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Abiotic stresses in grapevine: ecophysiology and application of dendro-ecological techniques

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Preface

The work presented in this thesis was mainly carried out at the Department of Agricultural Sciences of the University of Naples Federico II, from November 2020 to January 2024.

The project was funded by the Campania region within the program «Ricerca e Innovazione» 2015-2017 Asse “Capitale Umano”, del Fondo per lo Sviluppo e la Coesione, “Dottorati A Tema Vincolato – Aree Interne/Aree Marginalizzate” and supervised by Prof. Veronica De Micco.

The project was carried out in partnership with the "Società Cooperativa Agricola - La Guardiense" where a period of 8 months was spent under the supervision of Dr. Marco Giulioli. Furthermore, for a period of 4.5 months, the research activities were carried out at the UMR BIOGECO laboratory at the University of Bordeaux and INRAE (France) under the supervision of Dr. Sylvain Delzon. Other collaborations included: Prof. Chiara Cirillo at the Department of Agricultural Sciences of the University of Naples Federico II; Prof. Carmen Arena at the Department of Biology of the University of Naples Federico II; Prof. Alessandro Tarantino at the University of Strathclyde (both in person and remotely; 1.5 months); Dr. Antonello Bonfante at the “CNR Isafom – Istituto per Sistemi Agricoli e Forestali del Mediterraneo” of Portici (NA), and Dr. Arturo Erbaggio (freelance).

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Chapter 3: Effects of foliar distribution of basal dust on morpho-physiological traits and yield of 'Falanghina' grapevine in Southern Italy 43

Introduction

This thesis project entitled "Abiotic stresses in grapevine: ecophysiology and application of dendro-ecological techniques" has a multidisciplinary approach with the aim of creating a synergy between research and farms. The programme aimed to deepen the studies on identifying sustainable vineyard management techniques and transfer them to the growers.

Viticulture is undergoing important changes due to climate change. Climate change is producing negative effects on viticulture and therefore management is becoming more and more complex especially in areas where the effects of increasing temperatures and decreasing rainfall frequency is severe, such as in the southern Mediterranean area. Furthermore, in areas where the introduction of irrigation is limited by governance or water availability, management is even more complicated. The impact of water stress on the vine, produces negative effects on productivity and quality of grapes, also influencing the quality of final products, such as wines. The increase in temperature produces a decoupling of ripening with a high accumulation of sugars, reducing acidity especially due to the degradation of malic acid and can produce an advance in ripening. From a technological point of view, wines can also suffer consequences on the aroma and phenolic composition. Furthermore, a strong alternation of production can occur over the years. These conditions put producing companies in serious difficulty.

The effects of climate change produce consequences on the physiology of plants, influencing fundamental physiological mechanisms such as photosynthesis and changes in their anatomical structures. The ability of acclimation to environmental stresses is species- and cultivar-specific and plants' responses also vary based on the pedo-climatic conditions and cultivation practices. The acclimation strategies that plants adopt to face environmental stresses are targeted to maintain a high photosynthetic and hydraulic efficiency to complete the ripening of the fruit. The improvement of water use efficiency, therefore, especially in conditions of strong and prolonged stress, is essential to preserve the equilibrium between vegetative growth and yield, as well as to maintain high grape quality. In grapevine, to improve the water use efficiency and protect the plant from adverse effects of climate change, in recent years, the interest in the use of products as inert materials of mineral origin (particle films) has increased. However, the vines' responses depend on the cultivar and the severity of the stress. In this project, to identify strategies capable of mitigating the negative effects of abiotic stress, such as water stress and high light intensity, a three-year experimental trial was performed on *Vitis vinifera* sub. *vinifera* 'Falanghina' to study the effect of the distribution of basalt dust under two water availability conditions. The study was conducted at the commercial vineyard of the agricultural company "La Guardiense" located in Guardia Sanframondi (Benevento), Italy.

The thesis is organized as follows:

Chapter 1 is a review on the effects of the most used particle films in viticulture on the main eco-physiological parameters. After a focus on the control of leaf gas exchange and photoprotection, this chapter highlights the lack of information on the effects of particle film distribution on morpho anatomical traits despite their fundamental role in the acclimation capability of the different grapevine cultivars has been recognized. A literature analysis was conducted to evaluate the studies' distribution by geographical area and type of particle film used in the last 15 years. The manuscript has been published in Progress in Botany (Springer Nature Switzerland).

(De Micco et al., 2024 ; https://doi.org/10.1007/124_2024_84)

Chapter 2 focused on the characterization of the study area where the three-year experimental trial was conducted. The pedoclimatic study included: a pedological survey, geophysical approach, collection of meteorological data with a station installed in the field, and the evaluation of water availability for vines using TDR probes installed at different depths and indirect method measurements.

Chapter 3 presents the results of the experimental trial in which the effects of basalt dust distribution, combined with two different water regimes were evaluated on *Vitis vinifera* subsp. *vinifera* ‘Falanghina’ in Southern Italy (Guardia Sanframondi, Benevento, Campania region) over three years (from 2021 to 2023). The treatments were: NDR (No Dust distribution-Rainfed), NDI (No Dust distribution-Irrigated); DR (Dust distribution-Rainfed) and DI (Dust distribution-Irrigated). The impact of basal dust was evaluated on plant growth, eco-physiological performance, leaf and grape mineral content, yield and grape quality.

Chapter 4 is focused on the methodology applied to perform eco-physiological measurements during the evaluation of the effects of basalt dust on grapevine. This study aimed to evaluate whether the presence of dust on the leaves may affect PSII photochemical efficiency and gas-exchange measurements, thus leading to biased results. The manuscript has been published in Photosynthetica (International Journal for Photosynthesis Research).

(Petracca et al., 2023; DOI: 10.32615/ps.2023.013)

Chapter 5 focuses on the effects of basalt dust distribution, combined with two different water regimes (ref. chapter 3) on anatomical traits in leaves and stem-wood to highlight possible modulation of eco-physiological acclimation ability as mediated by the organ and tissue traits.

Chapter 6 reports the characterization of the vulnerability to embolism at the leaf and stem level of four autochthonous cultivars of the Campania region:

‘Aglianico’, ‘Fiano’, ‘Greco’ and ‘Falanghina’. The analysis was conducted with innovative methods such as the use of optical technique (leaf vulnerability) and flow centrifugation methods (stem vulnerability) at the UMR BIOGECO laboratory at the University of Bordeaux and INRAE (France). To evaluate whether structural traits influence embolism vulnerability and to to investigate possible trade-offs between anatomical traits and eco-physiological behaviour at the leaf and stem levels, anatomical analyses were performed too.

Chapter 1

Particle film to mitigate the negative effects of climate change on grapevine leaf eco-physiology as mediated by anatomical traits

This chapter has been published as:

De Micco, V., Petracca, F., Cirillo, C., Arena, C. (2024). Particle Film to Mitigate the Negative Effects of Climate Change on Grapevine Leaf Eco-Physiology as Mediated by Anatomical Traits. In: Progress in Botany. Springer, Berlin, Heidelberg.

https://doi.org/10.1007/124_2024_84

Particle Film to Mitigate the Negative Effects of Climate Change on Grapevine Leaf Eco-Physiology as Mediated by Anatomical Traits

Veronica De Micco, Francesca Petracca, Chiara Cirillo, and Carmen Arena

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Chapter 2

Soil and water availability characterization at the study site

1. Introduction

Forecasting models predict that the Mediterranean basin will be characterized by an increase in the frequency and severity of drought events, raising concerns for resource management in agriculture (Masson-Delmotte et al. 2021). In grapevine (*Vitis vinifera* L. sub. *vinifera*), these conditions cause damage from excessive heat, solar radiation and water stress to plant growth and physiological processes with negative effects on grape yield and quality (Bernardo et al. 2018). Given recent and future climatic adversities, studies are increasing on plant tolerance to water stress to define and apply sustainable management of crops and resources, to maintain the rainfed management of vineyards in Italy, especially in the PDO (Protected Designations of Origin) region. To plan precise and efficient vineyard management, it is essential to know the physical properties of the soil which influence the hydraulic properties and therefore the water availability for the plants. Furthermore, there is spatial variability in the soil even at the farm scale which influences grapes' quality, the potential wines, and thus applicable viticultural practices (Bonfante et al. 2014). For this reason, in recent decades, the micro-zonation of vineyards has become a good practice for precision agriculture to achieve the right balance among the use of resources, agricultural yield and quality of production. Soil spatial variability evaluation can be estimated through non-invasive methods such as electrical conductivity measurements using electromagnetic induction sensors. These instruments generate an electromagnetic field from a transmitting coil that passes through the soil and is then intercepted by the receiving coil, with an output voltage that is related to the soil's apparent electrical conductivity. The secondary field amplitude will differ from the primary field due to soil characteristics (clay content, water content and salinity) (Hendrickx and Kachanoski, 2002). For a more accurate investigation, these mobile instruments are equipped with GPS sensors (Global Positioning System) georeferencing the data to have a site-specific characterization through the Geographic Information System, GIS (Corwin and Lesch, 2005a). To evaluate the distribution of water in the soil at different depths, it is possible to apply different methods to measure the soil water content (SWC, percentage of water in a given volume of soil). In particular, the most used methodologies are based on thermogravimetric methods, or the dielectric properties of the soil (Gardner et al. 2000). In the first case, to measure gravimetrically soil water content, the wet samples are weighed before and after drying (Bittelli, 2011). Detection techniques based on dielectric permittivity involve the use, for example, of TDR (Time Domain Reflectometry) sensors. TDR probes use the travel time of a reflected wave of electrical energy along a transmission line based on the principle of soil electrical permittivity. The travel time is influenced by the water content in the soil. Furthermore, the use TDR probes allows both automation and the possibility of continuously monitoring the water content in the soil through measurements that can be carried out at intervals of up to 5 minutes (Gardner et al. 2000).

To characterize the soil of the study site, a geophysical-pedological approach was used to evaluate water availability for the vines. TDR probes were installed at different depths. Indirect method measurements were also applied to obtain a characterization of the SWC at multiple depth levels compared to TDR probes.

2. Materials and methods

2.1. Plant material and study site

The experimental trials of the PhD project were conducted in a commercial vineyard located in Guardia Sanframondi, BN, Campania, Southern Italy, at La

Guardiense farm (lat: 41.229231 lon: 14.592362, 84 m a.s.l.), over three years: 2021, 2022 and 2023.

The experimental field was in the Intermontana plain system, subsystem TET, soil unit TAS1 (soil series Taverna Starze; Terribile et al. 1996) on a Typic Calciustolls (P72) with a sandy loam texture and presence of anthropic material along the profile (Figure 1). The principal horizons are an Ap horizon (0-20 cm) with a developed sub-angular polyhedral structure, common rock fragments, and the presence of fine and medium roots, and a sub-superficial Bw horizon (from 20 to 70+) which differs from the surface horizon for the consistency of the aggregates which are moderately developed. Small iron-manganese concretions can be found along the soil profile.

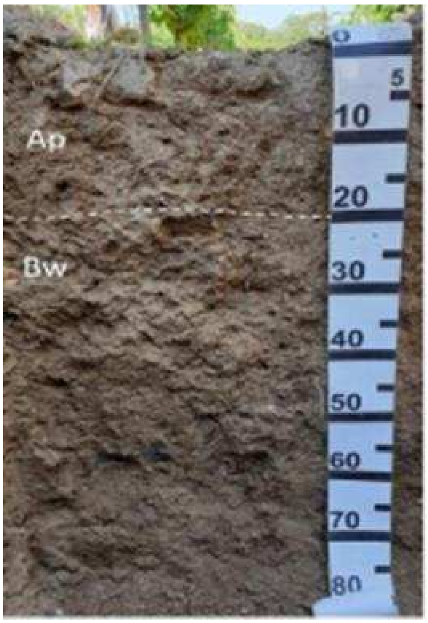

Figure 1. Representative soil of the experimental site.

Vines of *Vitis vinifera* L. sub *vinifera* 'Falanghina' are grafted on 157- 11 Couderc (*Vitis berlandieri* x *Vitis riparia*) rootstock. Vines, planted in 2010, are spaced by about 2.3 × 1.2 m (≈ 3600 vines ha^{-1}), and have E-W row orientation. The training system is double guyot, with about 20 buds per plant (72000 buds ha^{-1}). A plot of 200 m^2, including 6 rows (70 plants per row), was identified in the vineyard. The rows were divided into 4 experimental theses (1 row for treatment and 2 drift rows). The treatments were set by combining two factors: the presence or absence of basalt dust application and rainfed/irrigated water regimes. The trials were: NDR (No Dust distribution-Rainfed), NDI (No Dust distribution-Irrigated), DR (Dust distribution-Rainfed), and DI (Dust distribution-Irrigated (Figure 2).

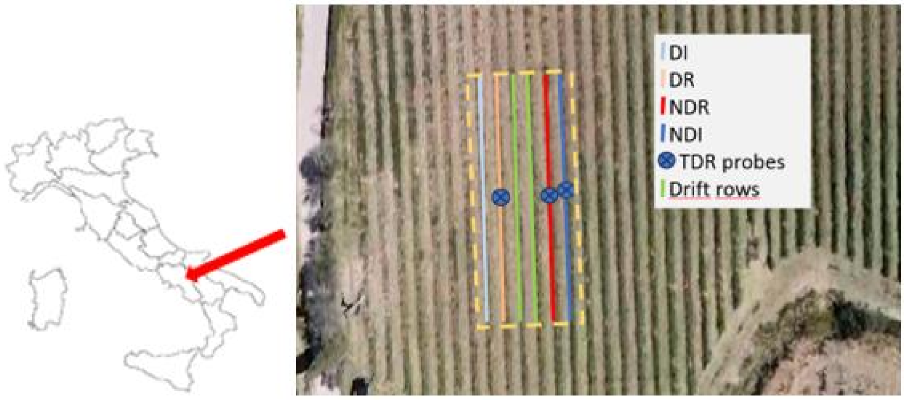

Figure 2. Experimental site and design at Guardia Sanframondi (BN), Southern Italy with four treatments: NDR (No Dust distribution-Rainfed), NDI (No Dust distribution-Irrigated), DR (Dust distribution-Rainfed), and DI (Dust distribution-Irrigated)

The particles of the dust had a diameter of less than 30µm. The basalt dust (Tecno beton, Giugliano in Campania, Naples, Italy) was dissolved in water (5% w/v; 5 Kg/100 L^{-1} water) and sprayed directly on the canopy with a shoulder sprayer pump (GeoTech SP 300 4T, Bruna, Perugia, Italy), equipped with three flat spray nozzles, operating at a pressure of 25 bar. During the experimental trials, the dust was distributed for a total of 7 treatments/season, always maintaining the same degree of leaf coverage, (from 24/05 to 08/09 in 2021; from 18/05 to 14/08 in 2022; from 19/06 to 12/09 in 2023) from flowering to post-veraison in the years 2021-2023, with additional distribution in case of leaching due to precipitation events. Two rows of vines were managed as rainfed, while two rows were irrigated to reintegrate water lost by evapotranspiration. The water volume for irrigation was calculated by:

$$ET_c = ET_0 * Kc$$

where ETc was crop evapotranspiration, Kc indicated the crop coefficient, ET_0 (reference evapotranspiration) was estimated by the Penman-Montheith method (Allen et al. 1998), using data and estimation model from the AgriSense loT Monitoring System (Netsens s.r.l., Calenzano - FI, Italy). Kc was estimated according to the plant type and growth. The volumes of water were calculated by subtracting the precipitation recorded through a meteorological station (ref. paragraph 2.3) from the estimated ETc.

2.2. *Pedological survey.*

A combined geophysical–pedological approach was used to manage the soil spatial variability on the selected vineyard and identify the experimental plots.

Soil survey (soil profiles and auger-drilling) was supported by geophysical survey techniques.

In this case study, the apparent soil electric conductivity (Eca; Corwin and Lesh, 2005b), was carried out by electromagnetometer (Profiler EMP-400; Codevintec Italiana Srl, Milano, Italy) both vertical (VDM) and horizontal dipole mode (HDM), in two depths (1.6m for VDM and 0.76m for HDM). The data recorded was georeferenced on a GPS-supplied data logger. Data post-processing was performed in SAGA software (https://saga-gis.sourceforge.io/). These vertical (VDM) and horizontal (HDM) ECa maps were used as baseline data for a pedological survey based on soil auger-drilling and soil profile description and to address the definition of field trial plots.

2.3. Meteorological data and SWC

Daily meteorological information (temperature, precipitation, wind, solar radiation) was collected through the Netsens AgriSens IoT weather station (www.netsens.com) installed at the experimental site. To analyze the state of water in the soil and the distribution of water, three TDR probes (TDR-310H) were installed at three different depths (-15, -35, -55 cm), for three treatments, DI, DR, and NDR.

In August 2023 (31/08/2023), SWC was also measured gravimetrically for DR and NDR, by taking soil samples between 12:00 and 14:00. Two samplings were carried out for each plot at different depths (-10, -20, -30, -40, -50, -60 cm). The samples were weighed to measure the fresh weight and then reweighed after oven dehydration (105°C) until stable weight. After estimating soil density and volume, SWC was calculated and compared to SWC measured with TDR probes.

3. Results and discussion

During the experiment, geophysical surveys and monitoring of the SWC were carried out using TDR probes to characterize the study area to better understand the plants water use behaviour. The results of the geophysical survey were used to identify a zone, in the selected vineyard, in which the expected soil spatial variability showed by ECa was the lowest (Figure 3).

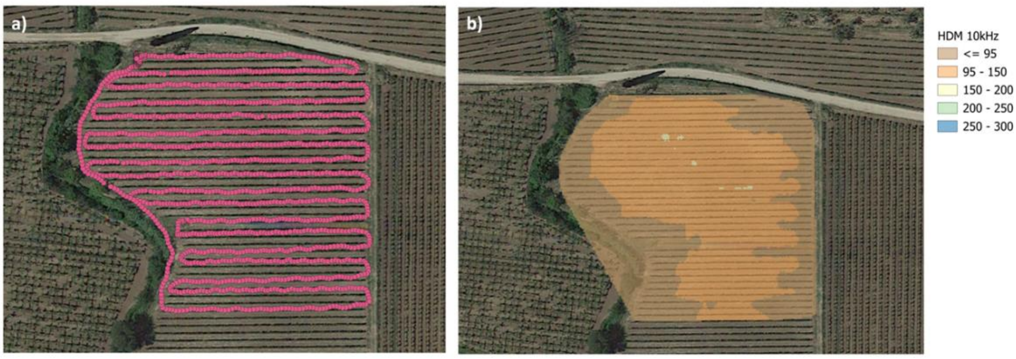

Figure 3. The geophysical survey was realized in the selected vineyard. In figure "a" the recorded points of measurement are reported while in figure "b" the spatialization of HDM ECa was measured at 10KHz.

The observation of data recorded through TDR probes in the three years of the experiment showed differences in the dynamics of water through the soil profiles at the NDR, DR and DI positions. As an example, Figure 4 shows the

SWC values detected at the three depths of -15, -35, and -55 cm, in the period 16/09/2021-30/09/21, after two rainfall events on 17/09 and 19/09 of 50.2 and 10.8 mm, respectively. The SWC at -15 cm depth of NDR probe showed lower values than DR and DI, with maximum values of 26.4% recorded on 20/09 which remains constant over time. Furthermore, for the DR probes, the values after 10 days of drought decreased by 4% from an initial value of 35.2 %, while DI had initial and final values of 36.4% and 34.4%, respectively. At -35cm the treatments showed the same trend with values at 20/09 of 23.1% of NDR, 42 and 40.8% of DR and DI. Also, at a depth of -55 cm, DR had the highest SWC with initial values of 52.3 % and final values of 44.6%. NDR had initial and final SWC values of 42.2 and 35% respectively.

The geophysical results confirmed a homogeneity among the rows and, therefore, the lower SWC values for NDR compared to DR and DI could be due to plant behaviour, especially at -15 and -35 cm, where fine and medium roots were present. This behaviour may be related to dust applications on the leaves. It has been reported that plants treated with dust have lower transpiration than untreated ones, thus preserving the good water status of the plants (Boari et al. 2015). Furthermore, during the monitoring period, the NDR kept the SWC constant in the top 35 cm. Probably the lower availability of water had led to a strong stomatal control of evaporative losses to save water and maintain constant leaf water potential (LWP) with an isohydric behaviour (Bucci et al. 2005).

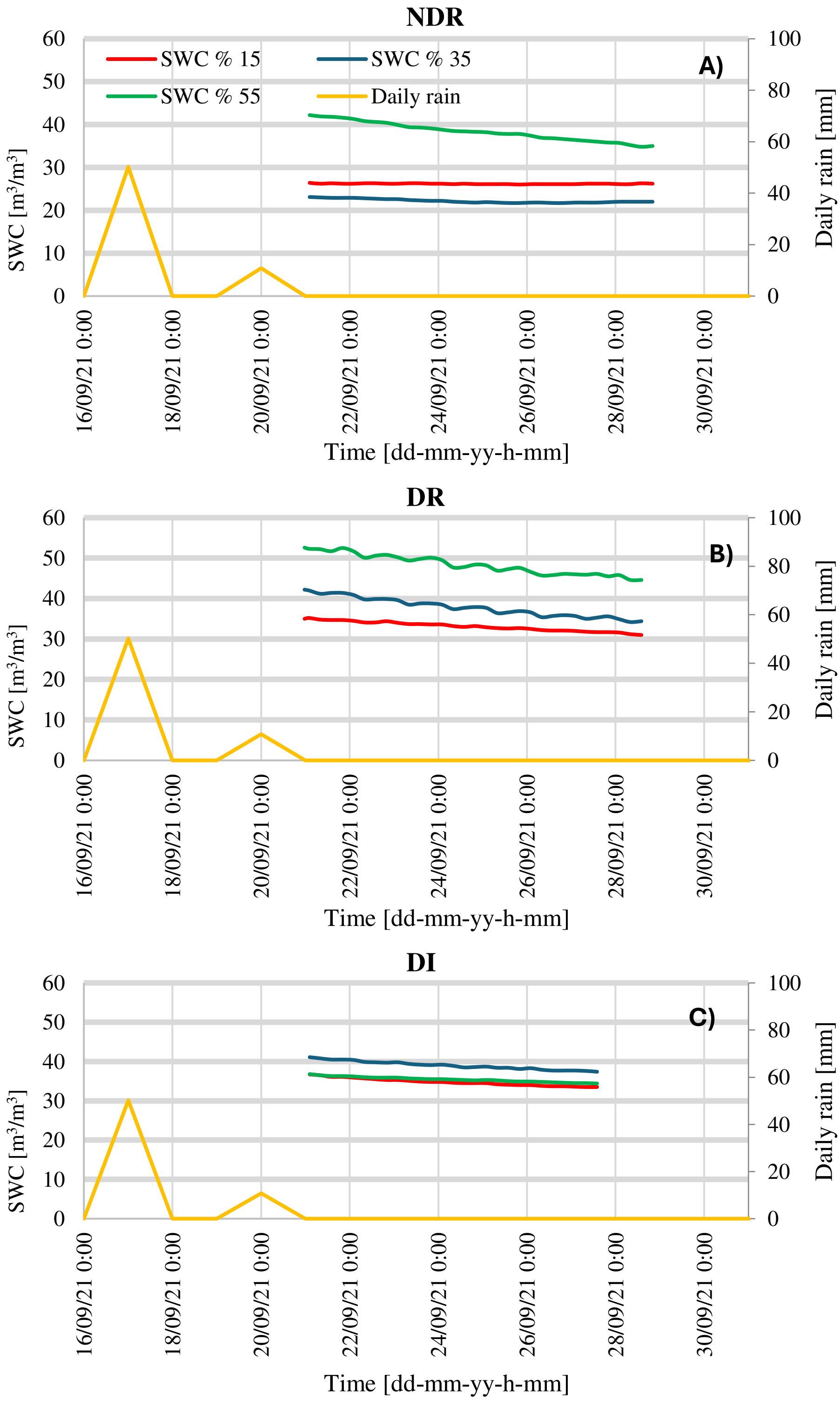

Figure 4. Soil water content (SWC) was measured with the dielectric method, with TDR probes, in three treatments: NDR (No Dust distribution-Rainfed, 4A), DR (Dust distribution-Rainfed, 4B) and DI (Dust distribution-Irrigated, 4C).

The graph in Figure 5 shows the values measured by the probes and the SWC calculated from the soil samples through the direct method on 30/08/2023. The measurements were carried out after a rainfall of 19 mm, which occurred in the previous days. As shown in the graph, they are completely overlapping in the range of depth included between 25 and 45 cm, whereas slight differences were observed in the soil between 15 and 25 cm (9%) and below 45 cm (4.24%).

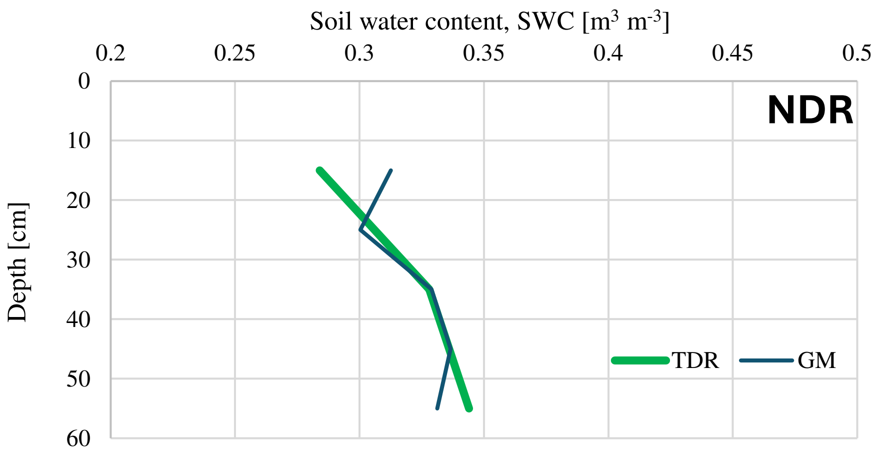

Figure 5. Comparison between soil water content values measured on 30/08/2023 with the dielectric method through TDR probes and gravimetric methods (GM). NDR (No Dust Rainfed).

Conclusion

The study area characterization through a geophysical-pedological approach, identified an area in which the spatial variability of the ECa was low, thus allowing excluding interference of the different soil properties on the effects of dust distribution and irrigation regime. The SWC data collected by the TDR probes helped us to evaluate the different water availability and vines' water use. The investigations on the soil characteristics and properties are the basis for interpreting the data relating to the vegetative-productive performances and the physiological aspects of the plants monitored during the entire experiment and for evaluating the effect of the basalt dust and irrigation management with a multi-level approach.

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Chapter 3

The combined effect of basalt dust foliar distribution and water availability on morpho-physiological traits and yield of 'Falanghina' grapevine in Southern Italy

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*NS, *, **, and ***, Not significant or significant at $p < 0.05$, 0.01, and 0.001, respectively. Different letters within each column indicate significant differences according to Duncan's multiple comparison tests ($p \leq 0.05$).*

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<u>Chapter 4</u>

The basalt dust deposition on leaves does not influence the measurement of ecophysiological traits in grapevine

This chapter has been published as:

Petracca, F., Arena, C., Cirillo, C., & De Micco, V. (2023). The basalt dust deposition on leaves does not influence the measurement of ecophysiological traits in grapevine. Photosynthetica, 61.

10.32615/ps.2023.013

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DOI 10.32615/ps.2023.013 PHOTOSYNTHETICA 61 (SI): 241-246, 2023

Special issue on Recent advances in photomodulation in higher plants, algae, and bryophytes

BRIEF COMMUNICATION

The basalt dust deposition on leaves does not influence the measurement of ecophysiological traits in grapevine

F. PETRACCA[*], C. ARENA[**,***,+], C. CIRILLO[*], and V. DE MICCO[*]

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Chapter 5

Leaf and wood anatomical responses to water stress in 'Falanghina' grapevine (*Vitis vinifera* sub. *Vinifera*) as mediated by basalt dust foliar distribution

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Chapter 6

Vulnerability to xylem embolism and wood functional anatomical traits in autochthonous Campania cultivars

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Conclusion

The work of this thesis was funded by the Campania region under the program -PON Dottorato Industriale- “Ricerca e Innovazione 2014-2020” /Piano Stralcio “Ricerca e innovazione 2015-2017“- Fondo per lo Sviluppo e la Coesione (FSC).- Dottorati A Tema Vincolato. The project was carried out in partnership with the "Società Cooperativa Agricola - La Guardiense" and in collaboration with the University of Bordeaux and INRAE (France), the Department of Biology of the University of Naples Federico II, the University of Strathclyde and the "CNR Isafom - Institute for Agricultural and Forestry Systems of the Mediterranean" of Portici. This project aimed to investigate the morpho-functional aspects of grapevine plants in response to drought to suggest possible mitigation strategies for sustainable vineyard management in the context of climate change. A field experiment was set up to evaluate the effects of the distribution of basalt dust on ‘Falanghina’ plants grown in southern Italy, in the Sannio wine district, combined with two levels of water availability. The results, obtained through eco-physiological monitoring, morpho-anatomical and vegetative-productive characterization, suggested that the use of particle film can mitigate the negative effects of water deficit. The responses were also influenced by climatic characteristics, confirming that vines have a high degree of plasticity and that their ability to adapt is also a function of soil-climate variability. A strong relationship between the anatomical traits of plants and their physiological acclimation mechanism was confirmed indicating the differential ability of Falanghina vines to modulate morpho-functional traits in years characterized by different climatic conditions. A deeper focus on leaf and wood functional anatomical traits and xylem vulnerability to embolism was devoted to four Campania Region autochthonous cultivars for which such information is completely lacking. Indeed, there is increasing awareness that considering the hydraulic traits related to plant drought resistance, thus vulnerability to embolism, is needed to recommend grapevine cultivars adapted to the local climate to allow viticulture sustainability under climate change.