). In particular, abiotic stresses such as drought, salinity, extreme temperatures, and nutrient deficiencies, are most intensely experienced by plants at the root-soil interface (George et al., 2024) – a unique zone that acts as the primary interface between plants and their surrounding environment, where roots interact with soil moisture, nutrients, and microbial communities attempting to alleviate those stresses. Increasingly climatic emergencies raise concerns for viticulture, as soil plays a critical role in the balance of ecosystems and the quality of wine (Costa et al., 2023). Soil temperature affects vine performance, influencing growth, yield, and grape composition (De Rességuier et al., 2023; Van Leeuwen et al., 2020), yet its impact under extreme climatic conditions is poorly understood. A deeper understanding of soil temperature impact on grapevine can improve vineyard management and help predict crop performance and soil microbiome dynamics in the face of climate stress. In traditional growing areas, grapevine is still grown with its own roots, particularly under organic farming (Biasi et al., 2023). Findings showed that under extreme weather conditions, root genotype concurred to improve grapevine resilience in terms of root elongation during the most favourable periods (spring and autumn) and with a major root growth rate, especially in topsoil. A more reactive and functional grapevine root system helped leaves to perform good photochemical functions, analyzed by JIP test - based on fast chlorophyll fluorescence (CHL) kinetics and derived parameters, a tool to screen the capacity of plant adaptation to climate change (Bussotti et al., 2010). In particular, leaves of own-rooted vines exhibited an increase of all the photosynthetic efficiencies: ψE_0 - the efficiency of electron transfer and ET_0/RC - electron transport flux per reaction centre; and an enhancing the photosynthetic performance index ϕ_{ABS} . Better efficiency in electron transport could be interpreted as an acclimation response under drought-stressed (Swoczynna et al., 2022) combined to a greater photosynthetic performance index to a reduction of thermal dissipation (DI_0/RC) and a greater conductance. All these resilience traits showed a significant correlation with root growth in topsoil (0-0.30 m), respectively positively with ϕ_{P_0} – the maximum quantum yield of primary photochemistry, trapping flux per PSII RC (TR_0/RC), and negatively with dissipation flux per RC (DI_0/RC) and -stomatal conductance (gs). TR_0/RC showed a positive correlation also with root growth at a deeper soil level (0.30-0.60 m). Previous studies (Biasi et al., 2023; Catalani et al., 2024) have also shown how own-rooted vine showed differences in arbuscular mycorrhizal fungal (AMF) colonisation of fine roots. Roots of ungrafted vines were rich in vesicles representing the final colonization strategies to promote nutrient uptake and carbon storage (Requena et al., 2007; Wang et al., 2021). This promotes for own-rooted vines a faster bi-directional movement of nutrients that activates the rooting process early in the season (Hammer et al., 2024), higher in the grapevine flowering stage (de Souza Kulmann et al., 2022). The different AMF colonization of the *Vitis vinifera* root system contributes to modulating the root growth rhythms and consequently improves the plant's physiological response to climatic alterations. The ungrafted vine shows better resilience traits - greater stomatal conductance and higher leaf chlorophyll content under the same abiotic stressors, soil management and physical conditions (Biasi et al., 2023). The 420A rootstock genotype appears to exhibit a stronger tendency for deep rooting, given its capacity to flourish in dry and rocky soils, as well as its known lower vigor (Smart et al., 2006). However, a thorough assessment of their impact on the scion, particularly regarding eco-physiological traits for more seasons, is necessary.

5. Conclusions

The challenges posed by climate emergencies and extreme weather conditions are increasingly impacting viticulture, as monocultural systems exhibit heightened vulnerability to abiotic stresses. The root-soil interface is essential for nutrient uptake and water absorption but also serves as a critical barrier against various stresses intensified by climate change. Understanding and addressing the complexities of this interaction is vital for creating effective strategies to improve plant resilience, secure food supplies, and maintain ecosystems in a rapidly evolving environment.

Our findings indicate that ungrafted vines, which maintain their original root systems, display enhanced resilience traits such as better photosynthetic efficiency and root growth under stress conditions. The positive correlation between root development and plant physiological responses reinforces the importance of root genotypes in adapting to climate variations. Moreover, the unique relationship between grapevine roots and arbuscular mycorrhizal fungi plays a crucial role in nutrient acquisition and stress mitigation, further emphasizing the potential benefits of cultivating own-rooted vines. Exploring and addressing the complexities of the root-soil interface will be crucial for developing effective strategies to enhance plant resilience, ensure food security, and regenerate degraded agrosystems. As viticulture navigates an uncertain future, leveraging the resilience of ungrafted vines and optimizing rootstock selection could provide sustainable solutions for maintaining ecosystem balance and wine quality in the face of ongoing climatic challenges.

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Conflicts of Interest: The authors declare no conflicts of interest.

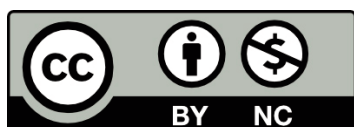
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