

Effects of Partial Rootzone Drying and Subsurface Irrigation on Vine Physiology and Fruit Composition in *Vitis vinifera* cv. Cabernet Sauvignon

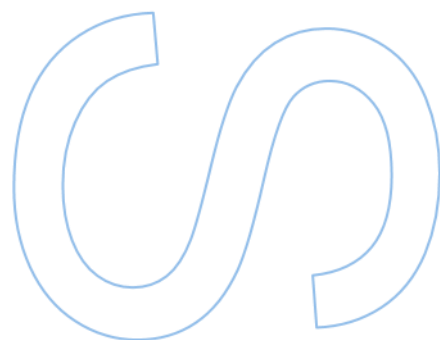
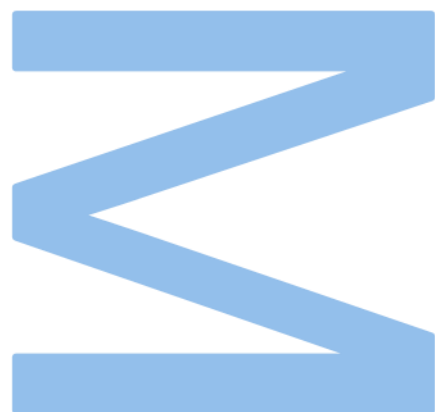
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Master in Agricultural Engineering

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Resumo

As alterações climáticas desafiam cada vez mais os ecossistemas mundiais. A viticultura na Califórnia enfrenta, quase todos os anos, crises devido à escassez de água e existe um aumento grande de leis e regulamentos rigorosos relacionados com o uso da água para cultivo, o que representa uma ameaça séria tanto para a produtividade como para a qualidade das vinhas, especialmente porque o setor é maioritariamente dependente de irrigação.

A Irrigação Radicular Alternada (PRD – Partial Rootzone Drying) é uma técnica de irrigação conhecida pelo seu potencial para reduzir o crescimento vegetativo e para melhorar a eficiência do uso de água (WUE), sem comprometer o estado hídrico e o rendimento das videiras. Fundamentalmente, PRD consiste em gerar diferenças espaciais na disponibilidade de água no solo, expondo diferentes partes do sistema radicular das videiras a ciclos alternados de secura e humidade. No entanto, a aplicação desta técnica tem enfrentado vários desafios com resultados extremamente variáveis, nomeadamente quanto à transição de ensaios em vasos e condições altamente controladas, para vinhas comerciais em plena produção.

Neste estudo, nós repensamos PRD, integrando esta estratégia com irrigação de subsuperfície de modo a criar uma abordagem inovadora destinada a maximizar a heterogeneidade espacial e temporal da água no solo, com o objetivo de replicar a uma escala comercial os benefícios descritos em ensaios de pequena escala. Esta nova abordagem consiste em irrigar as entrelinhas de uma forma alternada, utilizando tubos de irrigação gota-a-gota enterrados ao longo das entrelinhas, no centro das mesmas. Avaliámos os efeitos desta técnica em vários componentes de crescimento, rendimento e desempenho fisiológico das videiras, assim como o desenvolvimento e composição dos bagos, e a eficiência do uso de água ao nível intrínseco (WUE_i) e agronómico (WUE_c).

Em 2023, foi realizada uma experiência de campo numa vinha típica da Área Vitícola Americana (AVA) de Lodi, na Califórnia. A vinha, de Cabernet Sauvignon, foi plantada em 2014, conduzida em sistema de Cortina Dupla Horizontalmente Dividida (GDC – Geneva Double Curtain) onde cada videira tem uma configuração de cordão quadrilateral, muito típica do Central Valley da Califórnia. Quatro tratamentos de irrigação foram aplicados: Controlo/Irrigação Convencional (CTRL), Irrigação de Subsuperfície (SSI), Irrigação Radicular Alternada (PRD1) e Irrigação Radicular Fixa (PRD2). Todos os tratamentos foram irrigados a 80% da evapotranspiração da cultura (ET_c). Sensores de humidade do solo (WATERMARKS) e caudalímetros monitorizaram a eficácia da irrigação/tratamentos e os padrões de secagem do solo.

Os tratamentos de PRD estabeleceram com sucesso zonas radiculares “húmidas” e “secas” distintas. As videiras de PRD1 e PRD2 exibiram significativamente maior crescimento vegetativo, com medições de Fractional Cover (FC) 10–13% superiores às do CTRL. Os pesos da lenha de poda no final da temporada corroboraram estas observações, com ambos os tratamentos PRD apresentando um aumento médio de 1 kg/videira em comparação com CTRL. Estes dados indicaram que PRD1 e PRD2 já possuíam canópias maiores antes início da aplicação dos tratamentos, um potencial “carry-over” da adaptação das videiras a PRD no ano anterior.

Durante o período do ensaio, as variações no estado hídrico das videiras de todos os tratamentos (potencial hídrico de base ao meio-dia – Ψ_{stem} ao meio-dia; potencial hídrico foliar ao meio-dia – Ψ_{leaf} ao meio-dia; e temperatura da folha) foram principalmente influenciadas por variações nos padrões climáticos sazonais. As medições de condutância estomática sazonais ao meio-dia (g_{sw} ao meio-dia) apresentaram tendências mais claras e ligeiramente diferentes, com ambos os tratamentos PRD a exibir um intervalo de valores menor e mais consistente do que CTRL e SSI. Notavelmente, foram observadas alterações na relação diurna (medida por três pontos temporais distintos ao longo do dia) entre g_{sw} e Ψ_{leaf} em PRD1 quando comparado com todos os outros tratamentos, indicando benefícios fisiológicos derivados de PRD1 especificamente (PRD com a alternância entre zonas radiculares "húmidas" e "secas").

O rendimento aumentou significativamente em PRD1 e PRD2, representando um incremento de 68% e 63%, respetivamente, em comparação com CTRL e SSI, com os pesos dos bagos a seguirem as mesmas tendências. Os aumentos relativos dos componentes de rendimento foram principalmente impulsionados pelas diferenças no tamanho das videiras. A relação rendimento/peso de poda (Índice de Ravaz) não foi significativamente diferente entre tratamentos apesar de aumentar em PRD1 e PRD2, mas continuou boa o suficiente para as videiras conduzidas em GDC usadas neste estudo, resultando em níveis de qualidade aceitáveis em todos os tratamentos. Em ambos os tratamentos PRD, devido às canópias e bagos maiores, a acumulação de sólidos solúveis totais (TSS) foi atrasada e ocorreram reduções nas concentrações de compostos fenólicos na vindima. A WUE_i (A/g_{sw}), calculada a partir da taxa de assimilação líquida de CO_2 (A) e g_{sw} , não diferiu significativamente entre os tratamentos. Em contraste, WUE_c (rendimento/irrigação total) resultou em grandes melhorias significativas em ambos os tratamentos PRD.

Com base neste ensaio preliminar, esta abordagem inovadora de PRD com irrigação de subsuperfície à escala comercial aumentou o tamanho das canópias devido à realocação de água. Este aumento sugere que a realocação de água para a subsuperfície e para o meio das entrelinhas estimulou o crescimento radicular, resultando em raízes maiores e mais extensas, ajudadas em parte por um inverno e primavera mais chuvosos e frios. Os aumentos no tamanho das videiras revelaram grandes efeitos no rendimento, mas as canópias maiores prejudicaram a qualidade dos bagos. Serão necessários estudos adicionais para discernir completamente o efeito de PRD dos benefícios da irrigação de subsuperfície. Será também importante investigar os processos subjacentes à resposta a PRD (com, por exemplo, análises hormonais e mapeamento de raízes) e testar a estabilidade dos benefícios observados ao longo de vários anos com diferentes características climáticas.

Palavras-chave: Eficiência do Uso da Água, Práticas de Rega, Rega Espacial e Temporal, Heterogeneidade da Humidade do Solo, Sistema Radicular, Condutância Estomática, Trocas Gasosas, Estado Hídrico da Videira, Crescimento das Canópias

Abstract

Climate change is challenging the world's ecosystems across the globe. California's viticulture is increasingly troubled by water scarcity and stricter regulations related to water usage, which pose a significant threat to both crop productivity and quality, particularly because the sector depends so heavily on irrigation. Partial Rootzone Drying (PRD) is an irrigation technique known for its potential to enhance water use efficiency (WUE) without compromising plant water status and yield; however, its application at a commercial scale has faced many implementation challenges and has been shown to be unreliable.

In this study, we revised PRD by integrating it with subsurface irrigation to create a novel approach aimed at maximizing spatial and temporal soil moisture heterogeneity. We evaluated the effects of this novel PRD approach on grapevine performance, yield, grape composition, and WUE. In 2023, a field experiment was conducted in a classic, commercial, split-canopy (Geneva Double Curtain) trellised cv. Cabernet Sauvignon vineyard within the Lodi American Viticultural Area (AVA) in California, USA. Four irrigation treatments were applied: Control/Conventional Irrigation (CTRL), Subsurface Irrigation (SSI), Partial Rootzone Drying Alternated (PRD1), and Partial Rootzone Drying One-Side (PRD2). All treatments were irrigated at 80% Crop Evapotranspiration (ET_c). Soil moisture sensors and flowmeters monitored irrigation/treatment effectiveness and soil drying patterns; vine physiological responses, yield, and fruit composition were measured.

The PRD treatments successfully established distinct “wet” and “dry” rootzones. PRD-treated vines exhibited a significant increase in canopy growth: fractional cover (FC) measurements were 10–13% higher than CTRL; end-of-season pruning weights corroborated these findings with PRD vines averaging a 1 kg/vine increase versus CTRL. These data indicated that PRD1 and PRD2 already had larger canopies at the beginning of the season, a potential carry-over effect from their adaptation to PRD in the previous year. Seasonal variations in the water status of the treatments (midday stem water potential – midday Ψ_{stem} ; midday leaf water potential – midday Ψ_{leaf} ; and leaf temperature) were mostly driven by seasonal weather patterns. Midday stomatal conductance (midday g_{sw}) measurements led to slightly different trends, with both PRD treatments exhibiting a narrower range of values. Notably, changes in the diurnal relationship between g_{sw} and Ψ_{leaf} were observed in PRD1 compared to all other treatments, indicating benefits to vine physiology derived from the alternation of the “wet” and “dry” zones.

Yield was significantly increased in both PRD1 and PRD2, representing a 68% and 63% increment compared to CTRL and SSI, and berry weights followed the same trend as yield. Changes in yield components were primarily driven by differences in vine size. The yield-to-pruning weight ratio (Ravaz Index) was not significantly different between treatments. Still, it was optimal for the split-canopy trellis used in this study, resulting in acceptable quality levels across all treatments. In the PRD treatments, the larger canopies and bigger berries led to a delayed accumulation of total soluble solids (TSS) and resulted in lower concentrations of phenolic compounds at harvest. Intrinsic water use efficiency (WUE_i), calculated from gas exchange measurements (A/g_{sw}), did not differ significantly between treatments. In contrast, agronomic water use efficiency (WUE_c) resulted in large positive improvements in both PRD treatments.

Based on this preliminary test, the novel approach of combining PRD with subsurface irrigation at a commercial scale increased canopy size due to water relocation. Enhancing vine capacity showed large effects on yield, but the larger canopies negatively impacted quality. The increase in canopy size suggests that the relocation of water subsurface and into the interrows stimulated root growth and led to an overall bigger root system. Further work is required to fully discern the effect of PRD from the benefits of subsurface irrigation and test the stability of PRD benefits across various seasons with different weather fingerprints.

Keywords: Water Use Efficiency, Irrigation Practices, Spatial and Temporal Irrigation, Soil Moisture Heterogeneity, Root System, Stomatal Conductance, Gas Exchange, Vine Water Status, Canopy Growth, Fractional Cover, Crop Load

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List of Abbreviations

A – Net CO₂ Assimilation Rate
ABA – Absciscic acid
AVAs – American Viticultural Areas
CTRL – Control Irrigation
DI – Deficit Irrigation
DOY – Day Of The Year
DRZ – Direct Rootzone Irrigation
E – Transpiration Rate
ET_c – Crop Evapotranspiration
ET_o – Reference Evapotranspiration
FC – Fractional Cover
GDDs – Growing Degree Days
g_{sw} – Stomatal Conductance to Water Vapor
IBMP – 3-isobutyl-2-methoxypyrazine
K_c – Crop Coefficient
LSD – Fisher’s Least Significant Difference
MCARI – Modified Chlorophyll Absorption Reflectance Index
MOG – Material other than Grape
NDVI – Normalized Difference Vegetation Index
NOPA – Nitrogen by o-phthaldialdehyde
PDT – Pacific Daylight Time
PRD – Partial Rootzone Drying
PRD1 – Partial Rootzone Drying Alternated
PRD2 – Partial Rootzone Drying One-Side
PRI – Partial Rootzone Irrigation
PVC – Polyvinyl Chloride
PWs – Pruning Weights
RDI – Regulated Deficit Irrigation
SDI – Sustained Deficit Irrigation
SE – Standard Error
SOC – Soil Organic Carbon
SOM – Soil Organic Matter
SSI – Subsurface Irrigation
TA – Titratable Acidity
TSS – Total Soluble Solids
VM – Vine Measurements
VRI – Variable Rate Irrigation
WUE_c – Crop Level Water Use Efficiency
WUE_i – Intrinsic Water Use Efficiency
WUE_{inst} – Instantaneous Water Use Efficiency
WUE_{plant} – Whole-plant Water Use Efficiency
YAN – Yeast Assimilable Nitrogen
δ¹³C – Carbon Isotope Discrimination
Ψ_{leaf} – Leaf Water Potential
Ψ_{stem} – Stem Water Potential

1. Introduction

1.1 The Wine Industry and Climate Change: Global Perspectives, Californian Challenges and Water Management Pressures

2023 was a tough year for the global viticulture and winemaking sector: rising production and distribution costs driven by inflationary pressures led to higher wine prices and lower consumption; adverse climatic events and diseases culminated in historically low production volumes; vineyard surface area decreased globally, with the removal of vineyards across major vine-growing countries both for wine, table and dried grapes (OIV, 2023). Global wine export volume decreased, but the export value still reached 36000 million euros, with the average price per liter of exported wine hitting an all-time high of 3,62 euros (OIV, 2023).

The United States of America (USA) held its place as the fourth largest producer of wine in the world with 24,3 million hectoliters, accounting for 10,2% of global production, and maintained its position as the largest wine market and leading wine consumer (6680 million dollars, 33,3 million hectoliters) (OIV, 2023). The wine industry and its multiple branches are an essential and dynamic part of the US economy, accounting for 276000 million dollars in total economic activity (1,3% of Gross Domestic Product (GPD)), employing and supporting over 1,8 million Americans (WineAmerica, 2022).

In the USA, grape growing and wine production is predominantly located on the West Coast, with California accounting for 81% of the total production, generating a staggering 170000 million dollars in economic activity for the US economy (CAWG, 2022; TTB, 2023). California's climate is highly variable with a Mediterranean macroclimate and diverse mesoclimates – ranging from cool and coastal (i.e., North Coast American Viticultural Areas (AVAs)) to warm and inland (i.e., Central Valley AVAs), which provide a wide range of geographic conditions suitable for cultivating various wine grape varieties (Hannah et al., 2013; Parker et al., 2024). With over 100 wine grape varieties grown over 152 AVAs with characteristic *terroirs*, California offers a wealth of diverse and interesting wines.

Climate change poses many challenges to the world's ecosystems, economies, and societies, with adverse impacts in every region across the globe (IPCC, 2023). California is being actively threatened, and climate change indicators are becoming more evident (Cayan et al., 2008; Pathak et al., 2018). Temperature lows and highs are increasing at significant rates; heat waves are expected to become more frequent and severe; precipitation is becoming highly variable, and patterns are shifting; floods and droughts are increasing in number and magnitude (Cook et al., 2018; Gershunov et al., 2009; Swain et al., 2018). These trends are placing increasing pressure on California's agricultural industry (Pathak et al., 2018). Future impacts on California's crops may include crop yield declines, reduced accumulation of chill hours, increased water demand and water-induced stress, irregular and unreliable water supply, increased disease and pest pressure, and altered crop phenology (Elad & Pertot, 2014; Kerr et al., 2017; Lobell et al., 2011).

Unsurprisingly, climate change is impacting viticulture and winemaking all around the world, and a substantial body of review literature is available to assess its effects (Ollat et al., 2016a; Santos et al., 2020; van Leeuwen & Darriet, 2016; van Leeuwen et al., 2019; Venios et al., 2020).

Grapevine phenology, yield, and quality can all be affected by climate change (van Leeuwen et al., 2019). Rising temperatures have been linked with an earlier onset of phenological stages and a shortening of the growing season, leading to earlier grape ripening (Fraga et al., 2015; Santos et al., 2020). Higher temperatures during the growing season can lead to decreases in total berry acidity, higher trends of alcohol content, and a decoupling between technological and phenolic maturity (de Orduña, 2010; Venios et al., 2020). Reduced rainfall, increased drought events, or higher evaporative demand due to elevated temperatures create water deficits that can lower yield, berry quality, and bud fertility (Gutiérrez-Gamboa et al., 2021; van Leeuwen & Darriet, 2016). Shifts in grape berry moth phenology, changes in leafhopper distribution ranges, and the range expansion of grapevine mealybugs are now evident due to the direct and indirect effects of climate change. (Reineke & Thiéry, 2016; Santos et al., 2020).

Shifting spatial and temporal patterns of temperature and precipitation are expected to change viticultural suitability, reshaping the geographical distribution of wine regions and threatening wine typicity (Fraga et al., 2018b; van Leeuwen et al., 2019). One example of an endangered “wine identity” is the unique aroma, acidity, and relatively low alcohol content found in white wines from cool climate regions, such as Champagne and Rheingau. These characteristics are expected to be significantly affected by rising temperatures and increased water stress (Garcia de Cortazar-Atauri et al., 2017; Neumann & Matzarakis, 2011). However, the increased temperature and prolonging of the growing season may open the possibility of cultivating new grape varieties and integrating new wine styles (van Leeuwen et al., 2019). On the other hand, current winemaking regions in southern Europe (e.g., Alentejo, La Mancha, Campania) may become excessively dry and downsize, with increasing irregularity in yield and declining wine quality (Fraga et al., 2018b; Santos et al., 2018)

The winemaking sector must implement suitable measures to address the diverse impacts of climate change. This involves planning adequate adaptation strategies at the regional/local level to continue producing economically sustainable and quality wines (Ollat et al., 2016a; Santos et al., 2020). To specifically combat the temperature-driven advancing of grape phenology, growers should employ and combine techniques such as selecting cultivars, clones, and rootstocks that are later ripening, implementing late pruning, and adjusting the geometry and size of the canopy. This will help ensure that harvest dates remain in the optimal timeframe for the region (Ollat et al., 2016b; Santos et al., 2020). Generally, vineyard adaptations to drier growing conditions and increasing water scarcity can be made by planting drought-resistant/tolerant rootstocks and varieties, altering vine planting and training systems, implementing improved soil management practices, and applying irrigation (van Leeuwen et al., 2019; Venios et al., 2020). At the field scale, irrigation is one of the most effective methods for mitigating the negative impacts of water scarcity, and it is commonly used to enhance and standardize crop yield and quality, especially in many “New World” countries, such as the USA (Santos et al., 2020).

It is clear that current viticultural systems (and agriculture in general) need to keep evolving to meet current market demands in the face of climate change. Sustainable and efficient production must be prioritized, optimizing yields and reducing production costs, while maintaining or improving profitability. In California, agricultural efficiency is mainly improved through mechanization, automation, and better water management (Pathak et al., 2018). These factors are interconnected through advancements in irrigation technology.

The Californian wine industry is completely dependent on irrigation systems to compensate for insufficient rainfall. In the US, water demands have slightly increased despite big efficiency improvements across all water consumption areas. Both surface water (i.e., rivers and reservoirs) and groundwater (aquifers) use have intensified, and approximately 40% of all water withdrawn in the country is used for agricultural irrigation (ERS, 2023; PPIC, 2023b; TTB, 2023). Californian farms depend heavily on irrigation, using approximately 40 - 50% of the state's total water, and each year the amount of irrigated acreage rises (PPIC, 2023a). As groundwater extraction is increasingly relied upon, overdrafting - extraction beyond the aquifer's natural recharge limit - becomes challenging. Depletion of aquifers, impaired water quality, higher energy costs to pump water, diminished surface water flows, and land surface subsidence are the major detrimental impacts (Graham et al., 2021; PPIC, 2024). The San Joaquin Valley is the state's largest farming region, accounting for 50-60% of California's total agricultural production, and is suffering heavily from declining water supplies and overdrafting, exacerbated by climate change (PPIC, 2023c).

To address the prospect of long-term water shortages, over-extraction, and depletion of groundwater resources – especially amid prolonged droughts and climate change – California has implemented stringent water management legislation. Examples include the 2009 Senate Bill X7-7 (The Water Conservation Act) and the Sustainable Groundwater Management Act (SGMA) of 2014 (CDWR, 2022a, 2022b). The SGMA has a significant impact on agriculture. Its main goal is to protect groundwater resources by requiring local agencies to develop and implement strict plans for regulating and sustainably managing groundwater extraction. Consequently, agencies like the California Department of Water Resources (CDWR) enforce these regulations on various stakeholders involved in water management, such as irrigation districts. Irrigation districts must adhere to regulations and develop accurate water budgets that balance supply and demand. They need to collaborate with farmers and businesses to ensure more sustainable water practices. (CDWR, 2022b; PPIC, 2023d). In recent years, these regulatory requirements even gave rise to private companies such as LandIQ (LandIQ Inc., Sacramento, CA, USA) or NASA's OpenET (OpenET, Las Vegas, NV, USA), which use evapotranspiration-based models to approximate consumptive water use, providing reliable data to state regulators and growers/businesses alike, to help regulatory compliance (LandIQ, 2024; OpenET, 2024).

As the main water users, Californian farmers are facing mounting pressure from all sides – climate change and its many indicators; water scarcity and price increases; new legislation and regulatory constraints; escalating demands from the environmental and urban sectors; industry pressures – to transition to more sustainable water use, reducing crop water and/or improving efficiency (PPIC, 2023b, 2023c).

1.2 Enhancing Water Use Efficiency in Vineyards: Genetic Variability and Agronomic Practices

The term Water Use Efficiency (WUE) does not have a single precise definition, it's definition depends on the particular context in which it is being discussed (Hatfield & Dold, 2019). In current plant literature, WUE reflects a ratio between gains (carbon assimilated or crop yield) and costs (water transpired or water consumed by the crop), and can it be expressed and measured at different levels (leaf, plant or crop level) and at different time scales, from months to minutes (Hatfield & Dold, 2019; Tomás et al., 2014b).

At the leaf level, WUE is commonly estimated by measuring leaf gas exchange, made possible in the field by a portable photosynthesis system – e.g., the LI-6800 (LI-COR Inc., Lincoln, NE, USA). These instantaneous gas exchange measurements relate net CO₂ assimilation rate (*A*) to either stomatal conductance to water vapor (*g_{sw}*) to determine intrinsic water use efficiency (WUE_i, calculated as *A/g_{sw}*), or to the transpiration rate (*E*) to determine instantaneous water use efficiency (WUE_{inst}, calculated as *A/E*) (Medrano et al., 2015b; Schultz & Stoll, 2010).

Plant level WUE (WUE_{plant}) is the ratio between the plant's carbon/biomass accumulation (i.e., the plant's dry matter production) and its water consumption, representing a larger temporal and spatial scale (Flexas et al., 2010; Medrano et al., 2015b). Carbon Isotope discrimination (δ¹³C) – with the isotope ratio ¹³C/¹²C measured by isotope mass spectrometry – is a good (yet variable) indicator of WUE_{plant} and water stress in plants (Medrano et al., 2015b). Leaf samples can lead to high variability in results, so berries sampled at veraison prove to be the best for accurately assessing both plant water status and WUE_{plant} (Bchir et al., 2016).

When analyzing the relationship between leaf level instantaneous values and plant level values, a careful, context-specific approach is needed (Medrano et al., 2015b). Research has shown that in plants with complex canopies, like grapevines, these single leaf measurements don't accurately reflect whole plant photosynthesis and provide incomplete information. Factors such as leaf light exposure, position, age, interactions with other plant structures and other limitations (Tomás et al., 2014b), may explain the often reported lack of correlation (Douthé et al., 2018; Poni et al., 2009).

Crop WUE (WUE_c) can be described, from an agronomic perspective, as the amount of crop obtained (i.e., yield) per unit of water used (Gregory, 2004). Due to the direct practical balance between water input and crop yield, it is an effective metric for growers' decision making. That being said, studies of WUE at different levels (leaf and plant level) are necessary to explore improvements in WUE_c (Tomás et al., 2014b).

In the past few decades, a significant amount of research has been directed towards optimizing grapevine water use, with the existing literature roughly split into two fronts, as detailed by Medrano et al. (2015a) and Romero et al. (2022): one focusing on improving WUE through agronomic practices such as soil management, canopy management, and irrigation strategies, and the other exploring the potential of grapevine genetic variability. Irrigation strategies are the focus of this work.

Genetic Variability of WUE

Exploring the immense diversity in grapevine cultivars, clones, and rootstocks offers a promising solution to enhancing vineyard WUE. Current research trends are focused mainly on the intercultural and intracultural variability in the physiological response to water stress and the rootstock influence / rootstock scion interactions (Bota et al., 2022; Medrano et al., 2015a).

A bi range of genetic variability has been observed among grapevine cultivars in terms of their responses to drought, in regards to leaf photosynthesis, g_{sw} , and WUE (mainly at the leaf level) (Bota et al., 2022; Tomás et al., 2014a). The key mechanism in drought response is stomatal control, which regulates the compromise between water loss and CO₂ uptake, and grapevine cultivars have been traditionally categorized as isohydric (tighter stomatal control - "conservative") or anisohydric (less marked stomatal control - "optimistic") based on their stomatal behavior/regulation under drought, but recently this clear dichotomy is debated (Chaves et al., 2016; Medrano et al., 2015a). The hormone abscisic acid (ABA) plays a key role in stomatal control and is possibly responsible for the different levels of anisohydry observed among cultivars when faced with environmental changes (Marusig & Tombesi, 2020; Osakabe et al., 2014). Differences in ABA basal concentration, ABA sensitivity, and hydraulic related processes have been found among cultivars with contrasting water deficit responses (Martorell et al., 2015). There are other factors that, although showing high environmental plasticity, result in drought response variability between cultivars, such as the xylem architecture, hydraulic conductivity, and osmotic adjustment (Bota et al., 2022; Tortosa et al., 2016).

Studies on the genetic variability among clones have demonstrated significant differences in traits such as WUE (Tortosa et al., 2016). This large intracultural variability can account for most of the total WUE variability across multiple cultivars, and certain genotypes can perform consistently better than others under different growing conditions (Tortosa et al., 2016; Tortosa et al., 2020).

Rootstock selection plays a crucial role in grapevine cultivation, and rootstocks huge genetic variability provide adaptability to diverse growing conditions (Santos et al., 2020). Despite the complexity of interactions between rootstocks, scions, and environmental factors, studies show that rootstocks display traits such as heightened resistance to drought, cold, soil salinity, changed root architecture, and differences in water uptake and root-to-shoot signaling that affect plant water status and WUE (Bota et al., 2022; Ollat et al., 2016b).

Improvement of WUE Through Agronomic Practices

Soil management

Sustainable soil management usually comprises the minimization of soil organic carbon (SOC) losses through conservation agriculture techniques and the enhancement of soil organic matter (SOM) with the addition of organic materials (Romero et al., 2022). SOC directly improves surface soil aggregation and stability, and techniques that increase SOM improve the soil's water retention capacity and infiltration, reducing runoff and soil degradation risks (Hendgen et al., 2020; Lazcano et al., 2020). These practices

increase the usage of green water (water from natural sources like rainfall or snow), reduce evaporation and can reduce eventual irrigation requirements. As a result, appropriate vineyard soil management can improve WUE (López-Urrea et al., 2020; Medrano et al., 2015a; Santos et al., 2020). The addition of compost has been found to increase SOM, soil moisture retention, soil structural stability, and microbial activity, while also reducing soil bulk density and erosion (Medrano et al., 2015a). Zero tillage or reduced tillage can enhance soil carbon storage, improve soil structure and aggregation, increase hydraulic conductivity, water holding capacity, and WUE (Bai et al., 2018; Bordoni et al., 2019). Organic mulching is seen as an effective tool to improve WUE, as it enriches the soil with organic matter, prevents erosion, retains soil moisture, and improves the physico-chemical properties of the soil. It also regulates soil temperature, reduces soil water evaporation, and effectively controls weed growth (Gutiérrez-Gamboa et al., 2021). Cover crops, whether seeded or spontaneous, and in row spaces and/or under vines, can effectively increase SOM and improve soil quality through reduced water runoff and soil erosion, and enhancing soil fertility and structure (Romero et al., 2022). Cover crops may compete with grapevines for water and nutrients, especially in warmer countries, but reviews conclude that all vineyards should have soil covered by herbaceous cover crops at least during the rainy season (Medrano et al., 2015a; Santos et al., 2020).

Canopy management

Various methods of canopy management – e.g. pruning, leaf thinning – are widely employed to regulate the microenvironment surrounding grape clusters, enhancing fruit sanitary conditions, yield, and quality while also influencing light absorption by the canopy, thereby impacting photosynthesis, water loss, and ultimately WUE (Flexas et al., 2010; Medrano et al., 2015a).

Recently, the application of antitranspirants to the grapevine canopy has been studied, which may reduce transpiration losses, conserving water and preventing berry shrinkage (Gutiérrez-Gamboa et al., 2021). Treatments with engineered kaolin, a new and inexpensive particle film technology antitranspirant, increased grapevine WUE_i without affecting berry and cluster weight and quality (Brillante et al., 2016).

Planting density directly affects a vineyard's water consumption, with reductions in density allowing for higher control of grapevine water status and an increase in drought resistance (van Leeuwen et al., 2019). Modifications in training systems and row orientation optimize light interception by the canopy and radiation use efficiency, adopting training systems that promote shorter trunks increases vine WUE (Flexas et al., 2010; Santos et al., 2020).

1.3 Irrigation Strategies

Irrigation is used to enhance and standardize crop yield and quality, and extensive research has been conducted to enhance grapevine WUE through various irrigation technologies and strategies. Also, an important part of this research is related to the evaluation of irrigation scheduling and timing (Balint & Reynolds, 2014; Chalmers et al., 2010; Chaves et al., 2007; Dry et al., 2001; Keller et al., 2016; Schlank et al., 2024). Supplying irrigation to ensure the full evapotranspiration requirements of

grapevines may maximize vineyard productivity because it supports full canopy expansion, higher photosynthetic activity, and greater yields. However, this approach often comes at the expense of unsustainable water use and excessive vegetative growth which leads to lower grape quality parameters such as sugar concentration, phenolic compounds, and aroma precursors (Chaves et al., 2007; Medrano et al., 2015a).

Contemporary irrigation technologies and strategies can enhance WUE while simultaneously improving grape and wine quality (Chaves et al., 2007; Dry et al., 2001). Among these strategies, the application of water stress inducing approaches such as deficit irrigation – i.e sustained deficit irrigation (SDI), regulated deficit irrigation (RDI) – or partial rootzone drying (PRD) are the most significant (Romero et al., 2022). Even irrigation practices that minimize water use with acceptable impacts on production levels represent viable and cost-effective tools to cope with reduced water supplies (Montazar, 2021).

1.3.1 Deficit Irrigation

Deficit irrigation (DI) is a practice in which water is applied at rates that replace only a fraction of the grapevines' potential crop evapotranspiration (ET_c) (Chaves et al., 2007). From a physiological perspective, DI is commonly applied in a manner that maintains grapevine water status within a range of optimal thresholds (Romero et al., 2013). DI strategies need to be developed and optimized for each unique situation, and this involves considering various factors such as the edaphoclimatic conditions, the specific combination of variety/rootstock and its genetic variability, the availability of irrigation water, the established production limits, the desired style of grape and wine, etc.

Sustained Deficit Irrigation (SDI)

When water is consistently withheld (the same fraction of ET_c) throughout the entire growing season, this approach is referred to as sustained deficit irrigation (SDI) (Mirás-Avalos & Araujo, 2021). With SDI, the water deficit progressively increases as the season advances. This occurs due to a combination of uniformly applying a reduced amount of water and the depletion of soil water reserves. As a result, water stress develops gradually, allowing the vines to adapt to the deficits, especially in soils with significant water storage capacity (Feres & Soriano, 2007). For example, in a study by Buesa et al. (2017) on cv. Muscat of Alexandria, the SDI treatment irrigated at 50% ET_c maintained yields comparable to full irrigation while surpassing the other DI strategies in WUE_c and grape sugar accumulation. On the other hand, when Munitz et al. (2017) examined DI regimes in cv. Merlot, the SDI treatments were less effective than RDI, resulting in worse yields and wine quality.

Regulated Deficit Irrigation (RDI)

When a water deficit is enforced during specific plant phenological stages, it's known as regulated deficit irrigation (RDI). RDI is based on the principle that plant sensitivity to water stress – in terms of yield, quality, etc. – varies during the different phenological stages across the season and the extent of the stress (Medrano et al., 2015a).

RDI is an effective irrigation strategy for arid and semi-arid regions, as it shows significant potential for water conservation and improved irrigation efficiency. This method helps control excessive vine growth, enhances WUE_c and improves the quality of berries and wine (Chaves et al., 2007; Costa et al., 2007; Medrano et al., 2015a). Reducing the amount of irrigation water applied through RDI can lead to energy savings from lower energy consumption by the irrigation/pumping system (Romero et al., 2022), and water stress and reduced water availability from RDI can lessen pest and disease pressure in certain cases (Mirás-Avalos & Araujo, 2021).

Additionally, as reviewed by Romero et al. (2022), several studies have investigated various RDI strategies across multiple wine grape varieties and rootstocks. These studies consistently report improvements in WUE_c and berry quality when compared to conventional irrigation methods, SDI, and full irrigation. However, some noted yield losses between 10% and 24%, highlighting the importance of balancing water savings and quality gains against potential yield reductions when implementing RDI.

RDI is frequently applied during the two phenological stages that correspond with peak evaporative demand and vine water requirements – aimed at improving WUE_c and berry quality (Medrano et al., 2015a). Applying a water deficit from fruit set to just before veraison – where vegetative and reproductive growth phases overlap (Stage I) – primarily controls berry size and reduces vine vigor. When a water deficit is applied after veraison, during the berry ripening stage (Stage III), it mainly enhances the biosynthesis of anthocyanins and other phenolic compounds. Generally, a pre-veraison water deficit is more effective than a post-veraison water deficit in reducing vegetative and berry growth, but both pre and post veraison water deficits (light and moderate) can improve berry and wine quality in various ways, these include: improved cluster microclimate due to reduced canopy leaf area; reduced berry weight and size, along with a higher skin-to-pulp ratio; changes in endogenous hormone responses (such as an increase in ABA in roots, leaves, and/or berries); changes in the root-to-shoot ratio; enhanced expression of key genes involved in anthocyanin synthesis, sugar unloading and accumulation of amino acids (Alatzas et al., 2021; Romero et al., 2022).

A standard RDI methodology involves replacing a varying portion of the water lost through ET_c during the growing season (Costa et al., 2007). Irrigation scheduling management should be done through plant water status indicators, based on the vine's actual water demand, instead of only relying on methods such as soil moisture, weather or ET_c . Metrics like stem water potential (Ψ_{stem}), pre-dawn or midday leaf water potential (pre-dawn Ψ_{leaf} , midday Ψ_{leaf}), g_{SW} or WUE_i are used in physiologically driven effective RDI strategies (Barbagallo et al., 2021; Cifre et al., 2005).

Berry size reductions and thus cropping levels may often result from RDI treatments, and, although water application is reduced heavily during the RDI period, the effect on long-term WUE can be small or nonexistent, since post-RDI irrigation is normally applied and yields may be reduced. However, a sustained effect on stomatal conductance might be an alternative option (e.g., partial rootzone drying) (Davies et al., 2002).

1.3.2 Subsurface Drip Irrigation

Researchers define subsurface drip irrigation (SDI or SSI) as a method of delivering water below the soil surface, utilizing emitters and discharge rates similar to those used in surface drip irrigation. SSI has the potential to enhance WUE, plant health, growth, and yield compared to surface drip irrigation (Ayars et al., 2015).

Although not widely adopted, SSI is gaining broader acceptance for perennial crops with improved irrigation equipment and scheduling tools, especially when paired with the advantages of DI practices (Ayars et al., 2015; Zhang et al., 2017). SSI has been trialed in vineyards as part of water deficit irrigation management. In a study by Pisciotta et al. (2018), two methods of SSI were compared to conventional surface drip irrigation. The results showed that SSI reduced water consumption, yielding higher grape production with 20% less water with no significant differences in grape and must composition.

1.3.3 Direct Rootzone Irrigation

Direct root-zone irrigation (DRZ) is a novel and efficient subsurface irrigation system that delivers water directly to the rootzone at specific irrigation rates and delivery depths. This technique has recently been applied in cv. Cabernet Sauvignon (Ma et al., 2020b; Ma et al., 2019, 2020a) and cv. Chardonnay (Ma et al., 2023) through vertical PVC tubes at approximately 60 cm of soil depth, with a 15 cm length above ground.

Compared to conventional drip irrigation, DRZ improved cv. Cabernet Sauvignon grape yield by 9–12% and WUE_C by 9–11% while having minimal effects on berry composition and maintaining a better vine water status (Ma et al., 2020a). In cv. Chardonnay, DRZ maintained yield and enhanced berry quality while reducing the irrigation amount by 16–23%, improving crop water productivity by 23–34%, when compared to conventional surface drip irrigation (Ma et al., 2023). Additionally, DRZ altered root growth patterns by decreasing root development in the upper soil profile which encouraged deeper root growth. This shift towards deeper root systems potentially allows grapevines to extract water deeper soil layers, probably optimizing growth and reducing water stress during water scarcity events (Ma et al., 2023; Ma et al., 2020a).

Future research should focus on the detailed physiological responses of various cultivars to DRZ. Specifically, the relationship between rooting patterns and water distribution in soils, as well as a comprehensive economic analysis comparing DRZ to conventional subsurface and surface drip irrigation systems (Ma et al., 2023).

1.3.4 Partial Rootzone Drying

Partial Rootzone Drying (PRD), sometimes called Partial Rootzone Irrigation (PRI), is an irrigation technique that exposes plant roots to alternate “wet” and “dry” cycles. This method generates spatial differences in water availability within the soil, ideally ensuring that a part of the root system always experiences “dry” conditions, and at the same time, the irrigated or “wet” roots supply sufficient water to the rest of the plant to prevent severe water deficit. As a result, the drying roots trigger the production of ABA, and in the leaves, ABA induces partial stomatal closure as a mechanism to avoid drought stress. Overall, hydraulic signals, increased ABA levels, and changes in other hormones (cytokinins, ethylene), as well as the pH of xylem sap, influence the physiology of the shoots and leaves. In theory, PRD could save irrigation water and enhance WUE without

compromising plant water status and fruit yield (Dry et al., 2001; Romero et al., 2016a; Stoll et al., 2000b).

PRD was introduced in the Australian wine industry by 1995, formally in 1997 (Dry, 1997) and initially, the primary goal of PRD was to develop a strategy for controlling excessive shoot growth in irrigated vineyards. Dry (1997) conducted several experiments over three years using container-grown grapevines, which resulted in restricted vegetative growth but maintained crop level and improved fruit quality. Results of trials conducted in Australia using this system are available in Dry and Loveys (1999); Dry et al. (2000a, 2000b); Loveys et al. (2000).

Container split-rooted vines – where the root system is divided between two containers – and field split-rooted vines – where half of the root system is placed on either side of a plastic membrane buried in the field – were trialed by applying a continuous water deficit to one container/side (half of the root system) while ensuring the other one was well watered. This “half-drying” of the root system significantly reduced shoot growth rate and stomatal conductance, without altering shoot water status (Dry, 1997). Ensuing experiments demonstrated this effect to be transient, with shoot function eventually recovering without any intervention on the dried half of the root system. To extend the vigor-reducing effects of “half-drying”, a strategy was tested that involved alternating irrigation between the two halves of the root system approximately every 11 days, which was when the “dry” half was deemed dry enough (soil water content stopped decreasing), creating “wet” and “dry” cycles (Dry, 1997).

The strategy now known as “partial rootzone drying” successfully prevented shoot growth recovery and resulted in significant reductions in most measures of vegetative growth, all without negatively impacting vine yield. Grapevine water use was substantially reduced without compromising yield and even enhancing fruit quality, translating into a WUE that was double that of the control groups (Dry & Loveys, 1999; Dry, 1997).

Subsequent research has demonstrated that, under drying soil conditions, the production and transport of root-derived chemical signals such as ABA to the leaves seemed to play a central role in the grapevine response to PRD (Loveys et al., 2000; Stoll, 2000; Stoll et al., 2000a; Stoll et al., 2000b).

Stoll et al. (2000b) reported that, in PRD-treated vines, ABA concentration vastly increased in drying roots and to a lesser extent in leaves, when compared to fully irrigated vines, corresponding with a reduction in stomatal conductance. The production of ABA in the drying roots was also transient, with stomatal conductance recovery and shoot growth resuming when ABA production in the drying roots slowed down, reinforcing the need for a “continuous signal” (Loveys et al., 2000; Stoll et al., 2000b). The concentration of ABA in grapevine xylem exhibited a modest increase in response to PRD, and it has been shown that significant changes in stomatal conductance are linked to relatively small changes in xylem ABA concentration and the subsequent bulk leaf ABA (Stoll, 2000; Stoll et al., 2000b). These small changes may be accompanied by shifts in xylem sap pH and potential reductions in root cytokinin availability, all of which could contribute to the vine’s overall expression of stomatal conductance and response to environmental changes (Chaves et al., 2010; Stoll, 2000).

Considering these findings, PRD has proven to be quite effective with split-root grapevines grown under controlled conditions. In such setups, the separation between “wet” and “dry” irrigated areas is well-defined, either with the root system divided between two containers or by positioning each half of the root system on opposite sides of a plastic membrane buried in the field. Irrigation water is properly spatially applied, creating a system of calibrated water stress. However, a wider adoption of PRD strategies, with field-grown grapevines, across different environmental conditions, varieties, and soil types has yielded inconsistent results in the literature (Chaves et al., 2010; Intrigliolo & Castel, 2009; Romero et al., 2016b). Differences in evaporative demand, the volume of irrigation water applied to each rootzone side, the strength of chemical signaling during PRD, soil type or depth, variety and rootstock, or the timing of PRD alternation, are some of the factors that may have caused such diverse results (Dodd, 2009; Romero et al., 2014).

Almost all of the previously referenced PRD research in this section involved applying significantly lower volumes of irrigation water on PRD treatments compared to control irrigation treatments. This can make it unclear whether some of the observed effects, such as improved WUE or increased fruit quality, stem from PRD-specific mechanisms (e.g., root-derived chemical signaling) only or are mixed with the effects of reduced irrigation.

A PRD revision (Table 1) was conducted with the objective of finding several studies that examine the effects of partial rootzone drying on grapevines with as many different backgrounds and variables as possible, while staying relevant to the themes of this thesis. Some studies from the revision are described below just to highlight the variability in PRD responses.

Research has shown that intrinsic changes in vine physiology and berry secondary metabolism occur specifically due to PRD, in contrast to the effects of deficit irrigation. These changes have been reported in various field-grown grape varieties (Bindon et al., 2008; dos Santos et al., 2007; Romero et al., 2016b) and the hormonal changes induced by PRD appear to play a role. PRD irrigation may also impact root growth since PRD-treated vines tended to develop a deeper root system (dos Santos et al., 2007; Romero & Martinez-Cutillas, 2012b).

de Souza et al. (2005b) observed that the effects of PRD may be more pronounced in years with lower precipitation with a better drought responsive variety like cv. Castelão. Other studies indicate that PRD vines are more negatively affected by high temperature extremes during the growing season than RDI and SDI vines. This suggests that cooler and wetter years could improve the PRD response (Romero et al., 2016c).

Both Chaves et al. (2007) and dos Santos et al. (2007) showed vegetative growth restriction in PRD compared to equal irrigation DI, which led to reduced vigor and increased light interception in the cluster zone, potentially enhancing fruit quality with higher concentrations of anthocyanins and total phenols in berry skins. In dos Santos et al. (2007), this occurred without significantly reducing yield when compared to DI and FI (full irrigation), doubling FI's WUE. Other studies did not find any substantial changes in vegetative development or yield in PRD (Intrigliolo & Castel, 2009). De la Hera et al. (2007) only noted distinct alterations on vegetative development in the 3rd year of the study, when PRD was applied from budburst to harvest, possibly suggesting that an

earlier onset of PRD is preferable to a later one in order to enhance the PRD response. When compared to the equal irrigation conventional control, fruit set percentage and leaf area increased, contributing to increased yield without affecting berry weight or must quality.

Some field studies reported significant increases in WUE in PRD-treated vines (Conesa et al., 2015; Romero et al., 2016b). Others found no consistent improvements in WUE with PRD-treated vines (De Pau et al., 2011; Intrigliolo & Castel, 2009).

Several studies on field-grown grapevines on various locations (Baeza et al., 2005; Bravdo et al., 2004; Gu et al., 2004; Pudney & McCarthy, 2004) have reported no significant improvement in vine performance and vine water use when comparing PRD treatments and DI treatments at the same irrigation level. Differences in vine water relations between them were minimal and only occasionally observed. Physiological and viticultural effects were only evident when comparing the higher or lower irrigation levels – it was the irrigation amount that was the key factor in water stress, not the irrigation strategy. Similarly, several other studies did not find evidence for a more pronounced stomatal closure in PRD than in DI grapevines (De la Hera et al., 2007; de Souza et al., 2003; Rodrigues et al., 2008).

Antolín et al. (2006) and Poni et al. (2007) conducted controlled experiments with split-rooted grapevines in containers, applying PRD with a 50% reduction in water supply compared to conventional irrigation. In Antolín et al. (2006), despite both PRD and SDI receiving the same amount of water, PRD was significantly different in terms of vine performance, with higher yields, increased cluster and berry weight, larger leaf area and higher skin anthocyanin levels. It was suggested that the better performance might be related to the different pattern of berry ABA concentration in PRD. To assess the physiological responses of split-rooted container-grown Sangiovese grapevines, Poni et al. (2007) implemented PRD from pre-veraison until harvest to induce water stress. The PRD-treated vines showed several effects compared to the well-watered control, lower water status, fast cessation of shoot growth but no effect on crop level and yield components, steady increase in WUE_i, and PRD must showed higher concentrations of soluble solids and anthocyanins.

These contrasting results between field-grown grapevines and container-grown split-rooted grapevines point out the difficulties of implementing functioning PRD strategies in vineyards.

1.4 Study Objectives and Research Questions

PRD has shown significant potential for enhancing WUE while improving grape and wine quality. However, questions remain about how to optimize its application. As discussed before, a key aspect of PRD is its implementation in controlled conditions, such as the split-root container vines and the split-root field vines, where the separation between “wet” and “dry” zones is clearly defined. This distinct separation of the root system is fundamental for the application of calibrated water stress with the “wet” and “dry” areas, which seems to be necessary to achieve the beneficial physiological and agronomic responses sought after in PRD.

Another key aspect that needs to be clarified is the total amount of water applied. Some experimental designs fail to control for or equate irrigation volumes across PRD and the other irrigation treatments, making it challenging to isolate the specific effects of PRD. Numerous studies have noted this issue, as demonstrated in Table 1, highlighting that benefits of PRD, such as increased WUE and improved grape quality, may not be fully attributed to the unique physiological responses induced by PRD, but rather to the lower irrigation volumes associated with deficit irrigation. It's very important to standardize the amount of water applied across treatments to ensure that any observed vine performance improvements are precisely of PRD itself and not the reduced irrigation levels.

Based on these unclear aspects, the objective of this research was to address whether the physiological and agronomic responses associated with PRD under controlled conditions could be replicated in the field at the commercial scale. To achieve this, our PRD strategy consisted of maximizing the separation of the irrigation drip lines by placing them in the middle of the row spaces (interrows). With the study being conducted in a commercial vineyard, the drip lines were subsurface grade and were buried in the ground to allow mechanized vineyard operations and to protect the lines (subsurface irrigation).

The following research questions were formulated to guide the investigation, addressing both the theoretical underpinnings of PRD and the practical challenges of its field application. These questions aim to elucidate the efficacy, mechanisms, and scalability of PRD as a sustainable irrigation strategy for viticulture.

1. Is a novel approach to PRD, combining principles of subsurface irrigation and partial rootzone drying, capable of re-creating the benefits associated with PRD in controlled conditions at the commercial scale?
2. What are the benefits of applying PRD at a commercial scale using this novel approach in terms of vine performance, yield, and grape composition?
3. How does this new PRD approach affect water use efficiency at the intrinsic (WUE_i) and agronomic (WUE_c) levels?

Table 1 – Revision on the effects of Partial Rootzone Drying in grapevine physiology, yield and fruit quality

Reference	Country	Species and Variety	Growing Conditions	Treatments	PRD Method	Irrigation System and Amount	PRD-related key findings
Dry (1997) (PhD Thesis) (led to: (Dry & Loveys, 1999; Dry et al., 2000a, 2000b; Dry et al., 2001))	Australia	<u>Container-grown:</u> <i>Vitis vinifera</i> cv. Chardonnay, cv. Syrah; <i>Vitis champini</i> cv. Ramsey; <i>Vitis berlandieri</i> x <i>Vitis riparia</i> cv. Kober 5BB; <i>Vitis berlandieri</i> x <i>Vitis rupestris</i> cv. Richter 110 <u>transferred to the field:</u> <i>Vitis vinifera</i> cv. Cabernet Sauvignon	- Container-grown and kept (greenhouse and open field); - Container-grown, transferred to field (various soil types)	<u>Comparisons between:</u> - Both root sides irrigated, only one side irrigated, no irrigation - Both root sides irrigated, only one side irrigated - Both root sides irrigated, only one side irrigated (alternating irrigated side)	- Two drip emitters per container/vine - Split-rooted vines, root system divided between two containers - Split-rooted vines, half of the root system placed on either side of a plastic membrane buried in the field	- Drip irrigation based on volumetric soil water content - Alternation of "wet" and "dry" sides based on volumetric soil water content - PRD-treated vines received substantially reduced water amounts (approx. 50%)	- Half-drying (half of the root system slowly dried while the other half remained well-watered) led to significant reductions in shoot growth rate, stomatal conductance, and photosynthetic rate, compared to control plants - The effect of half-drying was transient, as both gas exchange and shoot growth rate returned to pre-treatment levels within 1-2 weeks, despite one-half of the root system remaining dry - Alternating rootzone drying between the two halves of the root system prolonged the inhibition of shoot growth - Partial rootzone drying substantially reduced grapevine vegetative growth and water use without any detrimental effect on yield and with fruit quality improvements
Stoll (2000) (PhD Thesis) (includes: (Stoll et al., 2000a; Stoll et al., 2000b))	Australia	<u>Container-grown:</u> <i>Vitis vinifera</i> cv. Chardonnay, cv. Cabernet Sauvignon <u>Field-grown vines:</u> <i>Vitis vinifera</i> cv. Cabernet Sauvignon, cv. Syrah, cv. Riesling	- Container-grown and kept (greenhouse) - Field (various soil types)	- Control (vines receiving water on both sides) - PRD (vines only received water on one side at any time)	- Split-rooted vines, root system divided between two containers, two drip emitters per container - Split-rooted vines, half of the root system placed on either side of a plastic membrane buried in the field, with two drip emitters per vine or a "commercial PRD design", meaning a drip line, installed parallel to the vine row, on the ground, positioned 0.4 m away from the trunk of the vine on both sides. - Conventional vines with a non-divided root system with the same "commercial PRD design"	- Drip irrigation (and alternation of "wet" and "dry" sides) based on soil moisture, rainfall, and temperature - In the field experiments, PRD-treated vines received either the same amount or half the amount of water as the control vines - In container vines, water was withheld from one pot at any time in PRD, which led to reduced water amounts	- Chemical signals, primarily originating from roots and modified under PRD, play a crucial role in root-to-shoot communication in grapevines - The transfer of water from "wet" to "dry" soil layers may help maintain chemical signaling in grapevines responding to PRD and support the activity of fine roots in the drying soil - PRD sustained reduced vegetative growth, particularly of lateral shoots, which altered the canopy structure (better light penetration), improving fruit quality - The reduction in irrigation applied did not significantly reduce grape yield, leading to increased WUE - Application of relatively low irrigation rates showed that PRD-treated vines had a greater tolerance to water stress and probably utilized water more efficiently

de Souza et al. (2003)	Portugal	<i>Vitis vinifera</i> cv. Moscatel (field-grown)	- Field (soil derived from podzols, mostly sandy, that transitions to clay)	- NI (rain-fed, non-irrigated) - DI (50% ETc to both sides, 25% on each side) - PRD (50% ETc to one side, alternating sides each 15 days) - FI (100% ETc to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0,3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters every 15 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts (50%)	- PRD maintained a vine water status closer to FI, which was attributed to reduced stomatal conductance with no significant impact on carbon assimilation. As a result, there was an increase in intrinsic water use efficiency and long-term water use efficiency - Although PRD vines had reduced transpiration and stomatal conductance compared to DI vines, the differences between both treatments were subtle
dos Santos et al. (2003)	Portugal	<i>Vitis vinifera</i> cv. Moscatel, cv Castelão (field-grown)	- Field (soil derived from podzols, mostly sandy, that transitions to clay) - 240 – 320 vines	- NI (rain-fed, non-irrigated) - DI (50% ETc to both sides, 25% on each side) - PRD (50% ETc to one side, alternating sides each 15 days) - FI (100% ETc to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0,3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters every 15 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts (50%)	- Both PRD and DI treatments led to the same increase in water use efficiency compared to FI, due to the small or non-significant effects on yield - PRD led to a significant decrease in vigor and a slight improvement in fruit quality compared to FI. However, the differences between PRD and DI were not consistent, but PRD exhibited notably lower values for leaf layer number, canopy width, and the number of water shoots. This may have allowed for greater light interception in the cluster zone, which contributed to improvements in certain berry quality characteristics.
Gu et al. (2004)	USA	<i>Vitis vinifera</i> cv. Sauvignon Blanc	- Field (sandy loam soil) - 384 vines	- 0.4-CI (40% ETc to both sides) - 0.4-PRD (40% ETc to one side, alternating sides every 7-14 days) - 0.8-CI (80% ETc to both sides) - 0.8-PRD (80% ETc to one side, alternating sides every 7-14 days)	- Two drip lines suspended 0,45 m above ground, parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk - PRD switching was done by alternating the irrigating drip lines every 7-14 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received the same amount of water	- When comparing both treatments at the same irrigation level, vine water relations were only occasionally affected, and there was no significant improvement in vine performance and vine water use - It was the irrigation amount, rather than PRD, that was the key factor in inducing water stress, and decreasing transpiration and vegetative growth
Bravdo et al. (2004)	Israel	<i>Vitis vinifera</i> cv. Merlot	- Field (deep soil, heavy in clay and silica) - 300 vines	- REG-50% (0,12 ETp to both sides) - PRD-50% (0,12 ETp to one side, alternating sides every 10,12,14 days) - REG-100% (0,25 ETp to both sides) - PRD-100% (0,25 ETp to one side, alternating sides every 10,12,14 days) - REG-200% (0,4 ETp to both sides) - PRD-200% (0,4 ETp to one side, alternating sides every 10,12,14 days)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk, each spaced 0,75 m - PRD switching was done by alternating the irrigating drip lines every 10,12,14 days	- Irrigation scheduled according to a calculated ETp - PRD-treated vines received the same amount of water	- There were no significant differences between the PRD and REG strategies at the same irrigation level - Physiological and viticultural effects were evident only when comparing the different irrigation levels

Chalmers et al. (2004)	Australia	<i>Vitis vinifera</i> cv. Syrah	- Field (sandy loam soil) - 168 vines	- 100% Control (normal drip irrigation for commercial vineyards in the region, two drip lines) - 50% Control (single drip line, deficit of 50% of control) - 50% PRD¹⁴ (50% of control to one side, alternating sides every 14 days) - 50% PRD² (50% of control to one side, alternating sides every second irrigation event)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk - PRD switching was done by alternating the irrigating drip lines every 14 days or every second irrigation event	- Irrigation based on the typical amount for commercial vineyards in the region - PRD-treated vines received substantially reduced water amounts (50%)	- Both PRD treatments and the 50% control showed similar physiological responses - No PRD effect observed - Only the controlled level of irrigation (from 100% to 50%) improved water use efficiency and some grape quality parameters
Pudney and McCarthy (2004)	Australia	<i>Vitis vinifera</i> cv. Chardonnay	- Field (sandy loam over a medium red clay) - 135 vines	- F (net water application of 4L per hour through a single drip emitter) - FPRD (net water application of 4L per hour through a single emitter, alternating active emitter based on soil moisture) - S (net water application of 4L per hour through two emitters) - D (net water application of 2L per hour through a single emitter) - DPRD (net water application of 2L per hour through a single emitter, alternating active emitter based on soil moisture)	- F and D were fitted with one emitter per vine, 0,05m away from the vine trunk - FPRD, S, and DPRD had two emitters per vine, each on one side of the vine, spaced 0,5 m away from the vine trunk - All emitters were suspended approximately 0,25 m above ground - PRD switching was done by alternating active emitters based on soil moisture	- Irrigation scheduled according to the depletion of soil water in FPRD vines - PRD-treated vines had controls with the same amount of water	- Yield depended more on the volume of water applied than on the application method - No PRD effect was observed - No significant differences were shown between the PRD treatments and their counterparts - Water use efficiency improved similarly in both of the reduced water treatments
Baeza et al. (2005)	Spain	<i>Vitis vinifera</i> cv. Cabernet Sauvignon	- Field - 216 – 324 vines	- T1 (applied water at 0,45 of ETo to both sides) - T2 (applied water to both sides at 0,2 of ETo until veraison, 0,45 of ETo after) - T3 (PRD, applied water at 0,45 of ETo to one side, alternating sides based on soil moisture)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk - PRD switching was done by alternating the irrigating drip lines based on soil moisture	- Irrigation scheduling estimated using ETo data - PRD-treated vines received the same amount of water as the control	- No significant differences were observed in almost all parameters measured, and the three irrigation treatments did not significantly affect the agronomic or physiological responses of the grapevines - T2 demonstrated the highest water use efficiency, achieving a similar yield while using less water. This indicates that the reduced amount of water applied before veraison did not negatively impact yield.

de Souza et al. (2005a) (follow-up study to de Souza et al. 2003)	Portugal	<i>Vitis vinifera</i> cv. Moscatel (field-grown)	- Field (soil derived from podzols, mostly sandy, that transitions to clay)	- NI (rain-fed, non-irrigated) - DI (50% ETc to both sides, 25% on each side) - PRD (50% ETc to one side, alternating sides each 15 days) - FI (100% ETc to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0,3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters every 15 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts (50%)	- PRD almost always exhibited higher values of pre-dawn leaf water potential than DI - There were no significant differences between PRD and DI in all the berry parameters - Both deficit irrigation treatments (PRD and DI) didn't negatively affect the growth and quality of the berries, compared to the FI treatment
de Souza et al. (2005b)	Portugal	<i>Vitis vinifera</i> cv. Moscatel, cv. Castelão (field-grown)	- Field (soil derived from podzols, mostly sandy, that transitions to clay)	- NI (rain-fed, non-irrigated) - DI (50% ETc to both sides, 25% on each side) - PRD (50% ETc to one side, alternating sides each 15 days) - FI (100% ETc to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0,3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters every 15 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts (50%)	- The water status of PRD-treated vines was closer to FI than DI, particularly at midday, possibly suggesting stomatal conductance restriction by PRD. However, the differences between PRD and DI were small - PRD displayed restricted shoot growth in cv. Castelão - The reduced stomatal conductance seen in some PRD vines had no significant negative effect on carbon assimilation - Both PRD and DI, had no negative impact on CO₂ assimilation, resulting in improved water use efficiency due to the less water applied - The differences between the measured parameters of PRD and DI were subtle overall
de Souza et al. (2005c)	Portugal	<i>Vitis vinifera</i> cv. Moscatel, cv. Castelão (field-grown)	- Field (soil derived from podzols, mostly sandy, that transitions to clay)	- NI (rain-fed, non-irrigated) - DI (50% ETc to both sides, 25% on each side) - PRD (50% ETc to one side, alternating sides each 15 days) - FI (100% ETc to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0,3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters every 15 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts (50%)	- The deficit irrigation treatments (PRD and DI) significantly enhanced water use efficiency compared to FI. This is evident in both the short term, as intrinsic water use efficiency (A/g_{sw}) and in the long term, through the increase in ¹³C in plant tissues - The response to deficit irrigation was also influenced by grapevine variety and the specific environmental conditions of each year, with differences between treatments being more pronounced under drier conditions - In the dry year of 2002, PRD led to higher leaf water potentials than DI, possibly due to the reduced leaf area and greater midday stomatal closure, however, gas exchange measurements and the carbon isotope composition of berries revealed no statistically significant differences in WUE
Antolín et al. (2006)	Spain	<i>Vitis vinifera</i> cv. Tempranillo	- Container-grown and kept (greenhouse)	- C (well watered, both containers irrigated and maintained at "field capacity") - SDI (50% of the water given to C is applied, with half to each container) - PRD (50% of the water given to C is applied to only one container at a time, alternating containers)	- One drip emitter per container - Split-rooted vines, root system divided between two containers - PRD switching was based on a water potential threshold (anything below 0,2 MPa) monitored daily with a soil moisture sensor	- Irrigation scheduled according to container "field capacity" - PRD-treated vines received substantially reduced water amounts (50%)	- Both SDI and PRD showed significantly lower yield and cluster weight when compared to C (although PRD sustained closer values than SDI) - SDI and PRD had significantly different effects on vine performance. PRD led to higher yields, increased cluster weight, larger leaf area, greater berry weight, and higher skin anthocyanin levels than SDI - This better performance may be due to the different pattern of berry ABA concentration observed in PRD versus SDI, where ABA levels increased earlier and faster during veraison

Chaves et al. (2007)	Portugal	<i>Vitis vinifera</i> cv. Moscatel, cv. Castelão (field-grown)	- Field (soil derived from podzols, mostly sandy, that transitions to clay) - 320 vines	- NI (rain-fed, non-irrigated) - DI (50% ETc to both sides, 25% on each side) - PRD (50% ETc to one side, alternating sides each 15 days) - FI (100% ETc to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0.3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters every 15 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts (50%)	- PRD and DI displayed subtle physiological differences from each other, such as marginal reductions in stomatal aperture in PRD compared to DI, but PRD showed a clear reduction of vegetative growth. PRD vines maintained similar or better water status than DI vines, but growth inhibition occurred despite that, suggesting the involvement of a non-hydraulic mechanism in regulating growth - No significant differences in photosynthetic rates, chlorophyll fluorescence parameters, or water use efficiency were observed between PRD and DI - The reduced vigor in PRD compared to DI resulted in increased light interception in the cluster zone, potentially enhancing fruit quality. PRD vines had higher concentrations of anthocyanins and total phenols in berry skins, compared to DI and FI
De la Hera et al. (2007)	Spain	<i>Vitis vinifera</i> cv. Monastrell	- Field (deep clay loam soil) - 976 vines	- CI (30% ETc applied uniformly through the single drip line) - PRD (30% ETc applied to one side, alternating sides every 14 days)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk - PRD switching was done by alternating the irrigating drip lines every 14 days - PRD treatment applied at different phenological stages in every year	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received the same amount of water as the control	- Throughout the experiment, leaf water relations and gas exchange parameters were not significantly affected by the PRD treatment - Only in the 3rd year of the study, when PRD was applied from budburst to harvest, reproductive and vegetative development was distinctly altered in PRD vines - PRD primarily increased the fruit set percentage and leaf area development, both contributing to increased yield - Yield and water use efficiency improved significantly under PRD, without affecting berry weight, must and wine quality. This suggests enhanced synthesis and accumulation of photoassimilates and metabolites in the PRD berries
dos Santos et al. (2007)	Portugal	<i>Vitis vinifera</i> cv. Moscatel	- Field (soil derived from podzols, mostly sandy, that transitions to clay) - 720 – 960 vines	- NI (rain-fed, non-irrigated) - DI (50% ETc to both sides, 25% on each side) - PRD (50% ETc to one side, alternating sides each 15 days) - FI (100% ETc to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0.3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters after one month, and then after every 15 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts (50%)	- Of the three irrigated treatments, PRD vines exhibited an improved cluster zone microclimate, with higher incident photosynthetic photon flux density - PRD and NI showed higher berry temperatures when compared with DI and FI, a result of the more open canopies versus denser ones - The cluster microclimate of PRD vines was better than FI due to reduced vine vigor, with lower leaf layers, leaf area, canopy width, water shoots and shoot weight - PRD vines tended to develop a deeper root system, whereas DI and FI showed a more uniform root distribution across the soil layers - PRD enhanced berry quality by increasing the concentration of flavor precursors and total phenols. This occurred without significantly reducing yield, when compared to DI and FI

Poni et al. (2007)	Italy	<i>Vitis vinifera</i> cv. Sangiovese	- Container-grown and kept (open greenhouse) - 8 vines	- WW (well watered, both containers irrigated and maintained at "field capacity") - PRD (water applied (to "field capacity") to only one container at a time, alternating containers every 7 days)	- Two micro drip emitters per container - Split-rooted vines, root system divided between two containers - PRD switching was done by alternating active emitters every 7 days	- Irrigation scheduled according to container "field capacity" - PRD-treated vines received substantially reduced water amounts (50%)	- Both stem water potential and mid-day leaf water potential were significantly lower in PRD vines compared to WW - During the six-week stress period, PRD vines exhibited a 16% seasonal reduction in leaf CO₂ assimilation rate, relative to WW vines, less pronounced than the decreases in leaf stomatal conductance and transpiration, at 41% and 25%, respectively. As a result, intrinsic water use efficiency (A/g_{sw}) was significantly increased in the PRD vines - Shoot growth completely stopped when water was withdrawn from one of the pots in the PRD treatments, but PRD didn't significantly affect crop level and yield components - Most must quality parameters were not affected by water stress, but PRD must showed a higher concentration of soluble solids and total anthocyanins
Bindon et al. (2008)	Australia	<i>Vitis vinifera</i> cv. Cabernet Sauvignon	- Field (sandy loam soil) - 84 vines	- Control (well watered, irrigation applied uniformly through the single drip line) - PRD (60% of Control applied to one side, alternating sides every 7 days)	- Two drip lines parallel to the vine row, each emitter spaced 0,6 m apart - The spatial arrangement of the drip lines allowed for two consecutive emitters to be "on" while two consecutive emitters were "off", creating the "wet" and "dry" sides - PRD switching was based on soil water content (approximately every 7 days), and done by alternating drip lines	- Irrigation scheduled according to soil water content and essentially considered supplemental rainfall - PRD-treated vines received substantially reduced water amounts (40% deficit)	- PRD reduced berry weight compared to the Control, but had no effect on total anthocyanin concentration. Nonetheless, beginning at veraison, PRD altered the berry anthocyanin profile, increasing the proportion of non-malvidin anthocyanins. - Vines produced from PRD showed no significant differences in total monomeric anthocyanin concentration compared to the control. However, PRD wines exhibited a 15% increase in wine color density, total tannins, and polymeric pigments. The contribution of non-malvidin anthocyanins to total anthocyanins was significantly higher in PRD wines, reflecting the grape composition. - The researchers concluded that the PRD-induced changes in anthocyanin composition cannot be attributed to changes in cluster microclimate, but are a likely result of variations in the methylation step of anthocyanin synthesis
Rodrigues et al. (2008)	Portugal	<i>Vitis vinifera</i> cv. Moscatel, cv. Castelão	- Field (soil derived from podzols, mostly sandy, that transitions to clay) - 240 – 320 vines	- NI (rain-fed, non-irrigated) - DI (50% ET_c to both sides, 25% on each side) - PRD (50% ET_c to one side, alternating sides each 15 days) - FI (100% ET_c to both sides, 50% on each side)	- Two drip emitters per vine, installed parallel to the vine rows, positioned 0,3 m away from the vine trunk on both sides - PRD switching was done by alternating active emitters after one month, and then after every 15 days	- Irrigation scheduled according to a calculated ET_c - PRD-treated vines received substantially reduced water amounts (50%)	- PRD plants had a better (Castelão) or similar (Moscatel) water status compared with DI vines as assessed by pre-dawn leaf water potential and midday stem water potential - Partial stomata closure occurred in both PRD and DI plants - Data suggests that both hydraulic feedback and root-to-shoot chemical signaling mechanisms can be involved in the control of grapevine stomata apertures in response to decreased soil water availability; however, interestingly, hydraulic signals appear to play the dominant role

Du et al. (2008)	China	<i>Vitis vinifera</i> cv. Rizamat	- Field (light sandy loam soil) - 45 vines	- CDI (100%, conventional drip irrigation) - ADI (50%, alternated partial rootzone drying) - FDI (50%, fixed partial rootzone drying)	- Two drip emitters per vine, in line with the vine row, positioned 40 cm from the vine trunk - PRD switching was done by alternating active drip emitter according to soil moisture (FDI never switched dripper)	- Irrigation scheduled according to a calculated field capacity - PRD-treated vines received substantially reduced water amounts (50%)	- ADI maintained net photosynthesis, reduced transpiration, increasing instantaneous water use efficiency (A/E) - No significant diurnal changes in leaf water potential between treatments - ADI improved some aspects of grape quality without showing significant differences in yield
Intrigliolo and Castel (2009)	Spain	<i>Vitis vinifera</i> cv. Tempranillo	- Field (deep clay loam) - 3,240 vines	- NI (rainfed, no irrigation) - 50 (50% ET_c to both sides, 25% on each side) - 50-PRD (50% ET_c to one side) - 100 (100% ET_c to both sides, 50% on each side) - 100-PRD (100% ET_c to one side)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk - PRD switching was done every 14 days	- Irrigation scheduled according to a calculated ET_c - PRD-treated vines received the same amount of water as the control	- PRD vines did not significantly impact physiological parameters, growth, yield, or fruit and wine quality, compared to the same amount of water in conventional irrigation - Researchers report that it was the irrigation amount rather than the irrigation system installed that affected vine performance
de Souza et al. (2009)	Brazil	<i>Vitis vinifera</i> cv. Moscato Canelli, cv. Syrah	- Field (low fertility, deep, clay-rich) - 540 vines	- PRD (100% ET_c to only one side after fruit set) - DI (100% ET_c, withholding water after veraison)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk, emitters spaced 1,5 m from each other - PRD switching was done approximately every 24 days	- Irrigation scheduled according to a calculated ET_c - PRD-treated vines received the same amount of water as the control	- Water status and stomatal conductance suggested absence of water stress - No effects of PRD compared to DI - Variety and rootstock type affected vine performance
Romero et al. (2012a)	Spain	<i>Vitis vinifera</i> cv. Monastrell	- Field (fine clay, with a calcareous hard soil layer at 70 cm) - 650 vines	- Control (60% ET_c both sides, 30% on each side) - DI-1 (30% ET_c fruit set to harvest, 45% from harvest to leaf fall) - PRD-1 (same as DI-1 but to only one side) - DI-2 (15% ET_c from fruit set to harvest, 45% ET_c from harvest to leaf fall) - PRD-2 (same as DI-2 but to only one side)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk, emitters spaced 0,6 m from each other - PRD switching was done approximately every 14-16 days, 3rd year 6-8 days	- Irrigation scheduled according to a calculated ET_c - PRD-treated vines received substantially reduced water amounts	- Distinctive physiological effects of PRD (e.g., greater stomatal closure), compared with DI treatments receiving the same irrigation volumes, depended on the total soil water content available - Compared to DI-1, PRD-1 maintained higher leaf area post-veraison and increased root water uptake, whole-plant hydraulic conductance, leaf transpiration, stomatal conductance, and photosynthesis, but decreased intrinsic water use efficiency - Compared to DI-2, PRD-2 increased leaf xylem ABA concentration and decreased root water uptake, whole-plant hydraulic conductance, leaf transpiration, stomatal conductance, and photosynthesis

Balint and Reynolds (2014)	Canada	<i>Vitis vinifera</i> cv. Cabernet Sauvignon	- Field (clay loam) - 200 vines	- Control (rainfed, non-irrigated) - 100ET (100% ETc to both sides, 50% on each side) - PRD (100% ETc to one side, alternating each side) - 50ET (50% ETc to both sides, 25% on each side) - 25ET (25% ETc to both sides, 12,5% on each side)	- Two drip lines parallel to the vine row, suspended at 0,4 m, and 0,75 m apart from each other, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk, emitters spaced 1,2 m from each other - PRD switching not specified	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received the same amount of water as the control	- 100ET had higher soil moisture, leaf water potential, and transpiration rate than PRD - Soil and vine water impacted vine physiology, yield, and berry composition, but seasons showed inconsistent patterns
Romero et al. (2016b)	Spain	<i>Vitis vinifera</i> cv. Monastrell	- Field (fine clay, with a calcareous hard soil layer at 70 cm) - 650 vines	<u>Seven-year research, several different ETc %</u> - Control (60% ETc to both sides, 30% on each, dropping to 40%) - DI-1 (Moderate DI, various %ETc) - PRD-1 (Severe PRD, various %ETc) - DI-2 (Severe DI, various %ETc) - PRD-2 (Moderate PRD, various %ETc)	- Two drip lines parallel to the vine row, with one emitter on one side of the vine's trunk and a second emitter (in the other drip line) on the other side of the vine's trunk, emitters spaced 0,6 m from each other - PRD switching was done approximately every 14-16 days, 3rd year 6-8 days - PRD switching was done almost always every 14-16 days	- Irrigation scheduled according to a calculated ETc - PRD-treated vines received substantially reduced water amounts	- Among the DI treatments, the strategy involving moderate water stress, PRD-1 was the one which most improved the yield, WUE, and berry anthocyanin content - PRD-2 and RDI-2 were the most restrictive strategies and provided limited improvements in WUE or berry quality - Significant interactive effects of irrigation volume (high vs. low) and irrigation placement (PRD vs. RDI) indicate distinct PRD effects on long-term yield, WUE, and berry quality, depending on the volume of water applied in the wet rootzone and the soil total water availability

Notes: Only trials that provided detailed information on the PRD strategy used, the amount of water applied and the effects of the PRD treatments on physiological and crop/yield related aspects were selected for this revision. Multiple literature searches of peer-reviewed published literature were conducted, conference proceedings and doctoral dissertations were also included in the revision due to overwhelming importance on the subject. A variety of search terms were used in multiple combinations across Clarivate's Web of Science™ Core Collection, PubMed (maintained by the National Center for Biotechnology Information) and Google Scholar: partial rootzone drying/grapevine, partial rootzone drying/viticulture, partial rootzone irrigation/grape, partial irrigation of the rootzone, PRD irrigation, PRI irrigation, deficit irrigation/viticulture, grapevine water use, PRD effects/impacts, irrigation strategies/grapevine, irrigation water, PRD *Vitis vinifera*; among other synonyms. The last comprehensive search was on May 2024. Most studies relevant to the revision were identified during the literature searches. However, some were inaccessible/unavailable due to journal subscription barriers or lack of access to full-text versions, mostly in pre-2000's articles.

2. Material and Methods

2.1 Vineyard Site Characteristics

The experiment was conducted in 2023 in a commercial vineyard within the Lodi AVA in California, USA (38°17'43"N, 121°06'17"W). The whole vineyard had a total size of 43,06 ha and the portion in which the experiment was conducted was approximately 3 ha. The experimental plot presented some topography, being imbedded in a small hill with a mild incline running east to west, with an elevation of 52 m at the highest point and 43 m at the lowest, averaging at 48-49 m overall (U.S. Geological Survey, 2023).

The climate of the region is classified as a hot-summer Mediterranean climate (Csa), according to the Köppen-Geiger climate classification (Beck et al., 2018), with warm, dry summers and cool, wet winters with an historical (30 year) average annual temperature of 16,3 °C and historical average annual rainfall of 482,7 mm (PRISM Climate Group, 2024).

The soil was classified as being mostly (85%) Corning series, with a yellowish red color. It was a sandy loamy (increasingly clay loamy texture with depth) moderately well-drained soil, with slow permeability and high runoff potential (Soil Survey Staff, 2024; Walkinshaw, 2022).

The cultivar of *Vitis vinifera* L. studied was Cabernet Sauvignon Selection 08 (FPS 08) grafted onto Selection Oppenheim #4 (SO4, *V. berlandieri* Rösséguier x *V. riparia*) rootstock.

The vineyard was planted in 2014 and trained as a Horizontally Divided Double Curtain (also called GDC type or Wye trellis) in a quadrilateral cordon configuration, with four permanent cordons from a single vine, two on each side of the canopy, at a height of 1,6 m, and 1 m between curtains. Vine spacing was 1,52 m and row spacing 3,35 m, the total vine density was 1965 vines/ha. Row orientation was east to west.

Vines were mechanically spur pruned for the second year in a row (in previous years they were hand-pruned to two bud spurs) and managed according to conventional growing practices of the region. These growing practices (fertilization, leaf removal, shoot trimming, pruning, mowing) were applied uniformly across the block. The occurrence of the phenological stages and viticultural practices in the 2023 season is reported in Table 7, Appendix A.

A conventional drip irrigation system was initially set up in 2014, and it was retrofitted for the experiment in 2022 with a Variable Rate Irrigation (VRI) system designed by Verdi.ag (Verdi Expeditions Inc., Vancouver, BC, Canada). This VRI system provides precise control over smaller sections of the block (so called “pixels”) through the Verdi Micro-Block controllers, and allows for remotely scheduled irrigation through a web browser dashboard.

2.2 Experimental Design, Irrigation and Soil Moisture

The experimental design (Figure 1A) consisted of four treatments with six replicates per treatment, installed in a standard randomized block design. Each replicate plot consisted of three rows of 20 vines (60 vines in total) allocated to sampling as exemplified in Figure 1B.

Four irrigation treatments were applied as described in Figure 2.

1) Control irrigation (CTRL), which was conventional drip irrigation located above ground on a support wire at 0,45 m from the ground, representing the standard irrigation practice for growers in the region. Water was applied with a Netafim UniRam™ AS pressure compensated drip line (Netafim Ltd., Tel Aviv, Israel) per row, with the following specifications: 18 mm inside diameter, 1,9 L/h emitter flow rate, 0,53 m spacing between emitters.

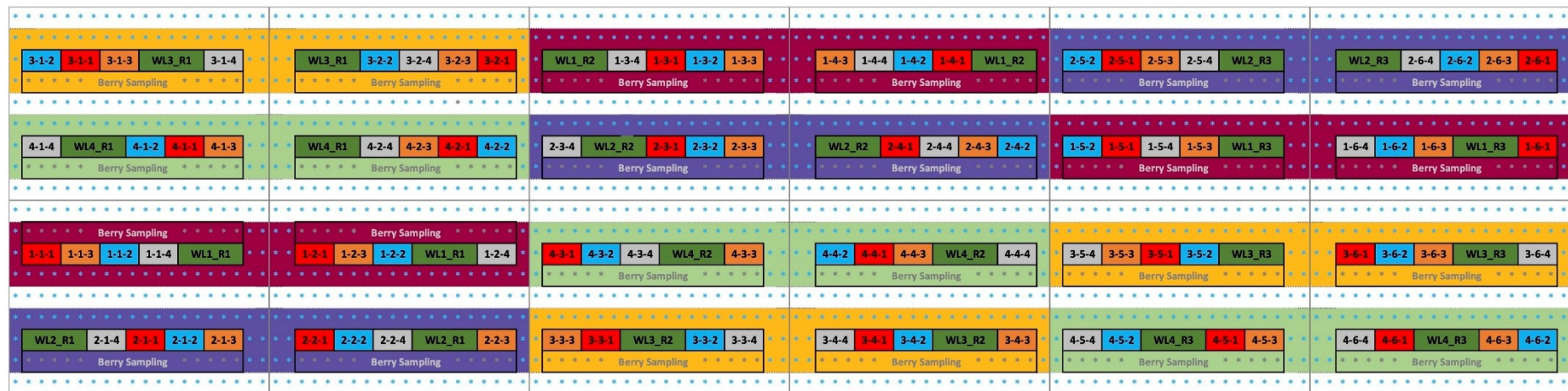
2) Subsurface irrigation (SSI), where water was applied directly under the vines at a depth of 0,3 m. This was achieved by a custom-made system that simulated a subsurface line but avoided disrupting the root system (Figure 2). The conventional drip line was replaced with a blind hose, and water was deviated underground using polyethylene tubes with a diameter of 6,4 mm connected to emitters at equal spacings and nominal flow rate as the control. These emitters were installed inside a PVC tube (0,022 m outside diameter and 0,3 m length) perforated at the bottom for water to permeate the soil at the defined depth.

3) Partial Rootzone Drying Alternated (PRD1), in which one Netafim UniRam™ XRS copper embedded pressure compensating drip line (20 mm inside diameter, 2,3 L/h emitter flow rate, 0,76 m spacing between emitters) was buried in the middle of each side of the row, called the interrow, at a depth of 0,3 m. Trenching and subsurface line installation occurred in May 2022. In PRD1, the interrows were irrigated in an alternated fashion, based on soil moisture readings, where 75 cbar was deemed dry enough to cause a switch.

4) Partial Rootzone Drying One-Side (PRD2), with a similar configuration to PRD1, but only one of the vine sides was maintained irrigated during the trial.

All treatments were irrigated at 80% crop evapotranspiration (ET_c), estimated as the product of reference evapotranspiration (ET_o) and crop coefficient (K_c). K_c values were estimated using a mixed method of Normalized Difference Vegetation Index (NDVI) and Fractional Cover (FC) values, where a model developed in-house was trained to calculate K_c with FC as a focal point and look at the correlations between NDVI and FC. The models were validated using Leaf Area Index (LAI) as points for ground truthing. Reference evapotranspiration values were collected from the on-site meteorological station. Irrigation started on June 27 (DOY 178) and ended September 23 (DOY 266). Daily irrigation was tracked through flowmeters (Saier Sensor Co. Ltd, Ronggui Town, China), and soil moisture values were recorded using IRROMETER WATERMARK 200SS sensors (Irrometer Company, Inc., Riverside, CA, USA). For two replicates (one on top of the hill and one on the bottom of the hill) of each treatment, six soil moisture sensors were installed to capture soil moisture at two depths (30 and 90 cm) and three locations, namely under the vine and in the middle of the interrow on each side of the vine. Flowmeter and soil moisture readings were collected every 15 minutes.

A



B

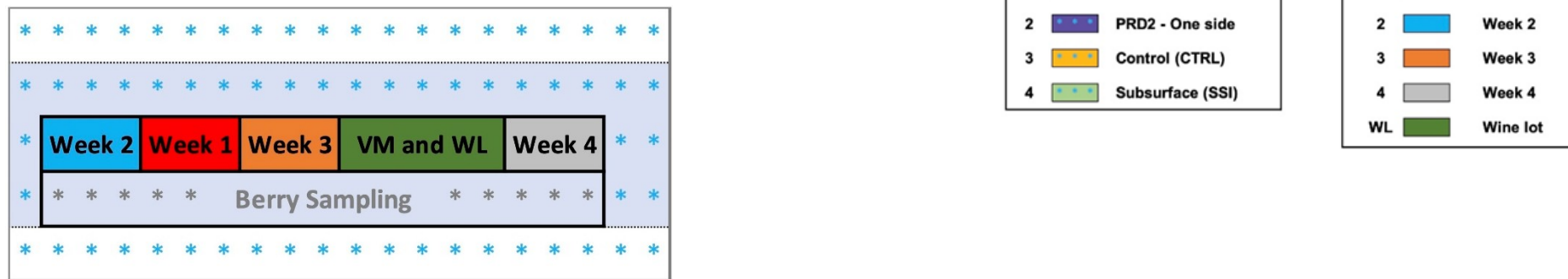


Figure 1 – Experimental design of the 2023 Cabernet Sauvignon trial:

1A) Trial layout representing the location of the 60-vine plots for each treatment and replicate (n = 6). Colors differentiate between treatments – CTRL (standard drip irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Uncolored rows represent vines left as buffer. The tri-codes represent treatment-replicate-week for weekly fruit sampling for chemical analysis.

1B) Example of the vine allocation within each replicate plot. In the middle row, four three-vine panels (Week 1-4) were allocated to weekly fruit sampling for chemical analysis and a five-vine panel (VM and WL) was used for vine measurements, yield components, research winemaking (data not shown), chemical analysis at harvest and pruning weights. The order of vine panels in the middle row was fully randomized. Berry sampling encompasses 17 vines from which berry maturity monitoring samples were collected. All rows were used for remote sensing procedures.

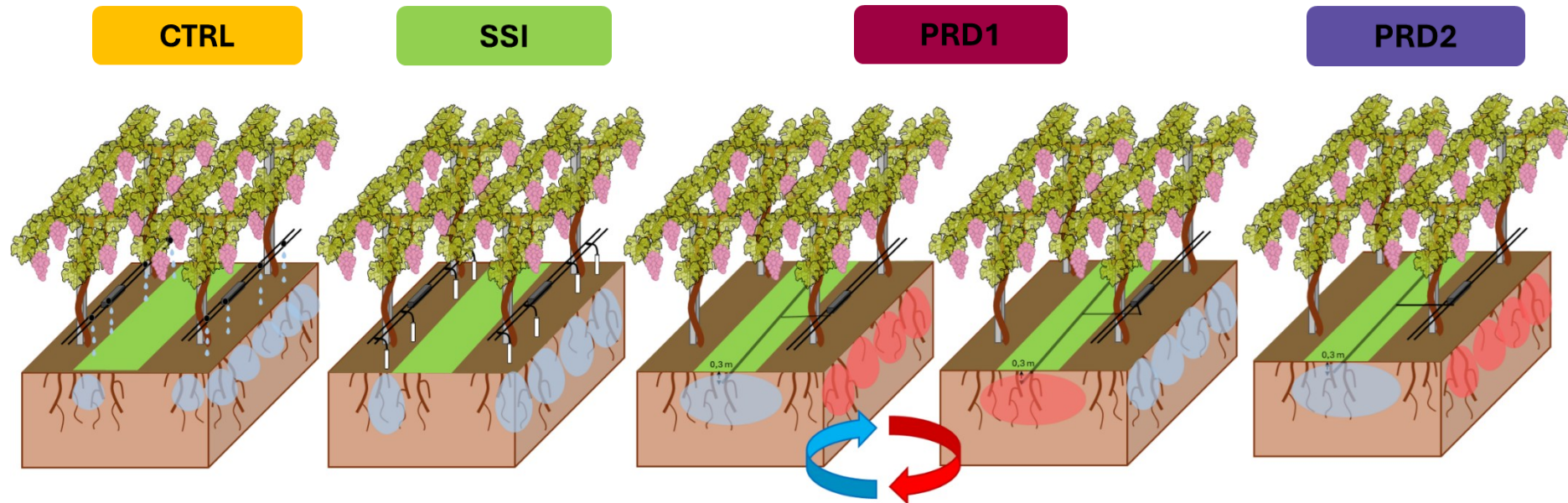


Figure 2 – Schematic diagram of the irrigation treatments applied in the 2023 Cabernet Sauvignon trial:

- 1) Control irrigation (CTRL), which was conventional drip irrigation located above ground on a support wire at 0,45 m from the ground, representing the standard irrigation practice for growers in the region.
- 2) Subsurface irrigation (SSI), where water was applied directly under the vines at a depth of 0,3 m. This was achieved by a custom-made system that simulated a subsurface line but avoided disrupting the root system.
- 3) Partial Rootzone Drying Alternated (PRD1), in which one Netafim UniRam™XRS copper embedded pressure compensating drip line was buried in the middle of each side of the row, called the interrow, at a depth of 0,3 m. The interrows were irrigated alternately based on soil moisture readings, where 75 cbar was deemed dry enough to cause a switch.
- 4) Partial Rootzone Drying One-Side (PRD2), in which one Netafim UniRam™XRS copper embedded pressure compensating drip line was buried in the middle of each side of the row, called the interrow, at a depth of 0,3 m. Only one of the vine sides (one of the interrows) was maintained irrigated during the trial.

2.3 Weather Data

Local weather data for the Borden Hills weather station (38°17'36"N, 121°06'22"W) (approx. 250 m from the experimental site) was retrieved from the Lodi Winegrape Commission Weather database (Lodi Winegrape Commission, 2024). Daily maximum, minimum and average temperature (°C), relative humidity (%), dew point (°C), wind speed (kph), precipitation (mm) and reference evapotranspiration (ET_o) (mm) for the 2023 season were used.

Historical daily data (1993-2023) of precipitation (mm) and temperature (°C) were obtained from the AN81d/AN91d dataset, at the PRISM Climate Group database (PRISM Climate Group, 2024). Historical and Seasonal Daily Growing Degree Days (GDDs) were calculated during the growing season (April 1 – DOY 91 to October 31 – DOY 304) using the following formula:

$$GDD (^{\circ}C) = \frac{(T_{max} + T_{min})}{2} - T_{base} \quad (1)$$

where $T_{base} = 10^{\circ}C$.

2.4 Vine Measurements

Vine measurements of water status and gas exchange were collected (in tandem) on, at least, two leaves in two vines per replicate plot, on a dedicated set of five vines (VM and WL – Figure 1B), later used for yield components, research winemaking and pruning weights. Vine and replicate selection were randomized throughout the season to characterize treatment effects over a broader range of vines and to prevent excessive vine defoliation.

Measurements started June 29 (DOY 180) and ended September 18 (DOY 261). They were conducted multiple days per week at midday (13:00-14:00 PDT) to monitor treatment effects. In parallel, diurnal measurements were taken on a weekly basis, by repeating measurements in three rounds of one hour each (10:00-11:00, 13:00-14:00 and 16:00-17:00 PDT) to characterize the daily variations of each parameter.

2.4.1 Plant Water Status

○ Leaf Temperature

Leaf temperature (°C) was measured with a MI-210 Handheld Infrared Radiometer (Apogee Instruments, Inc., Logan, UT, USA). Measurements were taken approximately 2-3 cm away from the leaf, in a sunny exposed section, making sure not to create shade or angle the radiometer incorrectly.

○ Leaf and Stem Water Potential (Ψ_{leaf} , Ψ_{stem})

Ψ_{leaf} (MPa) and Ψ_{stem} (MPa) measurements were taken using a 615 PMS Pressure Chamber Instrument (PMS Instrument Company, Albany, OR, USA). For Ψ_{leaf} , leaves were wrapped in a clear plastic bag, cut at the petiole, and immediately inserted in the pressure chamber. The pressure was gradually raised until the initial droplet of xylem sap appeared on the cut surface, at which point the chamber pressure was recorded. For Ψ_{stem} determination, leaves were enclosed in aluminum foil zip-lock bags

for 20-30 minutes before measurements, to stop transpiration and allowing for their water potential to balance with the stems. After that period, leaves were cut at the petiole and processed with the pressure chamber as fast as possible, in the same way as Ψ_{leaf} .

2.4.2 Leaf Gas Exchange

Net CO₂ assimilation (A ; $\mu\text{mol}/\text{m}^2/\text{s}$), stomatal conductance to water vapor (g_{sw} ; $\text{mol}/\text{m}^2/\text{s}$) and transpiration rate (E ; $\text{mol}/\text{m}^2/\text{s}$) were measured using the LI-6800 Portable Photosynthesis System (Li-COR Inc., Lincoln, NE, USA), with the 6800-01A 6,0 cm² broadleaf fluorometer chamber. Standard manufacturer protocol was adhered to during measurements, and environmental parameters reflected ambient conditions at the time of sampling. g_{sw} was also measured using the LI-600 Porometer/Fluorometer (Li-COR Inc., Lincoln, NE, USA). Due to the portability and ease of use offered by the LI-600, more g_{sw} measurements were taken compared to the LI-6800.

2.5 Yield Components

At harvest, yield components were taken from one datavine from each treatment replicate (VM and WL – Figure 1B). The treatments were harvested when TSS reached 25.0 °Brix. Due to treatments having different rates of ripening, the CTRL and SSI treatments were harvested a week earlier than PRD1 and PRD2 (CTRL, SSI – September 21 (DOY 264); PRD1, PRD2 – September 27 (DOY 270)). Vine yield (kg), clusters per vine and average cluster weight (g) were determined for all treatments.

2.6 Water Use Efficiency

The agronomic water use efficiency at the crop level (WUE_C) was calculated and expressed as the ratio between yield and the amount of water supplied (ton/ML). A and g_{sw} were used to calculate the intrinsic water use efficiency ($WUE_i = A/g_{sw}$) for treatment comparisons.

2.7 Vine Size/Vine Vigor

2.7.1 Normalized Difference Vegetation Index and Fractional Cover

High-resolution (5 cm/pixel) multi-spectral imagery was collected using an unmanned aircraft system (UAS) on a monthly basis for the entire trial. Fractional cover (FC), Normalized Difference Vegetation Index (NDVI) and Modified Chlorophyll Absorption Reflectance Index (MCARI) were calculated for each treatment and replicate plot (60 vines each). NDVI and MCARI were calculated using Equation 2 and 3.

$$NDVI = \frac{NIR - Red}{NIR + Red} \quad (2)$$

$$MCARI = ((RedEdge - Red) - 0,2 * (RedEdge - Green)) * (\frac{RedEdge}{Red}) \quad (3)$$

NDVI was used to separate green pixels from the background by applying a threshold. Then, vine pixels were separated from the green pixels based on MCARI, where pixels with values larger than 0,3 were considered vine pixels. Studies have shown (Basso et al., 2010) that the red edge band in MCARI effectively distinguishes cover crops, resulting in a more accurate classification of vine pixels versus green pixels in the background (e.g., grass). Ultimately, FC was calculated based on the number of vine pixels over the total count of pixels in each replicate.

2.7.2 Pruning Weights

Pruning weights (PWs) and the number of shoots per vine were recorded during vine dormancy on one datavine per treatment replicate (VM and WL – Figure 1B), the same one that was previously harvested to calculate yield components. The vines were hand-pruned back to two-bud spurs, the pruned canes were counted, weighed, and transported to the laboratory. Crop load was calculated using the yield to pruning weight ratio (Ravaz Index), dividing yield and pruning weight values in kg.

2.8 Berry Maturity Monitoring

From veraison to harvest (August 2, DOY 214 – September 27, DOY 270), one 100-berry sample was collected weekly from a flagged portion (17 vines) of each treatment replicate (Figure 1B). This berry sample was composed from groups of five berries, from various positions within the cluster, randomly collected across the 17 vines. The samples were stored in ice and transported to the laboratory, where the berries were weighed, counted to determine average berry weight, and subsequently crushed, using the juice to determine Total Soluble Solids (°Brix) with an ATAGO PAL-1 refractometer (ATAGO CO., Tokyo, Japan). The basic chemistry of the juice (Total Soluble Solids (°Brix), pH, titratable acidity (TA; g/L), malic acid (g/L), tartaric acid (g/L), nitrogen by o-phthaldialdehyde (NOPA; mg/L), and yeast assimilable nitrogen (YAN; mg/L)) was evaluated according to the methodology reported for cluster samples below (Grape Chemical Composition).

2.9 Grape Chemical Composition

Sequential sampling was conducted on a weekly basis starting at approximately 20 °Brix. Within each replicate plot, in the middle row (composed by 20 vines), four sets comprising three vines each were established and randomly assigned to a maturity level (Week 1-4 – Figure 1B). Upon reaching the target level, a cluster sample from each replicate was collected. This cluster sample was composed by randomly picked clusters from the three-vine set, representing various cluster positions and exposures, until half of the sample bag was filled (20-25 clusters). A composite sample was collected from each research winemaking lot at harvest, to better represent the composition of resulting wines. The cluster samples were weighed and stored on ice during transportation to the laboratory. Subsequently, the samples were stored in a cold room and analyzed within two days of collection. The grapes were destemmed and homogenized using a Vitamix blender (Vitamix Corporation, Cleveland, OH, USA).

Details of the grape analysis were previously described in Previtali et al. (2021), and a brief summary of the main aspects relevant to this research is reported below.

To determine basic biochemical parameters, a WineScan FT-120 Fourier transform infrared (FTIR) spectroscopy system (FOSS North America, MN, USA) was used for Total Soluble Solids (TSS; °Brix), pH, berry moisture content (%), titratable acidity (TA; g/L), malic acid (g/L), tartaric acid (g/L), nitrogen by *o*-phthaldialdehyde (NOPA; mg/L), and yeast assimilable nitrogen (YAN; mg/L). FTIR calibration functions for all the parameters were developed using standard primary analytical methodologies.

Free volatile compounds (3-isobutyl-2-methoxypyrazine (IBMP; ng/L), β -damascenone ($\mu\text{g/L}$), C_6 Compounds ($\mu\text{g/L}$), 1-octen-3-ol ($\mu\text{g/L}$)) were analyzed by headspace solid-phase micro-extraction gas chromatography-mass spectrometry (HS-SPME-GC-MS), using an Agilent 7890B gas chromatograph equipped with a multimode inlet coupled to a 5977B mass selective detector (MSD) (Agilent Technologies Inc., Santa Clara, CA), following standard procedures with some modifications.

The bound-form of β -damascenone ($\mu\text{g/L}$) was analyzed indirectly by isolating the glycosides using solid-phase extraction (with a Hamilton STAR robotic liquid handler (Hamilton Robotics, Reno, NV)), followed by acid hydrolysis and analysis of the released volatiles in a similar GC-MS system as described above.

The composition of phenolic compounds (total anthocyanins (mg/g berry), quercetin glycosides (mg/L), polymeric tannins (mg/L)) was assessed using an Agilent 1200 reversed-phase high-performance liquid chromatography (HPLC) system.

2.10 Statistical Analysis

All data were registered in Microsoft Excel (Microsoft Corporation, Redmond, WA, USA) version 2420 and analysis was conducted using SPSS (IBM, Armonk, NY, USA) version 29. Treatment effects on the physiological, growth and biochemical parameters were analyzed using a linear mixed model with one fixed effect (treatment) and one random factor (replicate). Means and standard errors (SE) were calculated, and significant interactions were separated using Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).

Charts and figures were constructed using Microsoft Excel and Microsoft PowerPoint, soil moisture summary plot was created with R Statistical Software version 4.3.1 (R Core Team, 2024) in RStudio (RStudio Team, 2024) using the "ggplot2" package. Spatial maps of fractional cover were also produced with RStudio using the "raster" and "sf" packages.

3. Results

3.1 Weather data

The local weather data retrieved from an on-site weather station is shown in Figure 3. The 2023 season was characterized by an average air temperature of 15,9 °C, with the average minimum temperature in the coldest month, February, dropping to 2,6 °C and the average maximum temperature in the warmest month, July, being 35,7 °C. Annual precipitation was 531 mm, with 422 mm falling from January to March, turning into an unexpectedly rainy winter. The Cumulative Growing Degree Days (GDDs) in the growing season were 2302, and the seasonal evaporative demand (ET_o) was 1072 mm (yearly 1335 mm).

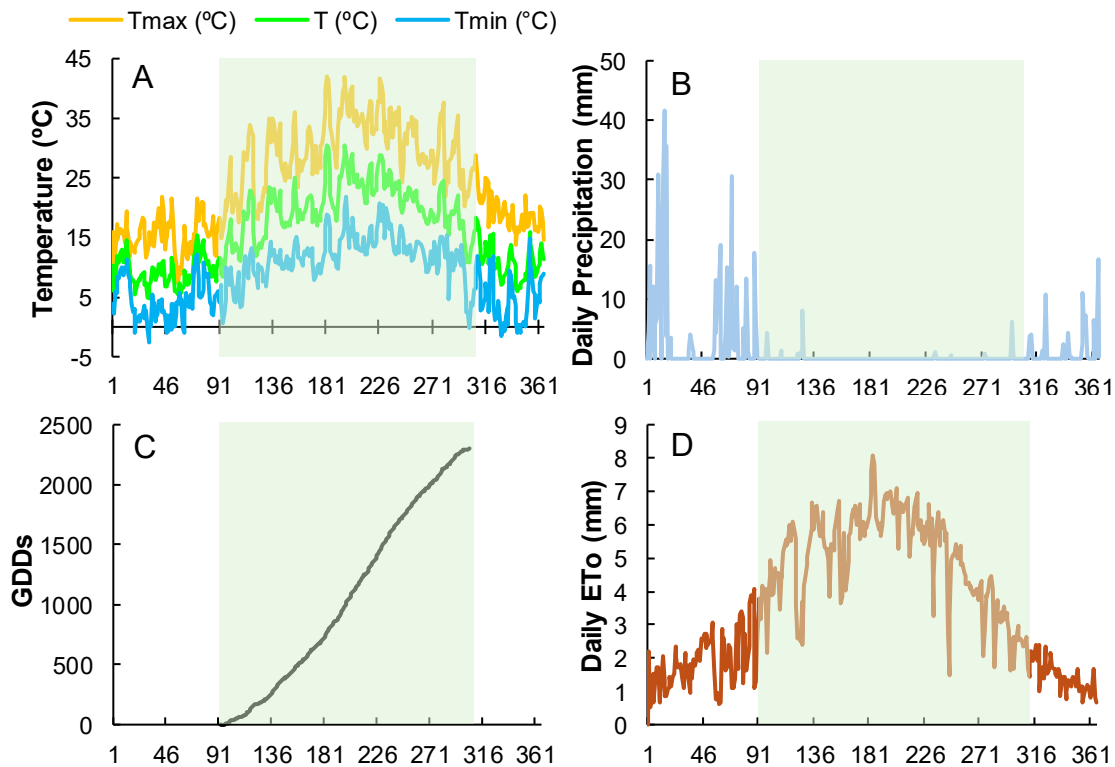


Figure 3 – Local weather data for the 2023 season expressed by day of the year (DOY). The growing season (April 1-DOY 91 to October 31-DOY 304) is highlighted in green.

3A) Daily temperature (°C) (maximum, average, minimum); **3B)** daily precipitation (mm); **3C)** cumulative growing degree days; **3D)** daily reference evapotranspiration.

Historical (1993-2023) monthly precipitation and mean air temperature of the experimental site, versus the 2023 season (both from PRISM), are represented in Figure 4. The historical average annual air temperature was 16,3 °C, with 3,5 °C as the average minimum temperature of the coldest month and 33,8 °C the average maximum temperature in the warmest month. Precipitation averaged 483 mm per year, concentrated in the fall and winter months, with typically dry summers.

The 2023 air temperature deviated from the long-term average, displaying a cooler winter and spring. Precipitation in 2023 was abnormally higher than the average in the winter. Compared to the previous five seasons and the historical average

(Appendix B, Figure 15), GDDs were lower in 2023, indicating that this was a cooler season overall.

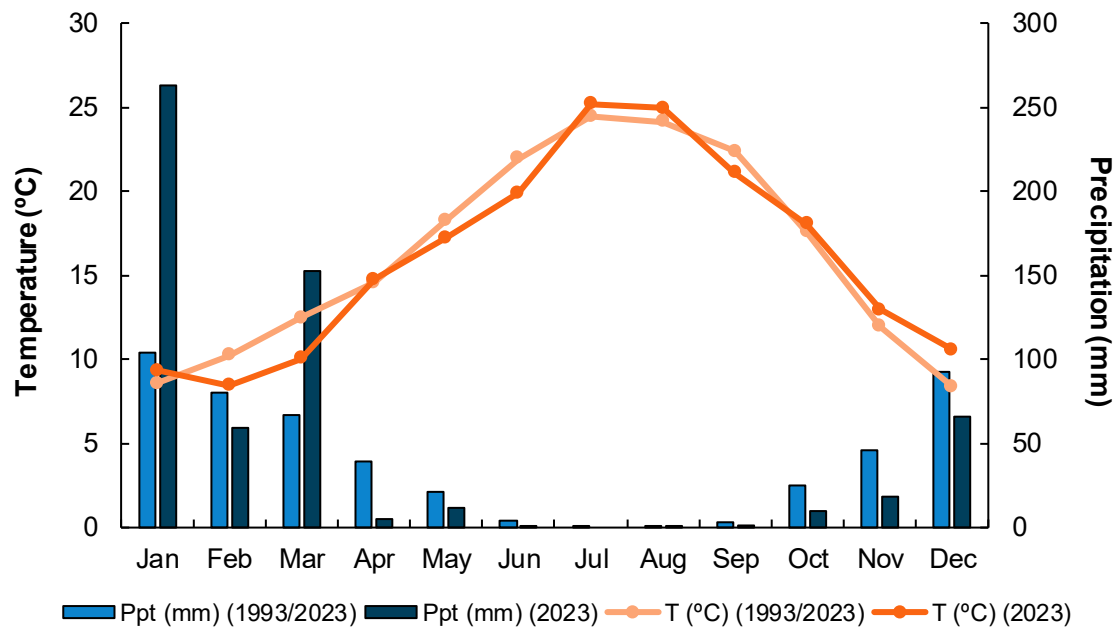


Figure 4 – Comparison of total monthly precipitation and average air temperature at the experimental site during the 2023 season and the past 30 years (1993-2023). Data sourced from PRISM Climate Group.

3.2 Irrigation and Soil Moisture

The irrigation amount supplied each treatment is displayed in Table 2 and the cumulative irrigation throughout the season in Figure 16, Appendix C.

Overall, there were no big differences between treatments. PRD1 R5&R6 had a jump in irrigation in the earlier stages of the trial due to a faulty valve opening, but it was resolved, and the amount was corrected. SSI showed slightly higher values due to water backflow and runoff, which required adjustments.

Table 2 – Total irrigation amounts supplied to Cabernet Sauvignon vines under different irrigation practices in 2023.

	CTRL	SSI	PRD1	PRD2
Irrigation amount (ML/ha)	2,54 ± 0,05	2,91 ± 0,08	2,82 ± 0,23	2,47 ± 0,03

Notes: Data are presented as means ± SE (n = 3) of flowmeter readings for each treatment: CTRL (control irrigation); SSI (subsurface irrigation); PRD1 (partial rootzone drying alternated); PRD2 (partial rootzone drying one-side).

Soil moisture trends in response to irrigation events over the course of the trial are represented in Figure 5.

CTRL exhibited a conventional drip irrigation pattern, with the readings of the middle (under the vine) sensor indicating that the water was being applied correctly. In a similar fashion, SSI also showed the intended constant moisture level under the vines. When comparing CTRL and SSI patterns under the vine row, CTRL readings displayed a more marked diurnal drying effect compared to SSI. As for the interrow sensors, readings showed equally dry conditions in both CTRL and SSI.

On PRD1, the north (row space on the north side) and south (row space on the south side) sensors moisture readings conveyed alternated wet and dry cycles between the interrow spaces, occurring close to the intended 75-100 cbar threshold. PRD2 showed that the north side was being correctly irrigated and maintained moisture throughout the trial, while the south side was kept dry and non-irrigated.

There were differences in the soil drying patterns of Rep 1 and Rep 2. Rep 1 (located on the top of the hill) showed a faster and more uniform drying pattern, really evident in PRD1, while Rep 2 (located on the bottom of the hill) had slower permeability and held more water, drying in a different manner in every treatment.

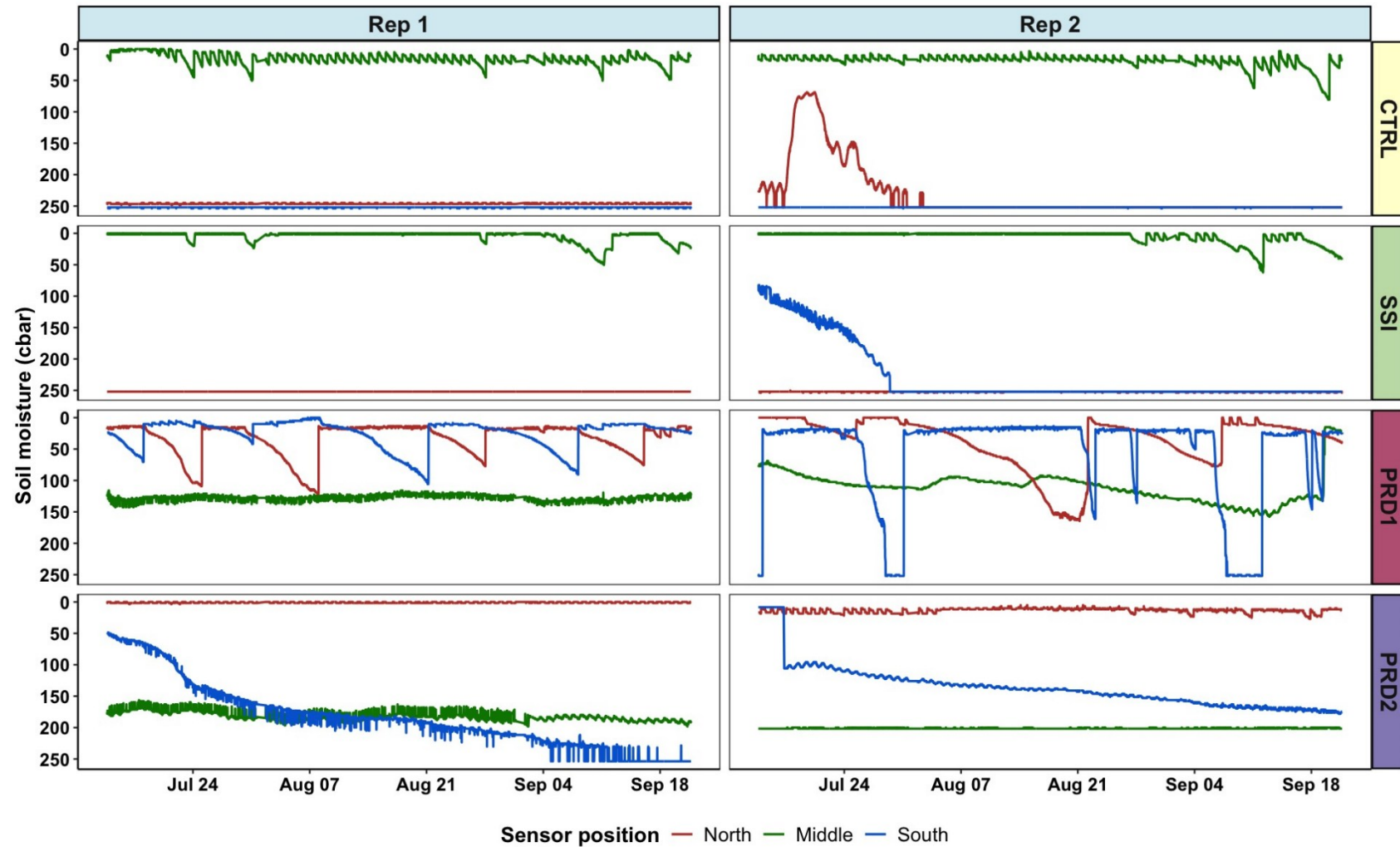


Figure 5 – Temporal trends of soil moisture in response to irrigation events in Cabernet Sauvignon vines under different irrigation regimes in 2023: CTRL (control irrigation); SSI (subsurface irrigation); PRD1 (partial rootzone drying alternated); PRD2 (partial rootzone drying one-side). Colors represent sensor positions in relation to the vine: North (row space on the north side), Middle (under the vine) and South (row space on the south side). Sensor depth was 30 cm. Data are shown for two replicates of each treatment. Irrigation started June 27 (DOY 178) and ended September 23 (DOY 266).

3.3 Vine Measurements

Seasonal changes in vine performance measurements for the four treatments are shown in Figure 6.

Midday ψ_{stem} and midday ψ_{leaf} trends paralleled each other. Overall, ψ_{leaf} (Figure 6D) varied between -0,8 MPa and -1,45 MPa throughout the season, with PRD2 trending for the lowest values until DOY 240, where a sharp increase was shown. SSI displayed a narrower range of ψ_{leaf} values when compared to CTRL, -1,28 to -0,79 MPa and -1,38 to -0,84 MPa. Both PRD1 and PRD2 showed similar ψ_{leaf} ranges -1,42 to -0,88 MPa and -1,46 to -0,84 MPa, respectively.

Midday ψ_{stem} (Figure 6E) varied between -0,44 MPa and -1,03 MPa throughout the season, and all treatments showed approximately the same trends as ψ_{leaf} . CTRL and SSI ψ_{stem} values displayed similar ranges, from -0,88 to -0,53 MPa and -0,81 to -0,53 MPa, respectively. PRD1 and PRD2 showed a wider scope of values when compared to CTRL and SSI, with PRD2 fluctuating from -1,03 to -0,47 MPa and PRD1 exhibiting a narrower range of -0,89 to -0,44 MPa.

Leaf temperature variations throughout the season were fairly consistent between treatments (Figure 6B). PRD2 showed slightly higher values most of the sampling dates and the highest values overall (38,9 °C). CTRL, SSI and PRD1 varied similarly over time.

Midday g_{sw} (Figure 6C) varied in the range of 0,1 mol/m²/s to 0,5 mol/m²/s. CTRL and SSI trended for higher values than PRD1 and PRD2 in the beginning of the season, but gradually shifted to having the lowest towards the later stages. CTRL and SSI exhibited a broader range of values (0,468 - 0,103 mol/m²/s and 0,488 - 0,159 mol/m²/s) than PRD1 and PRD2 (0,398 - 0,167 mol/m²/s and 0,351 - 0,131 mol/m²/s), generally showing more variability.

Overall, there were significant differences between treatments in all parameters; however, the treatment separation/differentiation was specific to the sampling date.

The relationship between gas exchange measurements and plant water status is shown in Figure 7, based on g_{sw} and ψ_{leaf} data collected at three different timepoints of the day. Overall, the g_{sw} and ψ_{leaf} were linearly correlated, with decreases in g_{sw} corresponding to more negative ψ_{leaf} (i.e. more water stress).

In round 1 (10:00-11:00), all treatments exhibited a significant linear correlation between variables except PRD1 ($y = 0,02x + 0,33$; $R^2 = 0,03$; $p = 0,508$), where g_{sw} was maintained constant even at lower levels of ψ_{leaf} . Additionally, there were differences in the respective ranges of g_{sw} and ψ_{leaf} measured in the four treatments. PRD2 displayed the biggest range, with the lowest ψ_{leaf} values and equally low g_{sw} values (i.e., -1,46 MPa corresponding to 0,090 mol/m²/s). Regarding round 2 (13:00-14:00), CTRL and SSI display a smaller range of values when compared to PRD1 and PRD2, not showing a linear correlation between variables ($y = 0,10x + 0,32$; $R^2 = 0,02$; $p = 0,544$ and $y = 0,12x + 0,36$; $R^2 = 0,07$; $p = 0,276$). PRD2 exhibited the same behavior as in round 1, reaching the lowest g_{sw} and ψ_{leaf} . In round 3 (16:00-17:00), all treatments behaved similarly; PRD1 and PRD2 revealed again a broader range of values, with PRD2 showing the lowest. CTRL and SSI showcased very similar behavior and data range across all rounds.

The relationship between net CO₂ assimilation (A) and the leaf water potential (ψ_{leaf}), measured at three different time points per day, is displayed in Figure 8.

Round 1 presented the same pattern as $g_{\text{SW}} \times \psi_{\text{leaf}}$ round 1, with PRD1 not linearly correlated ($y = 1,79x + 18,2$; $R^2 = 0,03$; $p = 0,497$), with higher values of A on average for each ψ_{leaf} . CTRL, SSI and PRD2 were significantly correlated ($p = 0,023$, $p = 0,049$ and $p = 0,013$). On round 2, only PRD2 was significantly correlated ($y = 14,6x + 30,9$; $R^2 = 0,55$; $p < 0,001$) showing a broad range of observations with more extreme values. PRD1 displayed a highly stable A with increasing ψ_{leaf} . In round 3, PRD2 showed again a significant linear correlation ($y = 9,19x + 22,7$; $R^2 = 0,39$; $p = 0,013$) while the other three treatments did not. Overall, PRD1 revealed higher A values on average and PRD2 showed more extreme high and low values across all rounds.

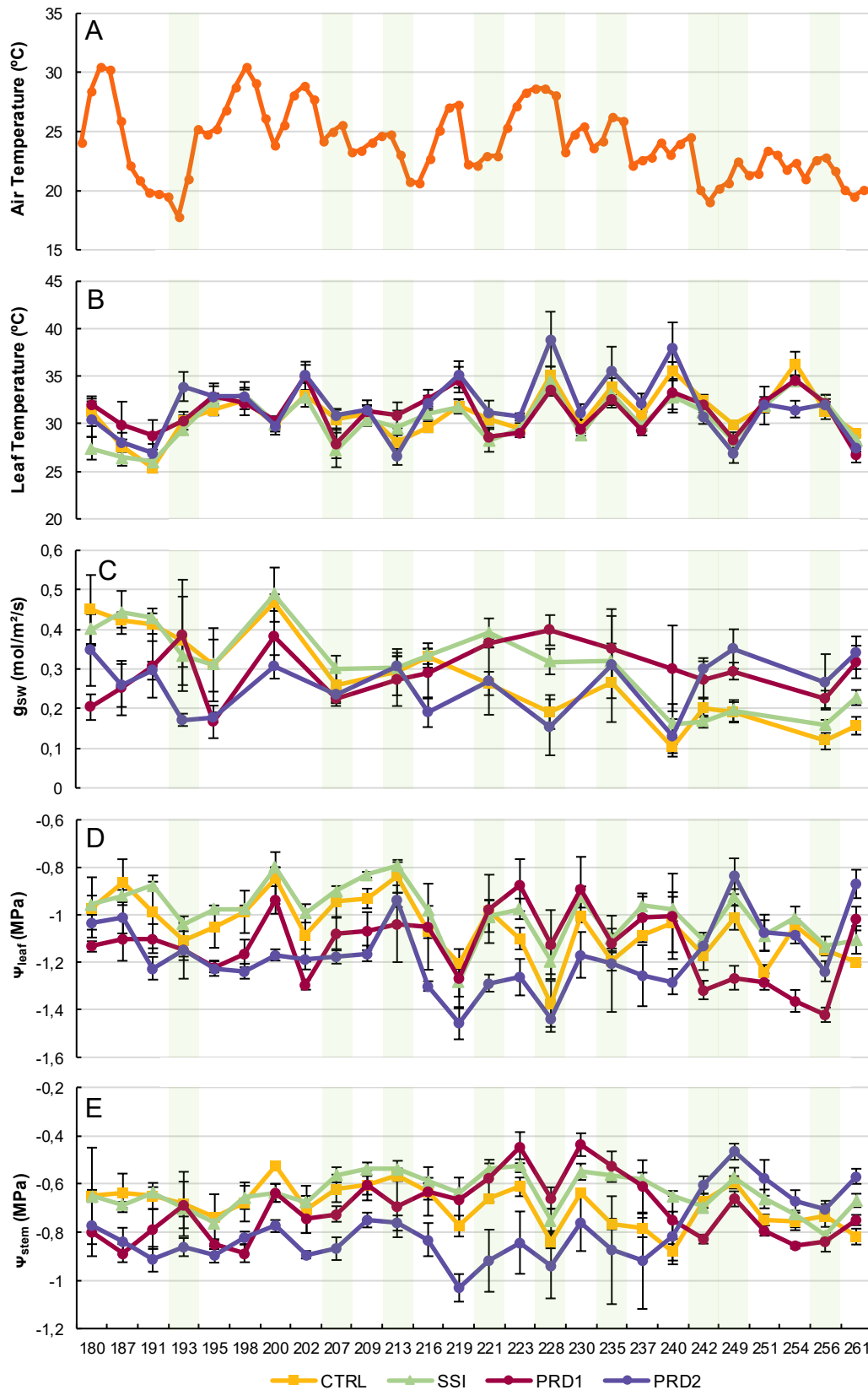


Figure 6 – Seasonal variation of average air temperature (A), leaf temperature (B), midday stomatal conductance (g_{sw}) (C), midday leaf water potential (Ψ_{leaf}) (D) and midday stem water potential (Ψ_{stem}) (E) of Cabernet Sauvignon vines under different irrigation treatments in the 2023 season. Data presented as means of $\pm$ SE ($n \geq 4$) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Measurements represented are from June 29 – DOY 180 to September 18 – DOY 261. Days where measurements were taken in a diurnal fashion (three rounds of one hour each: 10:00-11:00, 13:00-14:00 and 16:00-17:00 PDT) are highlighted in green.

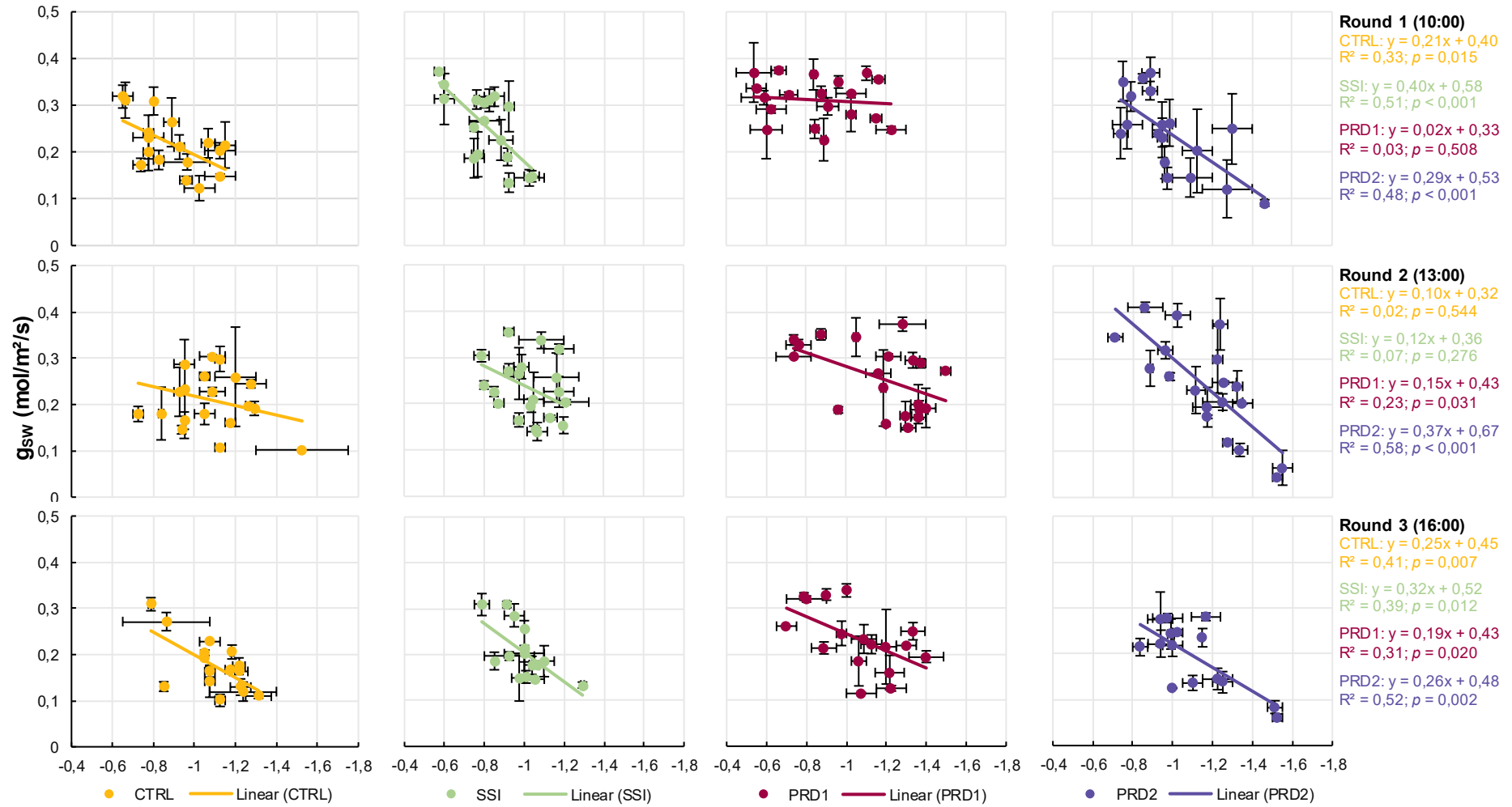


Figure 7 – Relationship between stomatal conductance to water vapor (g_{sw}) and leaf water potential (Ψ_{leaf}) in Cabernet Sauvignon vines under different irrigation treatments in the 2023 season. Treatment abbreviations: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Measurements were taken in a diurnal fashion (three rounds of one hour each: 10:00-11:00, 13:00-14:00 and 16:00-17:00 PDT). Each data point is the mean $\pm$ SE of at least two individual leaves in a replicate. Linear regression equations and their significance are shown for each round and treatment.

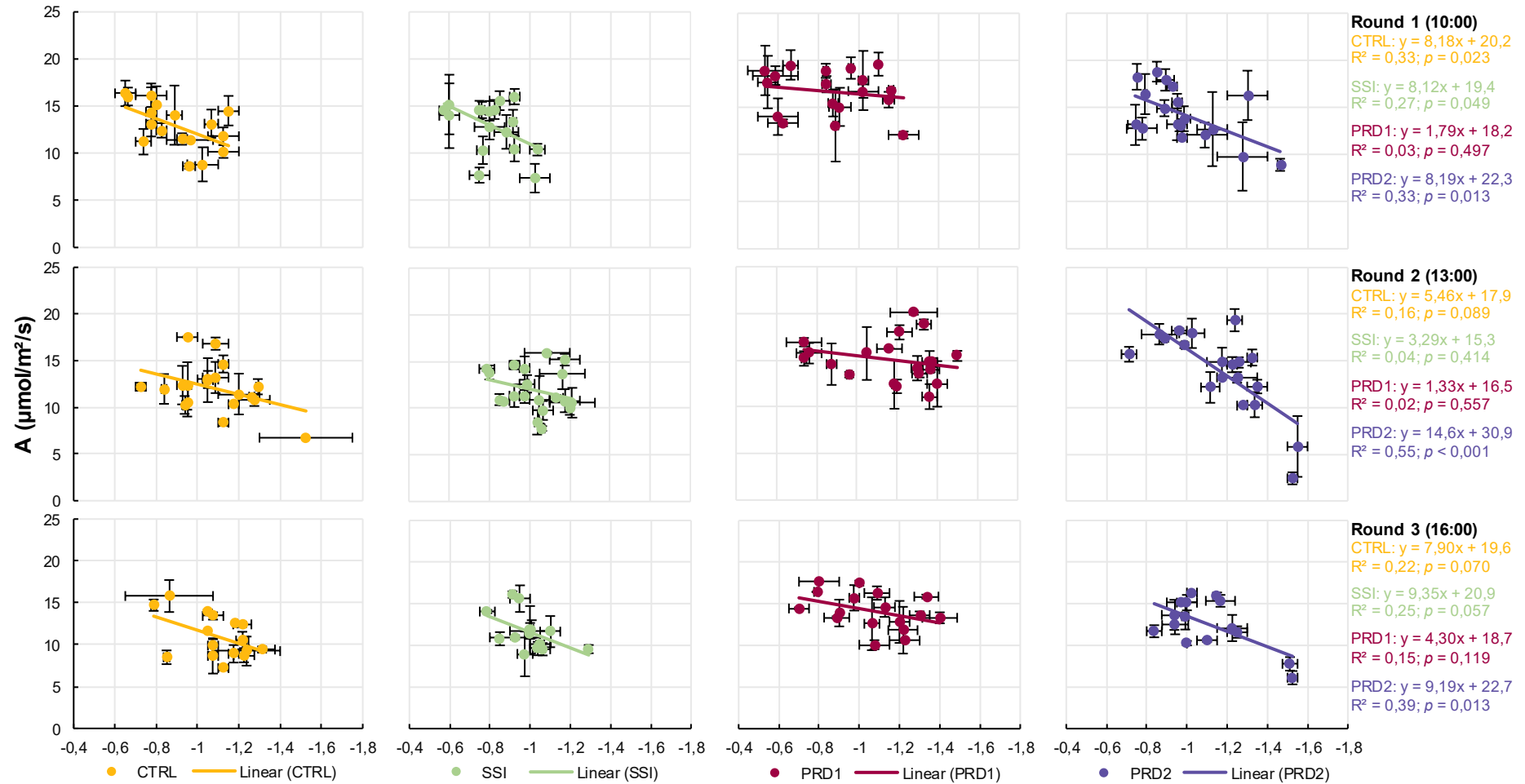


Figure 8 – Relationship between the net CO₂ assimilation (A) and the leaf water potential (Ψ_{leaf}) of Cabernet Sauvignon vines under different irrigation treatments in the 2023 season. Treatment abbreviations: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Measurements were taken in a diurnal fashion (three rounds of one hour each: 10:00-11:00, 13:00-14:00 and 16:00-17:00 PDT). Each data point is the mean $\pm$ SE of at least two individual leaves in a replicate. Linear regression equations and their significance are shown for each round and treatment.

3.4 Yield Components

All yield components were affected by the irrigation treatments, as reported in Table 3.

Vine yield was significantly higher in PRD1 and PRD2 compared to CTRL ($p = 0,018$), while remaining unchanged in SSI. In PRD1 and PRD2, vine yield was increased by 6,26 kg and 5,75 kg compared to the CTRL, which represents a 68% and 63% increment, respectively. Yield differences were mostly attributed to a higher berry weight at harvest ($p < 0,001$), which showed a similar separation between the treatments to vine yield. On average, PRD1 and PRD2 berries were 0,20 g heavier than CTRL and 0,17 g heavier than SSI. The number of clusters per vine and the cluster weight were also significantly increased in both PRD treatments ($p = 0,009$ and $p = 0,037$), although to a lesser extent. No significant differences were observed between PRD1 and PRD2, but PRD1 showed trends for higher yield, clusters/vine, and berry weight, with the exception being cluster weight in PRD2.

Table 3 - Yield components of Cabernet Sauvignon vines under different irrigation practices in 2023.

	CTRL	SSI	PRD1	PRD2	p^a
Yield (kg/vine)	9,20 ± 1,70 b	9,17 ± 1,83 b	15,46 ± 1,82 a	14,95 ± 1,40 a	*
Clusters/vine	164 ± 14 b	154 ± 9 b	201 ± 7 a	188 ± 9 ab	**
Cluster weight (g)	53,54 ± 6,14 c	57,48 ± 8,21 bc	76,37 ± 7,00 ab	79,48 ± 6,87 a	*
Berry weight (g)	1,053 ± 0,025 b	1,088 ± 0,047 b	1,261 ± 0,066 a	1,249 ± 0,069 a	***

Notes: Data presented as means ± SE (n = 6) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side).

^a p values were calculated using a linear mixed model with one fixed effect (treatment) and one random factor (replicate). Asterisks *, ** and *** indicate significant differences at $p < 0,05$, 0,01 and 0,001, respectively.

^bIn each column, values followed by different letters are significantly different according to Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).

3.5 Water Use Efficiency

The agronomic water use efficiency at the crop level (WUE_c) is reported in Table 4. WUE_c values were significantly different between treatments ($p = 0,013$) with PRD2 showing the highest values, with an increase of 4,7 ton/ML (65,7%) when compared to CTRL, and SSI showing the lowest values. Although no significant differences were observed between PRD1 and CTRL, PRD1 showed a 3,4 ton/ML increase compared to the latter.

Table 4 – Agronomic water use efficiency (WUE_c) of Cabernet Sauvignon vines under different irrigation practices in 2023.

	CTRL	SSI	PRD1	PRD2	p^a
WUE_c (ton/ML)	$7,16 \pm 1,34^{bc}$	$6,14 \pm 1,15^c$	$10,54 \pm 1,42^{ab}$	$11,87 \pm 1,11^a$	*

Notes: Data presented as means $\pm$ SE ($n = 6$) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side).

^a p values were calculated using a linear mixed model with one fixed effect (treatment) and one random factor (replicate). Asterisk * indicates significant differences at $p < 0,05$.

^bIn each column, values followed by different letters are significantly different according to Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).

As plant water status and leaf gas exchange were monitored throughout the season, the intrinsic water use efficiency (WUE_i) and its relationship with stem water potential (Ψ_{stem}) are reported in Figure 9. We observed that WUE_i and Ψ_{stem} were linearly related, with increases in WUE_i as Ψ_{stem} became more negative ($y = -43,161x + 34,069$; $R^2 = 0,2981$; $p < 0,001$).

CTRL exhibits a less inclined slope, with lower increases in WUE_i per Ψ_{stem} . The behavior of SSI, PRD1 and PRD2 was similar, with a higher increase in WUE_i per Ψ_{stem} . PRD1 and PRD2 exhibited higher values of Ψ_{stem} and correspondingly higher values of WUE_i , with PRD2 having the highest, and a more pronounced slope overall.

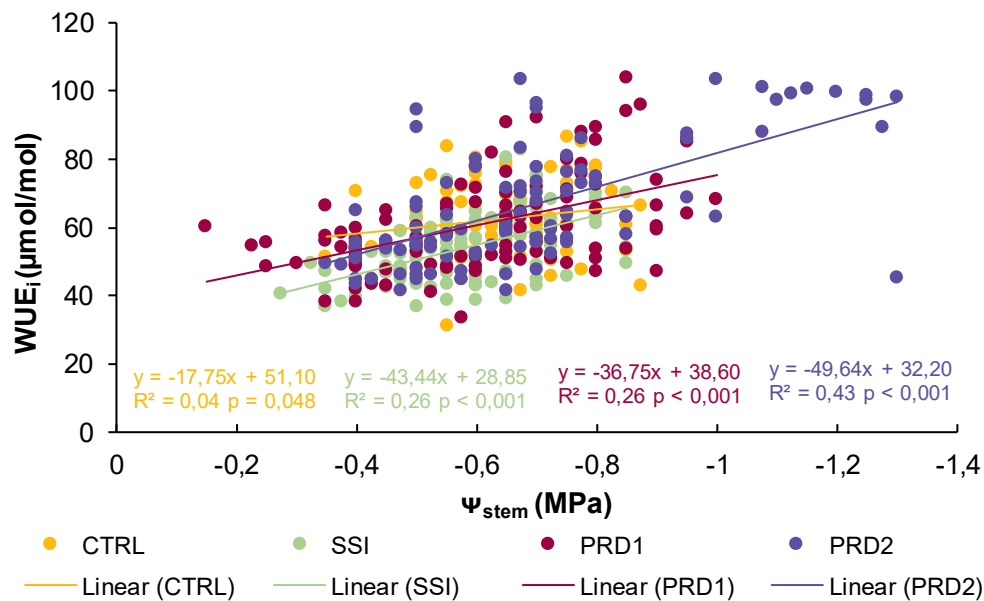


Figure 9 – Relationship between the intrinsic water use efficiency (WUE_i), calculated as A/g_{sw} , and the stem water potential (Ψ_{stem}) of Cabernet Sauvignon vines under different irrigation treatments in 2023. Treatment abbreviations: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Measurements represented are from July 26 – DOY 207 to September 13 – DOY 256. Linear regression equations and their significance are shown for each treatment

3.6 Vine Size/Vine Vigor

3.6.1 NDVI/FC

Remotely sensed FC values by irrigation treatment are reported in Figure 10. FC calculated from imagery collected on July 3 (DOY 184) (Figure 10A), representing the time of maximum canopy size, showed a large significant increase in PRD1 and PRD2 over CTRL ($p < 0,001$), on average 10–13%. SSI did not exhibit any significant differences when compared to CTRL and had 9,5% lower values on average when compared to PRD1 and PRD2.

A FC map of the entire experimental plot on July 3 is shown in Figure 11, where each panel represents one treatment replicate with the average FC (in %) reported above. PRD1 and PRD2 displayed higher percentages of FC when compared to CTRL and SSI, particularly evident in replicates 4 and 5. Additionally, Figure 11 highlights the denser canopy of PRD1 and PRD2, with a greater number of vine pixels. Both of these figures indicate that the PRD vines were already bigger than CTRL and SSI vines very shortly after the irrigation treatments started (June 27 – DOY 178).

The FC progression through the 2023 season is represented in Figure 10B, based on high-resolution imagery collected on a monthly basis. PRD1 and PRD2 sustained a large significant difference between CTRL and SSI in every month ($p < 0,001$), with PRD1 exhibiting the highest values of FC. A dip in overall FC was shown in August, due to mechanical trimming in mid-July.

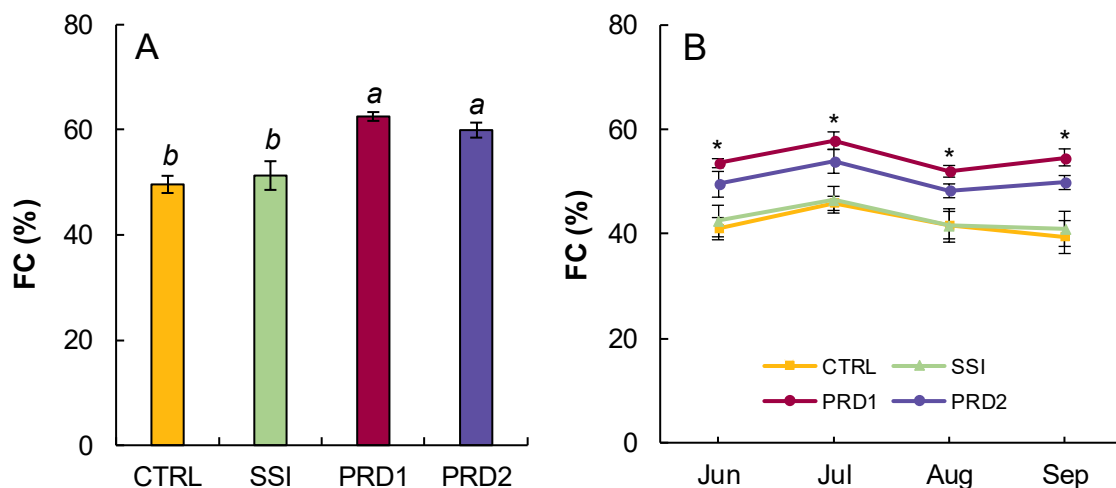


Figure 10 - Fractional cover (FC, in %) of Cabernet Sauvignon vines under different irrigation treatments. Data are presented as means $\pm$ SE ($n = 6$) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side).

10A) FC by irrigation treatment based on imagery collected on July 3 (DOY 184). Different letters indicate statistically significant differences according to Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).

10B) FC progression throughout the experimental trial, with imagery collected once every month (June 7 – DOY 158; July 3 – DOY 184; August 2 – DOY 214; September 11 – DOY 254). Asterisks * indicate significant differences ($p < 0,001$) between treatment means for the sampling date, analyzed using a linear mixed model with one fixed effect (treatment) and one random factor (replicate).



Figure 11 – Fractional cover (FC, in %) map of Cabernet Sauvignon vines under different irrigation treatments. FC was calculated from imagery collected on July 3 (DOY 184). Map of pixels classified as 1 (vine) or 0 (no vine), and the % is calculated as proportion of 1 vs 0 values. Each panel represents one treatment and replicate (60 vines each). The average FC of each treatment and replicate is reported above each panel. Treatment abbreviations: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side).

3.6.2 Pruning Weights

The effect of the irrigation treatments on vine size and vigor parameters is shown in Table 5.

Pruning weights (kg/vine) were impacted by both PRD treatments, with a significant increase compared to CTRL ($p = 0,008$). On average, pruning weights in PRD1 and PRD2 vines were 1,12 kg and 0,81 kg higher than the CTRL, which represents a 44,6% and 32,3% increase, respectively. The number of shoots per vine also showed significant differences ($p = 0,025$) compared to CTRL, with PRD2 exhibiting the highest values. Both pruning weights and number of shoots per vine were affected by the two mechanical trimmings that occurred in July and September. Average shoot weight was unchanged across the treatments ($p = 0,054$), but there was a tendency for PRD1 to have heavier shoots. The Ravaz Index (yield/PW) was higher in both PRD treatments, although no significant differences were observed between them and CTRL and SSI, the latter treatments exhibiting greater variability among replicates. SSI revealed no significant differences compared to the CTRL in all the measured parameters.

Table 5 - Vine size/vigor parameters and Ravaz Index in Cabernet Sauvignon vines under different irrigations practices in 2023.

	CTRL	SSI	PRD1	PRD2	p^a
Pruning Weight (kg/vine)	2,51 ± 0,09 ^b	2,53 ± 0,20 ^b	3,63 ± 0,37 ^a	3,32 ± 0,24 ^a	**
Shoots/vine	71 ± 3 ^c	76 ± 5 ^{bc}	87 ± 6 ^{ab}	90 ± 4 ^a	*
Shoot weight (g)	35,16 ± 1,7	33,30 ± 1,9	41,84 ± 2,9	36,78 ± 1,5	<i>n.s.</i>
Ravaz Index (kg fruit/kg PW)	3,67 ± 0,70	3,63 ± 0,86	4,25 ± 0,31	4,51 ± 0,28	<i>n.s.</i>

Notes: Data presented as means ± SE (n = 6) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side).

^a p values were calculated using a linear mixed model with one fixed effect (treatment) and one random factor (replicate). Asterisks * and ** indicate significant differences at $p < 0,05$ and $0,01$, respectively. *n.s.* indicates non-significant differences.

^bIn each column, values followed by different letters are significantly different according to Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).

3.7 Berry Maturity Monitoring

The treatments applied altered berry growth dynamics, as depicted in Figure 12A. Both PRD1 and PRD2 exhibited a higher berry weight when compared to CTRL and SSI, starting at DOY 235, with significant differences throughout the sampling dates ($p \leq 0,017$ at all sampling dates). The largest separation between treatments was observed at DOY 264 (first harvest), when PRD1 and PRD2 exhibited a 0,25 g average berry weight increase in comparison to CTRL ($p < 0,001$), representing a difference of 24,1%. Post-veraison berry growth plateaued earlier and at a lower berry weight in CTRL when compared to both PRD treatments, showing on average 16,3% smaller berries throughout the significantly different sampling dates. SSI showed virtually equal values to CTRL, with no significant differences.

Both PRD treatments and SSI affected the progression of berry ripening when compared to CTRL, as shown in Figure 12B. From DOY 228 to DOY 270, significant differences were observed between CTRL and all other treatments ($p \leq 0,012$). PRD1 and PRD2 showed an overall slower TSS accumulation when compared to CTRL (0,9 °Brix on average), with the biggest contrast observed at DOY 256 with a 1,14 °Brix difference. SSI also exhibited slower TSS accumulation when compared to CTRL, albeit with smaller differences (2,4% on average).

The temporal progression of basic biochemical juice parameters is displayed in Figure 13 and the detailed statistically significant differences in Figure 17, Appendix D. Overall, differences between treatments were subtle in most parameters, except NOPA, YAN and TSS. There was a trend for SSI to display higher acidity parameters throughout the sampling dates, but differences weren't consistent. PRD1 and PRD2 exhibited higher values of YAN when compared to CTRL, but only PRD2 was significantly different in almost all sampling dates ($p \leq 0,042$) with an average of 14 mg/L higher values. Similarly, PRD2 showed higher values of NOPA when compared to CTRL, significantly different in four of the six sampling dates ($p \leq 0,041$). TSS showed the same pattern described above, with both PRD treatments and SSI displaying slower TSS accumulation.

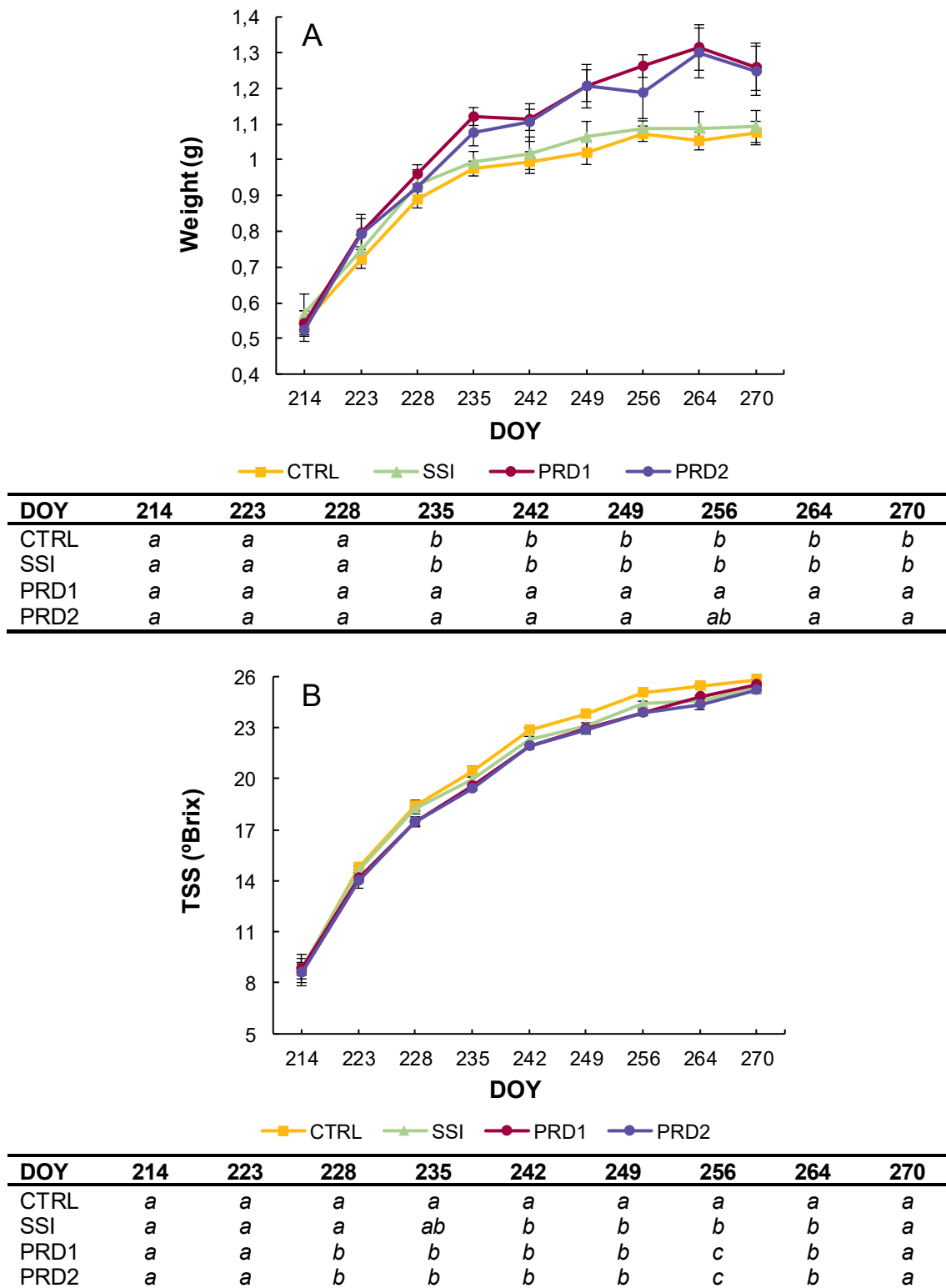


Figure 12 – Temporal progression of berry weight (**A**) and TSS (**B**) from veraison (August 2 – DOY 214) to harvest (September 27 – DOY 270) for the 2023 season. Data presented as means $\pm$ SE ($n = 6$) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Tables indicate the significant differences among treatment means for each sampling date according to a linear mixed model with one fixed effect (treatment) and one random factor (replicate), followed by Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).

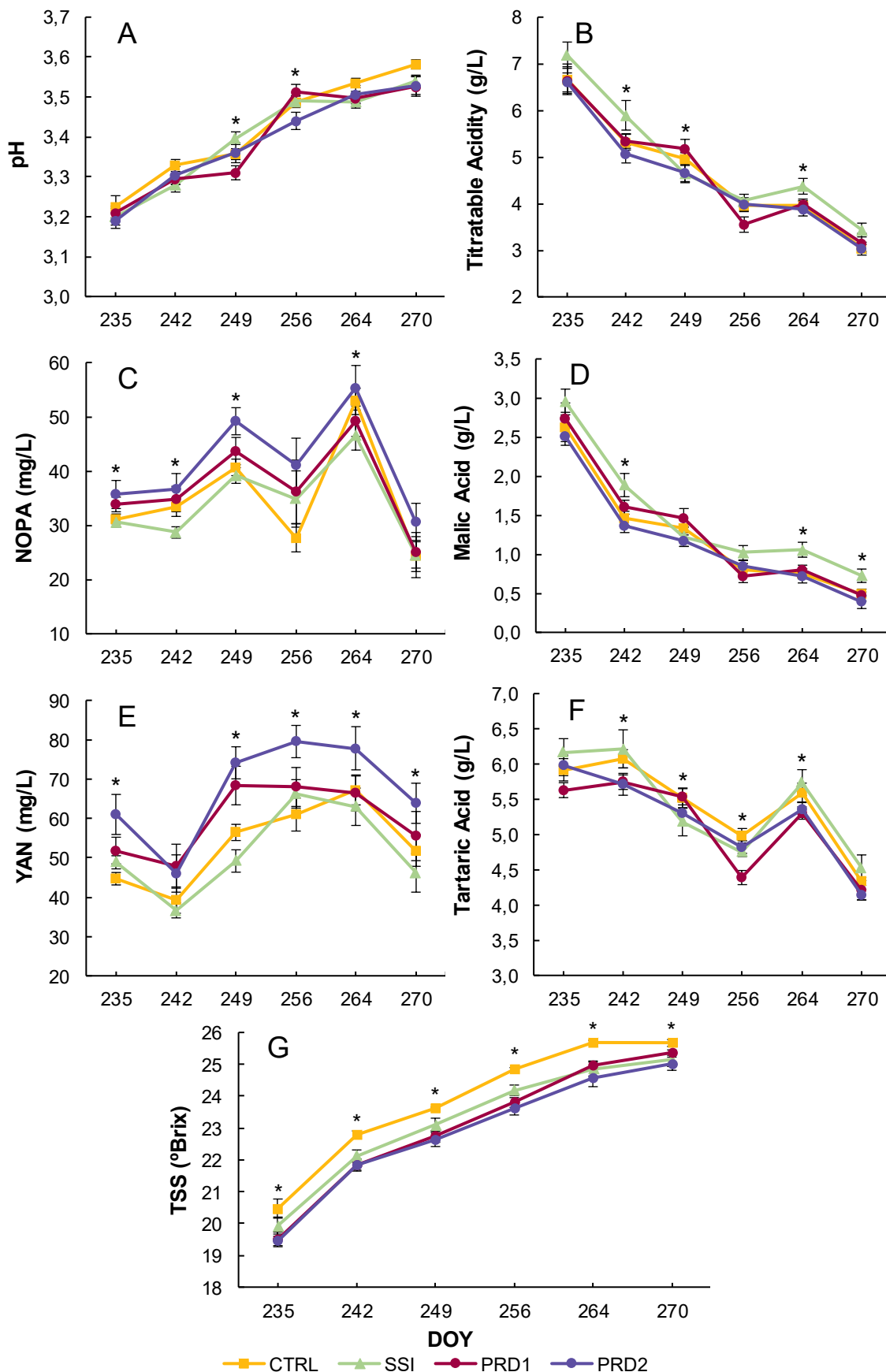


Figure 13 – Temporal progression of basic biochemical parameters (**A – G**) of Cabernet Sauvignon juice during ripening. Samples collected in the later stages of ripening (DOY 235 – DOY 270) of the 2023 season. Data presented as means $\pm$ SE (n = 6) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Asterisk * indicates significant differences ($p < 0,05$, $0,01$ or $0,001$) between treatment means for the sampling date, analyzed using a linear mixed model with one fixed effect (treatment) and one random factor (replicate). Harvest date for CTRL, SSI – September 21 (DOY 264) and for PRD1, PRD2 – September 27 (DOY 270). Full list of treatment means differences in Appendix D.

3.8 Grape Chemical Composition

The chemical analysis of the sequential grape cluster sampling is depicted in Figure 14.

Regarding basic biochemistry (A – E and M), only TSS and berry moisture were significantly different throughout all the sampling dates ($p \leq 0,016$ and $p \leq 0,012$). CTRL and SSI showed faster TSS accumulation (0,92 °Brix on average) than PRD1 and PRD2, with CTRL displaying consistently the higher values and PRD1 the lowest. Inversely, PRD1 and PRD2 exhibited an average of 1,13% higher berry moisture values when compared to CTRL and SSI, with PRD1 showing the highest and CTRL the lowest. YAN values started significantly lower for CTRL and SSI when compared to PRD1 and PRD2 ($p \leq 0,023$) but caught up in Week 4 – DOY 264.

Most volatile compounds (F – I) didn't show clear trends. SSI displayed higher values of 1-octen-3-ol in all weeks when compared to the other treatments ($p \leq 0,014$), with CTRL matching in Week 4. Both showed an average increase of 2,9 µg/L when compared to PRD1 and PRD2. SSI also displayed the lowest IBMP values in Week 4 ($p = 0,007$).

Phenolic compounds (J – L) showed a clear trend for overall lower concentrations in PRD1 and PRD2 across all weeks. Total anthocyanins (0,21 mg/g berry on average, $p \leq 0,008$), quercetin glycosides (52,3 mg/L on average, $p \leq 0,007$) and polymeric tannins (537,6 mg/L on average, $p \leq 0,002$) were lower in PRD1 and PRD2, when compared to CTRL and SSI.

The grape composition at harvest is shown in Table 6.

Concerning basic biochemistry, no significant differences between treatments were observed in most parameters, with the exception of tartaric acid ($p = 0,038$) and YAN ($p = 0,031$), which were highest in PRD2 and lowest in SSI.

Regarding volatile compounds, the analysis showed a large significant decrease (828 µg/L on average, $p = 0,016$) in the concentration of C₆ compounds in PRD1 and PRD2 compared to CTRL and SSI, and a significant decrease ($p = 0,003$) in the concentration of 1-octen-3-ol. IBMP levels were unchanged and extremely low across the four treatments ($p = 0,270$).

Significantly lower concentrations of all the tested phenolic compounds were observed in PRD1 and PRD2, including total anthocyanins (0,24 mg/g berry on average, $p = 0,023$), quercetin glycosides (58,75 mg/L on average, $p < 0,001$) and polymeric tannins (745,53 mg/L on average, $p < 0,001$).

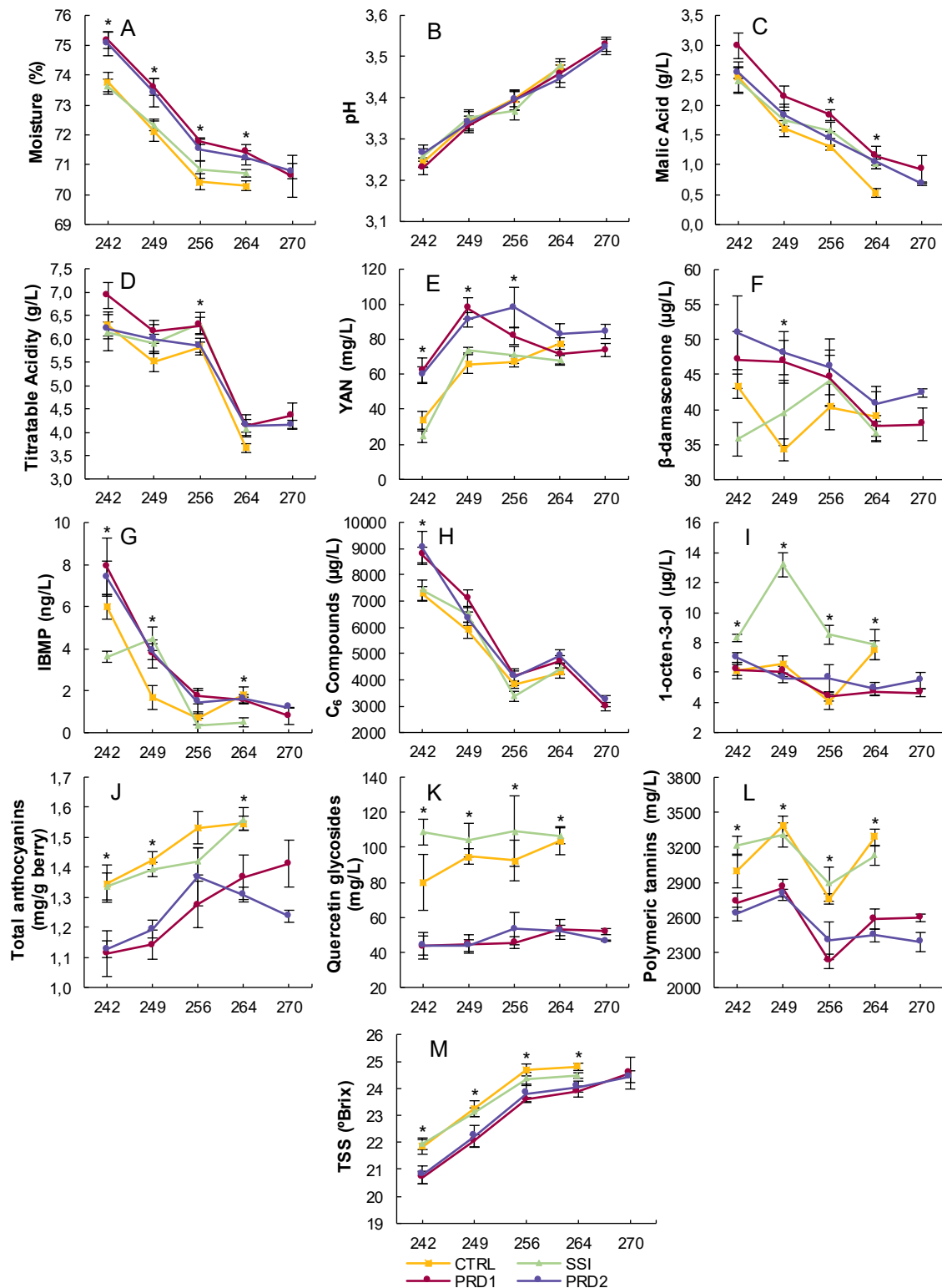


Figure 14 - Weekly maturity level chemical analysis (A – M) of Cabernet Sauvignon cluster samples during the later stages of ripening (DOY 242 – Week 1; DOY 249 – Week 2; DOY 256 – Week 3; DOY 264 – Week 4 & VM and WL; DOY 270 – VM and WL) of the 2023 season. Harvest date for CTRL, SSI – September 21 (DOY 264) and for PRD1, PRD2 – September 27 (DOY 270). Data presented as means $\pm$ SE (n = 6) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Asterisks * indicate significant differences ($p < 0.05$, 0.01 or 0.001) between treatment means for the sampling date, analyzed using a linear mixed model with one fixed effect (treatment) and one random factor (replicate). Full list of treatment means differences in Appendix E.

Table 6 – Grape chemical composition at harvest of Cabernet Sauvignon vines under different irrigation practices in 2023.

	CTRL	SSI	PRD1	PRD2	<i>p</i> ^a
Basic biochemistry					
Total Soluble Solids (°Brix)	24,9 ± 0,2	24,4 ± 0,1	24,6 ± 0,6	24,4 ± 0,2	<i>n.s.</i>
Moisture (%)	70,11 ± 0,18	70,72 ± 0,15	70,61 ± 0,70	70,78 ± 0,25	<i>n.s.</i>
pH	3,48 ± 0,02	3,48 ± 0,02	3,53 ± 0,02	3,52 ± 0,02	<i>n.s.</i>
Titrateable Acidity (g/L)	3,62 ± 0,24	4,03 ± 0,23	4,35 ± 0,27	4,16 ± 0,10	<i>n.s.</i>
Malic Acid (g/L)	0,53 ± 0,22	0,99 ± 0,14	0,92 ± 0,23	0,68 ± 0,03	<i>n.s.</i>
Tartaric Acid (g/L)	5,36 ± 0,14 ^{ab}	5,09 ± 0,13 ^b	5,48 ± 0,10 ^{ab}	5,72 ± 0,12 ^a	*
NOPA (mg/L)	50,33 ± 9,67	26,33 ± 3,28	36,00 ± 2,31	41,67 ± 1,20	<i>n.s.</i>
YAN (mg/L)	74,73 ± 3,36 ^{ab}	63,35 ± 4,21 ^b	73,84 ± 3,73 ^{ab}	84,44 ± 4,08 ^a	*
Volatile compounds					
β-damascenone (μg/L)	41,55 ± 6,75	37,51 ± 1,81	37,91 ± 2,33	42,37 ± 0,60	<i>n.s.</i>
IBMP (ng/L)	1,26 ± 0,39	0,87 ± 0,45	0,80 ± 0,41	1,18 ± 0,01	<i>n.s.</i>
C ₆ Compounds (μg/L)	3965,4 ± 197,9 ^a	3869,6 ± 198,9 ^a	2984,5 ± 159,8 ^b	3194,1 ± 211,0 ^b	*
1-octen-3-ol (μg/L)	6,54 ± 0,54 ^a	7,12 ± 0,12 ^a	4,65 ± 0,26 ^b	5,47 ± 0,54 ^b	**
Phenolic compounds					
Total anthocyanins (mg/g berry)	1,53 ± 0,06 ^a	1,59 ± 0,06 ^a	1,41 ± 0,08 ^{ab}	1,24 ± 0,02 ^b	*
Quercetin glycosides (mg/L)	103,73 ± 11,71 ^a	112,31 ± 11,20 ^a	51,85 ± 1,74 ^b	46,69 ± 0,35 ^b	***
Polymeric tannins (mg/L)	3265,5 ± 122,3 ^a	3213,2 ± 127,4 ^a	2597,3 ± 32,5 ^b	2390,3 ± 82,0 ^b	***

Notes: Data presented as the means ± SE (n = 3) per treatment: CTRL (control irrigation), SSI (subsurface irrigation), PRD1 (partial rootzone drying alternated), PRD2 (partial rootzone drying one-side). Harvest date for CTRL, SSI – September 21 (DOY 264) and for PRD1, PRD2 – September 27 (DOY 270)

^a *p* values were calculated using a linear mixed model with one fixed effect (treatment) and one random factor (replicate). Asterisks *, ** and *** indicate significant differences at *p* < 0,05, 0,01 and 0,001, respectively. *n.s.* indicates non-significant differences.

^b In each column, values followed by different letters are significantly different according to Fisher's Least Significant Difference (LSD) test at the 95% confidence level (*p* < 0,05).

4. Discussion

1. Is a novel approach to PRD, combining principles of subsurface irrigation and partial rootzone drying, capable of re-creating the benefits associated with PRD in controlled conditions at the commercial scale?

The fundamental objective of PRD is to create intentional soil moisture heterogeneity, both spatially and temporally, within the grapevine root system. In theory, this heterogeneity modulates root-to-shoot signaling and consequently changes the physiology of the shoots and leaves (Dry et al., 2001; Stoll, 2000). Research indicates that, in general, a successful PRD irrigation experiment must fulfill two criteria: a minimum threshold of soil water content to trigger a physiological response; and a clear distinction between the “wet” and “dry” rootzones (Poni et al., 2007; Romero et al., 2014; Stoll, 2000; Stoll et al., 2000a). Properly drying part of the root system is necessary to achieve calibrated water stress to trigger modulation of vine physiology that can lead to improved WUE. In addition, alternation of the “wet” and “dry” zones seems to be necessary to recreate such benefits over time and avoid that such positive responses disappear. In previous research, PRD-specific physiological and agronomic changes cannot always be discriminated (Intrigliolo & Castel, 2009; Marsal et al., 2008; Rodrigues et al., 2008). These unsuccessful attempts at PRD could be explained by inconsistencies in achieving a separation between the “wet” and “dry” portions of the rootzone. This supports the large significant effects observed when the rootzone was split by the use of a physical barrier, such as two distinct pots in controlled conditions or plastic dividers in field-like conditions (Antolín et al., 2006; Dry, 1997; Poni et al., 2007; Stoll, 2000). If such separation is necessary for PRD to cause physiological effects of interest, this could provide a reasonable explanation for the failed attempts at PRD through the alternation of irrigation in adjacent vine spaces (Baeza et al., 2005; Balint & Reynolds, 2014; Bravdo et al., 2004; Chalmers et al., 2004; Du et al., 2008; Gu et al., 2004; Pudney & McCarthy, 2004). The latter represents an option easily implemented at the commercial scale by adding a second drip line with emitters in alternated vine spaces. In this system, the “wet” and “dry” zones are directly adjacent and water movement in the soil may avoid the proper formation of such zones.

In the present study, a novel approach to PRD was used that combines spatial and temporal alternation in irrigation with subsurface irrigation. This novel technique was directly aimed at maximizing the separation between the “wet” and “dry” zones in commercial vineyards. By moving the drip lines to the middle of the interrows on both sides of the vines, the distance between the two irrigation zones was maximized (3,35 m). Even though subsurface irrigation became needed to protect the drip lines newly installed in the middle of vine rows, the combination of subsurface irrigation and PRD may represent a strategy to further maximize WUE.

This new PRD approach resulted in significant spatial and temporal variation. Soil moisture data represented in Figure 5 confirmed the success of establishing distinct “wet” and “dry” areas and cycles at the targeted soil depth of 30 cm, showing a heterogeneous distribution of soil moisture in the interrows where the PRD drip lines were strategically buried. The use of the WATERMARK sensors at three positions in relation to the vine row provided feedback on the drydown progression in the “dry” zones and irrigation

effectiveness in the “wet” zones of PRD1. In PRD2, readings validated the soil drydown of the south row and effective irrigation of the north row, with the middle sensor data demonstrating a clear separation of “wet” and “dry” zones. In the CTRL and SSI treatments, irrigation was applied directly under the vines, which was verified by soil moisture data showing adequate levels directly under the vine (Middle sensor) and a drying effect throughout the season for the two sensors placed in row middles (North and South sensors). The WATERMARK (tensiometer) measurements were used to understand the amount of water available for the vines and to trigger the alternation of the “wet” and “dry” zones in PRD1, while the precise amounts of water applied to each treatment were tracked with flowmeters. One limitation of using tensiometers is that their return information relies on point-specific measurements. Although three sensors were positioned in a row to enhance soil representation, variability in the soil at the study site may provide some interference. Future work could benefit from integrating soil moisture modeling techniques to create comprehensive moisture maps, such as Tao et al. (2023).

The alternation of irrigated interrows on PRD1 was based on the dryness level of each side instead of fixed temporal intervals (e.g., Bravdo et al. (2004); De la Hera et al. (2007)) because, as thought of initially and confirmed throughout the experiment, Rep 1 (located on top of the hill) and Rep 2 (located on the bottom of the hill) showed very different soil drying patterns (Figure 5). These differences were due to variations in soil composition, which progressed from sandy loamy to more clay-dominant downslope, and soil depth, which created different water retention dynamics in the two spots. Some studies suggested that sandy-type soils may be better for PRD application because they allow for higher water infiltration rates and deeper rootzones (Chaves et al., 2007; dos Santos et al., 2007). The switching thresholds in our study (75-100 cbar) were based on the soil type and moisture, and were established from pre-existing knowledge of the response of the vines to water deficit (IRRROMETER Company, 2022). The observed drying patterns matched our expectations. Over a period of 10 to 15 days, the soil moisture data indicated a clear progression towards a critical threshold in the drying rootzone, highlighting the need to reapply irrigation. A separate study could be performed to determine the optimal PRD switching points in this vineyard.

The second prerequisite for the success of PRD is the creation of calibrated water stress, achieved via partial drying of the rootzone, to induce physiological responses linked to WUE (Dry et al., 2001; Stoll et al., 2000b). This aspect is often overlooked in previous literature, where the lack of irrigation volume tracking makes it difficult to substantiate if the irrigation thresholds set for the study were met. In this study, we decided to compare treatments irrigated at the same level, set as 80% of ET_c, and daily irrigation volumes were recorded using flowmeters (Table 2). The VRI system installed allowed the scheduling of irrigation events as well as the verification of their completion by means of flow monitoring (Figure 16, Appendix C). VRI systems have been proven very effective in vineyards, enabling significant water savings through precise irrigation according to vineyard spatial variability (Ortuani et al., 2019). Overall, irrigation volumes applied were in line with the targeted values, with the exception of a few replicates where the system failed temporarily. PRD1 R5&R6 had an unexpected increase in irrigation volume during the early stages of the trial due to a faulty valve opening. The issue was resolved and the water volume adjusted, and the majority of the excess water was lost as runoff to non-experimental areas of the vineyard. The increase in the total irrigation volume of SSI was driven by difficulties in using this new prototype system. Larger water

applications were due to damages to the small polyethylene tubes caused by animals, which resulted in occasional leaks that were still recorded by the flowmeters. SSI provided additional challenges and required several adjustments to the system due to the vineyard soil characteristics. The mostly fine-textured soil demonstrated slower-than-expected water permeability, which created challenges in managing the irrigation volumes applied. Specifically, the applied water exceeded the soil's infiltration capacity, which led to backflow and runoff through the top of the PVC pipes that constituted the system. As a result, the water savings that could be expected when SSI is applied were lowered in this study, as discussed below. Ma et al. (2020a) reported no irrigation difficulties when implementing a similar subsurface irrigation strategy (i.e., DRZ) in sandy soil; however, they did apply it at a much smaller scale and at different insertion angles. Adjustments were, therefore, made to each irrigation event while ensuring the same water volume applied as the other treatments. Modifications to this prototype should be made in the future to maximize the benefits associated with moving from above-ground to subsurface irrigation.

Our approach successfully demonstrated distinct and well-defined patterns of “wet” and “dry” cycles with precise irrigation application that ensured that the intended treatments were accurately applied. These results confirm that the experimental system was sound and robust enough to successfully study PRD at a commercial scale.

2. What are the benefits of applying PRD at a commercial scale using this novel approach in terms of vine performance, yield, and grape composition?

One of the objectives of this study was to determine whether a novel PRD approach, which combines principles of subsurface irrigation and PRD, could positively impact vine performance, yield, and quality. Data collected throughout the growing season showed that PRD profoundly altered vine performance, fruit yield, and overall quality.

FC data from July 3 (DOY – 184), just seven days after irrigation began on June 27 (DOY – 178), already showed differences in canopy growth on both PRD treatments, represented by a 10-13% increase in FC when compared to CTRL (Figure 10A). FC is a proxy index for vine size based on remotely sensed data (which inherently has a degree of error) (Pereira et al., 2020). Pruning weight data collected at the end of the season (Table 5) corroborated the FC values computed from drone imagery, with an average increase of 1 kg/vine in both PRD treatments compared to CTRL. Pruning weight values are to be considered underestimated due to canopy trimming being performed right before harvest (Table 7, Appendix A) to limit MOG (material other than grape) levels. Pre-harvest trimming reduces, on average, 20-25% of the canopy; however, this could be more and could be different between treatments when the vine size has been impacted, such as in this case. In spite of limitations, pruning weight levels were statistically different, signifying a larger effect or indicating differences in shoot diameter and not only in shoot length. It is highly unlikely that the irrigation treatments caused such rapid growth within a week, especially when all treatments were irrigated at the same rate. Since this trial was started in 2022, it can be speculated that this was a potential carry-over effect from the previous year. In the first year of PRD application, the vines were stressed as they adapted to the new irrigation strategy, as the water was placed

further away (data not shown). As a result of water redistribution, changes in root growth and allocation were most definitely imposed, and in our trial year (2023, the second year), the PRD vines likely began the season with a more extensive and established root system. The timing of our measurements, around maximum canopy size and treatment differences, showing an increase in both PRD1 and PRD2, points to an important effect of relocating water to the subsurface and in the interrows. Newer roots were likely formed in response to the first year of PRD, adding to pre-existing roots and leading to an overall more extensive root system. The unusually cool and rainy winter weather in 2023 (Figure 3 and Figure 4) may have also inflated these differences, given the non-limiting conditions. While PRD1 and PRD2 were statistically similar, a trend for higher FC was always observed. This and other trends for improved effects in PRD1 compared to PRD2 discussed below suggest that there may be some important benefits associated with the temporal alternation of “wet” and “dry” zones. The hypothesis we have formulated regarding larger rootzones in PRD treatments should be verified by directly analyzing root density or using root mapping techniques.

In addition to distinct canopy growth patterns across treatments, seasonal variations in the vine water status were observed. These were characterized by measuring water potential as well as leaf temperature (Figure 6). Seasonal trends of midday Ψ_{stem} and Ψ_{leaf} of both PRD treatments were usually more variable and with more negative values than CTRL and SSI. This may suggest that the larger vines of the PRD treatments were more prone to becoming water stressed. Midday g_{sw} measurements led to slightly different trends, with both PRD treatments exhibiting a narrower range of values, possibly indicating tighter stomatal control. At the same time, CTRL and SSI shifted from having the highest g_{sw} values in the early season to the lowest in the later stages. Seasonal fluctuations were mostly driven by seasonal weather patterns, as the 2023 growing season was characterized by early heat spikes in July followed by cooler-than-normal conditions leading to harvest (Figure 4 and Figure 6A). In addition to this, PRD1 trends showed additional disturbance from the irrigation manipulation, due to imposing additional stress by switching “wet” and “dry” zones based on soil moisture data, on average every 10 to 15 days.

Interestingly, effects specific to PRD1 were highlighted when gas exchange parameters were related to water status ones (Figure 7 and Figure 8). The relationship between g_{sw} and Ψ_{leaf} has been investigated before in grapevines, and it is a negative linear relationship where g_{sw} decreases in response to more negative Ψ_{leaf} , which is explained by stomatal closure occurring as water status becomes more limiting (Williams & Baeza, 2007; Williams et al., 2010). In our analysis, g_{sw} and Ψ_{leaf} data were pooled together, and their relationship by treatment was analyzed at three times of the day, namely 10:00, 13:00, and 16:00 PDT. All treatments and rounds reflected the expected inverse relationship reported in the literature. The only exception was PRD1 in the first round (10:00-11:00), where g_{sw} appeared to be maintained, even with more limiting conditions of water stress. This trend, specific to PRD1 and possibly driven by measurements taken after the switch in the irrigation zone, suggests differences in how stomata regulate water use in response to vine water status under PRD. This observation contrasts with the findings of Dry (1997) and Stoll (2000) who characterized PRD as a strategy that reduces g_{sw} to regulate vigor. Differences in PRD implementation strategies are likely the reason behind this discrepancy, as PRD irrigation water was reduced by 50% by these authors while our PRD method maintained the same irrigation volume as

CTRL. In both cases, an improvement was observed in the PRD-treated vines, characterized as: a less than proportional decrease in g_{sw} to water reduction in prior studies; and improved g_{sw} or A in response to the same water input in the case of our analysis. The time of the day at which this improvement was observed is another important aspect. In field-grown, split-root cv. Cabernet Sauvignon vines, Stoll (2000) reported that the biggest effect on g_{sw} was between 09:00 and 11:00. Also, during a similar time frame (10:00-12:00), PRD-treated vines exhibited significantly higher concentrations of ABA in xylem sap compared to the control. We found a similar difference when we related the g_{sw} and ψ_{leaf} in the first round (Figure 7), which was at a similar time of the day (10:00-11:00). This suggests that there is a modulation of physiological activity under PRD at this time of the day, observed explicitly in PRD1 and not in PRD2, likely linked to the switching. Future work should investigate the mechanisms underlying this phenomenon and could benefit from hormonal and transcriptomic analyses to fully elucidate the link between effects at the root and canopy level.

Linking vine performance data to the final yield, the effect of the treatments on the final yield was clearly dependent on canopy size. The separation in yield values between the treatments (Table 3) reflected the differences in FC and pruning weights, with PRD1 and PRD2 displaying higher yields than CTRL and SSI. This difference was statistically significant and up to 68% in PRD1. Such an increase in yield is enticing to growers, especially in situations in which remunerations are mostly dependent on yield, and when said yield increases by 68% while maintaining irrigation to 80% ETc. Berry weight data was in agreement with the final yield, as berry weight values at harvest differed in a similar manner to vine yield (Table 3). Berry growth curves (Figure 12A) highlighted that PRD1 and PRD2 berries continued growing even after SSI and CTRL berry growth plateaued, leading to a 0,20 g increase in berry weight at harvest.

Similar to data collected for FC and pruning weights, there was a trend for increased yield in PRD1 compared to PRD2. When yield data was collected using a yield monitor mounted on a mechanical grape harvester (data not shown), the increase in PRD1 yield was over 2 tons/ha, and became statistically significant due to the bigger sample size, compared to the limited number of manually harvested vines (Table 3). It is also possible that, in future seasons, drier conditions in the early season (compared to the extremely wet conditions in 2023) may lead to a bigger separation between PRD1 and PRD2, as the latter was the treatment where the most considerable water stress was measured in 2023. Adaptation is also expected to occur over time when irrigating only on a single side, as the pre-existing roots become less and less functional if not irrigated over several seasons (Romero et al., 2016b). All these considerations point to the alternated “wet” – “dry” cycles being a strategy that affects shoot growth, canopy biomass, and vine yield. The improved relationship between g_{sw} and ψ_{leaf} may be connected to these results, as trends for A rates were in line with g_{sw} results (Figure 8).

The impact of source-sink relations, particularly crop load, on grape ripening has been extensively documented (Kliewer & Dokoozlian, 2005). Crop load is widely accepted as a good measure of vine balance (Howell, 2001), and in our study, we characterized crop load using the Ravaz index (yield/PW ratio). Crop load was not significantly different between the treatments investigated in this study. The Ravaz Index was just below 5, which is close to the optimal range suggested for GDC trellised

Cabernet Sauvignon grapes in the region (5-10 kg of fruit/kg of PW; Kliewer and Dokoozlian (2005)). Considering the effect of pre-harvest canopy hedging discussed above ($\pm 20\%$ of PW), the vines would have fallen in the perfect cropping conditions according to the literature. The unchanged yield-to-PW ratio indicates that increases in yield were proportionally matched by the increases in vegetative growth, as discussed above. These results further support that the increased vine capacity in PRD treatments due to the relocation of water overrode the effects of switching sides in PRD1. A higher number of shoots per vine was also observed in PRD1 and PRD2 compared to CTRL, which might signify an improved fruitfulness from the increases in canopy biomass and root growth discussed before.

Previously, it has been demonstrated that crop load directly affects berry ripening kinetics (Bravdo et al., 1985; Miller & Howell, 1998). In general terms, advanced ripening occurs when vines are under-cropped, and delayed ripening is associated with over-cropped vines, where photosynthates produced by the canopy do not meet the fruit demand. In our study, significant differences were observed for both TSS and berry weight, mainly at the end of ripening. Most, if not all, of the berry growth progression differences between CTRL, SSI, and both PRD treatments, shown in Figure 12A, can probably be attributed to berry cell enlargement (increase in cell volume), not new cell growth, since significant weight differences only manifested throughout veraison, and not at the start – DOY 214, 36 days after the irrigation treatments began. The number of cells in a grape berry is established during the first three weeks after anthesis (flowering) and almost no further cell division occurs after this period (Christensen, 2000; Coombe, 1976), and berry weight was virtually equal in all treatments at the start of veraison. The differences in the plateau of berry growth between the treatments suggest that the berries have a greater ability to expand in PRD1 and PRD2. TSS accumulation (Figure 12B) largely reflected changes in berry weight, with bigger berries accumulating sugar at a slower pace, resulting in delayed ripening in PRD1 and PRD2. This delayed ripening should not be considered a negative effect, as it has been shown to counteract the uncoupling between sugars and secondary metabolites in grapes (Previtali et al., 2021a). In fact, strategies to delay ripening are an important subject of study in the current viticultural research space (Previtali et al., 2021b). Due to the delayed maturation process, both PRD treatments were harvested one week later than CTRL and SSI, to obtain fruit at comparable TSS levels (1st harvest – DOY 264; 2nd harvest – DOY 270).

Balancing yield and quality is a primary goal for grape growers. In the case of wine grapes, it is still the case that growers are remunerated based on tonnage, except for premium wine regions where quality is monetarily recognized through grade scales. In our study, fruit composition was assessed on a weekly basis from 20 °Brix leading up to harvest (Figure 14). The most significant differences were once again observed between both PRD treatments and CTRL and SSI, primarily impacting phenolic compounds. Anthocyanins – responsible for red wine color and a key driver of consumer acceptance in cv. Cabernet Sauvignon (Dadmun & Mansfield, 2023; de Freitas et al., 2017) – quercetin glycosides and polymeric tannins – both responsible for mouthfeel-like sensations (Gutiérrez-Escobar et al., 2021) – were all present at lower concentrations in PRD1 and PRD2 treatments. However, these concentrations were still more than acceptable for cv. Cabernet Sauvignon standards in the region. Weekly measurements showed that there was a slower accumulation of these compounds. This may be partially due to differences in berry moisture levels, which indicates that the CTRL and SSI

treatments were more prone to berry dehydration. There is little work on the effect of PRD on the development of aroma compounds in grapes (Romero et al., 2019).

Research has shown that changes in the canopy microclimate are closely related to variations in fruit composition, such as anthocyanins, phenols, titratable acidity, and TSS (Poni et al., 2018). Increased canopy growth affects the structure of the canopy and its microclimate, which subsequently impacts light penetration within the canopy (Dokoozlian & Kliewer, 1996). Clusters exposed to direct sunlight (compared to canopy-shaded clusters) generally exhibit higher levels of TSS and phenolic compounds. Conversely, they tend to have reduced titratable acidity levels, malic acid, pH, and berry weight (Dokoozlian & Kliewer, 1996). Previous attempts at PRD have linked the benefits of PRD with better fruit composition, proposing that quality improvements were mediated by increased sunlight exposure. Stoll (2000) reported that fruit from PRD-treated vines showed a higher titratable acidity and similar TSS to control vines, together with better light penetration into the canopy, which was correlated with increased anthocyanin and phenolic content in berries. dos Santos et al. (2007) showed in cv. Moscatel that, with the same amount of water applied, PRD was more effective than DI at reducing vegetative growth by achieving more open canopies. This resulted in a cluster microclimate conducive to a higher concentration of phenolic compounds and flavor precursors without any yield reductions.

Most discrepancies with our results should be once again attributed to the different experimental approach, including inconsistencies between standard or different water amounts in PRD and control, different irrigation rates, and variable effects at the physiological level. It should still be noted that in our study, canopies were increased under PRD due to water relocation, as discussed above. Enhancing vine capacity seemed to have large positive effects on yield; however, we confirm that in our study, larger canopies negatively impacted quality, as previously reported by Bergqvist et al. (2001); Kliewer and Dokoozlian (2005). Several management strategies may be implemented to restore quality levels equal to those in the CTRL. First, harvest could have been further delayed to increase hang time and allow controlled dehydration of the fruit for a balanced result in terms of yield and quality. Specific and appropriate canopy management, including trimming and defoliation, could be applied to improve fruit microclimate in PRD vines, given all viticultural practices were executed uniformly across the season for this initial trial.

3. How does this new PRD approach affect water use efficiency at the intrinsic (WUE_i) and agronomic (WUE_c) levels?

The first and foremost reason to study PRD is the potential increase in WUE arising from its application, which is more attractive than ever due to water scarcity and irrigation regulations arising from the challenges of drought. Interestingly, PRD in grapevines has only been truly successful under controlled conditions (e.g., Poni et al. (2009); Stoll (2000)), where WUE_i is often rapidly and positively impacted, and WUE_c has been shown to increase by up to 85% (Stoll, 2000). PRD applications in the field have always encountered some difficulties. Some field studies reported significant increases in WUE in PRD-treated vines (Conesa et al., 2015; Romero et al., 2016b), while others found no consistent improvements in WUE (De Pau et al., 2011; Intrigliolo & Castel, 2009).

In this paper, we describe a novel approach to PRD that aims to bridge the gap between successful results in controlled conditions and several failed attempts to apply this technique at a more commercial level. When we examine WUE_i and its relationship with ψ_{stem} , no significant effects of PRD1 were directly observed (Figure 9). While some transient effects were found, they were not consistent across the season. It should be noted that water deficit itself can cause a transient improvement in WUE_i , and as such, this effect should be considered mixed in the outcome of applying PRD as a half or a fraction of the water of the control, as seen in other studies (Poni et al., 2007; Poni et al., 2005). The lack of significant findings regarding PRD1 can be explained by a variety of hypothetical reasons. First, the effect was not strong enough; however, this is highly unlikely considering the drying patterns and flow data presented earlier in this study. It is also unlikely that the effect was not captured considering the high frequency of measurements, repeated at multiple times of the day and several days a week.

Certainly, differences in stomatal regulation and carbon fixation in response to changes in water potential point to important differences between treatments, particularly for PRD1 and PRD2 (Figure 7 and Figure 8). PRD2 was generally more stressed (more negative ψ_{leaf}), and stomata quickly reacted to decreases in water availability. At the same time, PRD1 had a much narrower range ψ_{leaf} , and g_{SW} was maintained at a fairly high level, even under more limiting water conditions (when sometimes ψ_{leaf} was higher than in PRD2). It is possible that the response of PRD1 may be even larger under stricter water availability, given the low range of ψ_{leaf} , and the response of PRD2 be worse.

Although we did not describe any direct effects in terms of improved WUE_i , the preliminary application of PRD in conjunction with subsurface irrigation described in this research study resulted in positive improvements to WUE_c . Also, as described before in Medrano et al. (2015b) and Tomás et al. (2014b), research has shown that in plants with complex canopies, like grapevines, single leaf instantaneous measurements frequently don't accurately reflect whole plant photosynthesis and often WUE_i lacks correlation to WUE_{plant} or WUE_c . The final output captured using WUE_c encompasses several factors spanning multiple seasons. We reported a large increase in yield, stemming from a potential spatial redistribution and increased density of roots when applying PRD treatments for the second year on the same vine. Linking intrinsic patterns to final outputs should remain a goal for this project, as well as validating the mechanism underlying the positive effects detected in preliminary observations. Adaptation to the new conditions is expected over time, therefore, this study should be repeated over several years to fully characterize the effectiveness of this novel PRD approach.

5. Conclusion

In this study, we revisited the application of Partial Rootzone Drying (PRD), a technique known for its potential to save irrigation water and enhance water use efficiency (WUE) without compromising fruit yield and quality. While PRD has shown promise in controlled environments, it has faced implementation challenges when applied in the field.

The aim of this work was to revise the way PRD is applied by combining it with the principles of subsurface irrigation to maximize the separation between the irrigated and non-irrigated zones within the root system. Four treatments were trialed: Conventional Drip Irrigation (CTRL), Subsurface Irrigation (SSI), and two distinct PRD treatments, Partial Rootzone Drying Alternated (PRD1) and Partial Rootzone Drying One-Side (PRD2).

Our first objective was to determine whether our novel approach to PRD was sound and robust enough that it could replicate the benefits observed in controlled environments when applied at a commercial scale. Soil moisture data and irrigation data showed we were successful. By relocating the drip lines to the middle of the interrows and burying them underground, and through a precise irrigation system, we were able to meet PRD's two key criteria: first, the soil water content must reach a minimum threshold to trigger a physiological response in the plant; second, there must be a clear distinction between the "wet" and "dry" rootzones.

Secondly, we addressed whether there were benefits of applying our revised PRD on vine performance, fruit yield, and quality. Fractional cover (FC) data, corroborated with pruning weight data (PW), revealed that the canopies in the PRD treatments were already larger at the beginning of the study, likely due to carry-over effects from the previous year's adaptation to PRD. This bigger canopy size suggests that the relocation of water subsurface and into the interrows stimulated root growth and led to an overall larger root system, helped by the cool and rainy weather in 2023.

When we measured gas exchange parameters and water status in a diurnal fashion, we found that the relationship between stomatal conductance (g_{sw}) and leaf water potential (ψ_{leaf}) during the morning suggested differences in how stomata regulate water use in response to water status when PRD was implemented and the "wet" and "dry" zones were switched based on soil moisture data, as seen in the PRD1 treatment. The same trend was found between CO_2 assimilation (A) and ψ_{leaf} . These differences at the physiological level point to added benefits achieved by alternating irrigation sides, which deserve to be further researched to identify the underlying mechanisms.

Yield components and crop load were improved or maintained in PRD vines. PRD treatments exhibited a large yield increase when compared to CTRL, especially in PRD1. Berry weight data was consistent with the overall yield increase. The effect of the treatments in these parameters was clearly dependent on canopy size. Crop load, characterized using the Ravaz Index (yield/PW), was not significantly different between the treatments and was within the optimal range for the variety and site selected, which explains the good level of quality achieved across all treatments.

Regarding grape composition, the increased canopy growth and berry size in both PRDs primarily led to delayed ripening and lower concentrations of the measured phenolic compounds (anthocyanins, quercetin glycosides, and polymeric tannins). This is most likely attributed to a change in the vine microclimate due to the larger canopies, reducing cluster sunlight exposure. Although this is a negative effect, we have discussed viticultural practices that a concerned grower could apply to counteract detriments to quality. Large increases in yield would have a much bigger positive effect on grower remuneration in the current situation at the trial location.

The third and final objective was to try to understand how this novel PRD approach affected WUE, the main reason for implementing PRD in the first place. We evaluated PRD effects on water use efficiency both at the intrinsic (WUE_i) and agronomic (WUE_c) levels. Our findings revealed no significant changes in WUE_i , which were transient and occasional throughout the season. However, WUE_c was largely increased, and this increase stemmed from increasing yield without additional water input.

Based on our findings, continuing to work on PRD is strongly justified, as the study clearly demonstrated successful implementation at a commercial scale, with substantial increases in yield and WUE_c without additional water input and acceptable quality. It shows a lot of potential for long-term benefits, and it has room for innovation. While our study is worth pursuing further, some areas require deeper investigation to fully understand and optimize this novel PRD approach.

Effects on WUE_i are not fully understood, and future research should focus on verifying the effects at the hormonal level, such as the role of abscisic acid (ABA) and other signaling molecules in stomatal regulation and root-to-shoot communication. Investigating gene expression changes in leaves and roots could provide insights into the molecular responses driving the observed physiological effects. Exploring root growth patterns and distribution using root mapping techniques or imaging technologies would help verify our hypothesis that the PRD treatments largely changed root growth and allocation. Certainly, conducting multi-year studies would be essential to assess the long-term effects and adaptability of vines to this irrigation method, and this would determine whether the benefits observed are sustained over time and under varying climatic conditions, as well as any potential cumulative effects. Various suspicions arose during the trial that could potentially be clarified under different climatic conditions, such as PRD2 during a warmer, drier year, which we suspect would perform very differently and “crash” due to water stress. The SSI application prototype could also benefit from several improvements, such as the angle at which the PVC emitter is buried. And while it may still be too early, a somewhat comprehensive cost-benefit analysis could provide some good insight into further implementation at scale.

Overall, this study represents an important milestone in refining irrigation practices for commercial vineyards, offering a potential strategy to save water while maintaining crop performance. However, further work is needed to fully verify the long-term benefits and understand the underlying processes that govern WUE in these complex systems.

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Appendices

Appendix A

Table 7 – Occurrence of the main phenological stages and vineyard viticultural practices at the Cabernet Sauvignon experimental vineyard, in 2023.

	Date of occurrence
Phenological Stages	
Budburst	10/04/2023
Flowering	1/06/2023
Veraison	2/08/2023
Harvest	21/09/2023 and 27/09/2023
Viticultural Practices	
Mechanical leafing (north row side only)	19/06/2023
Mechanical trimming	Mid-July
Mechanical trimming (before commercial harvest)	3 rd week of September

Appendix B

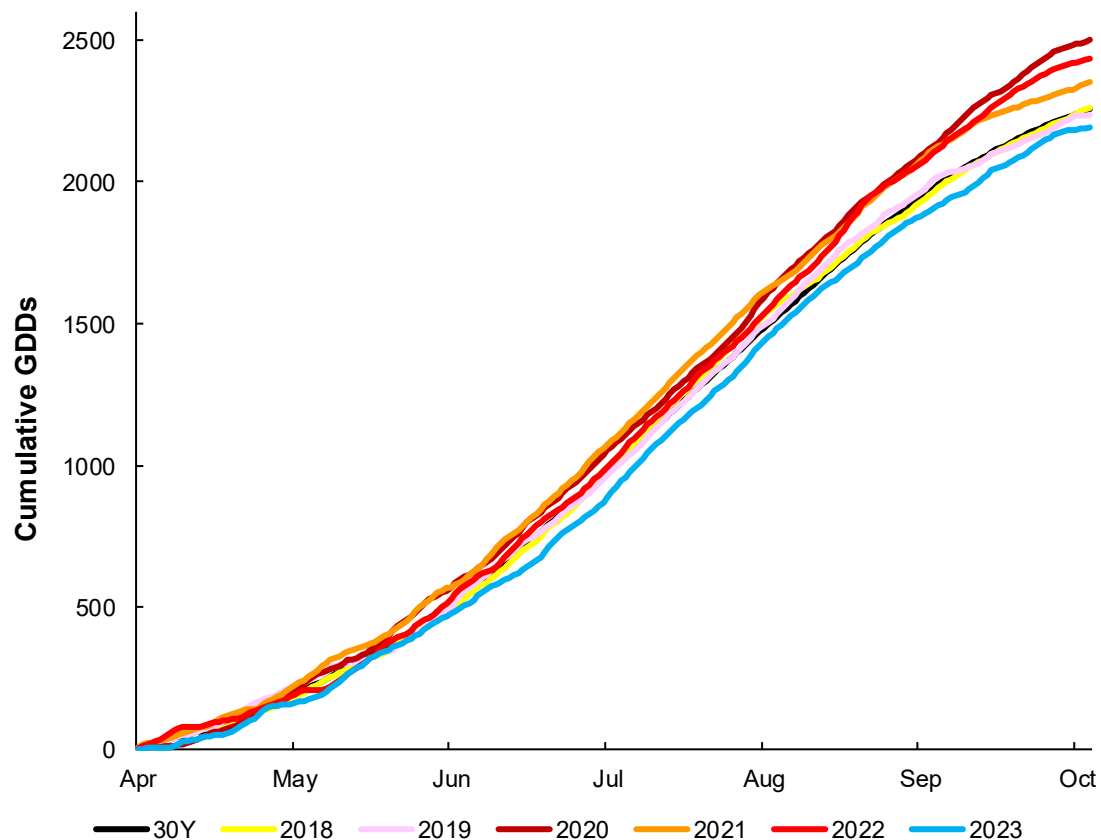


Figure 15 - Comparison of seasonal heat accumulation as growing degree days (GDDs) in six consecutive seasons and the 30-year normal. GDDs are calculated from April 1 (DOY 91) to October 31 (DOY 304).

Appendix C

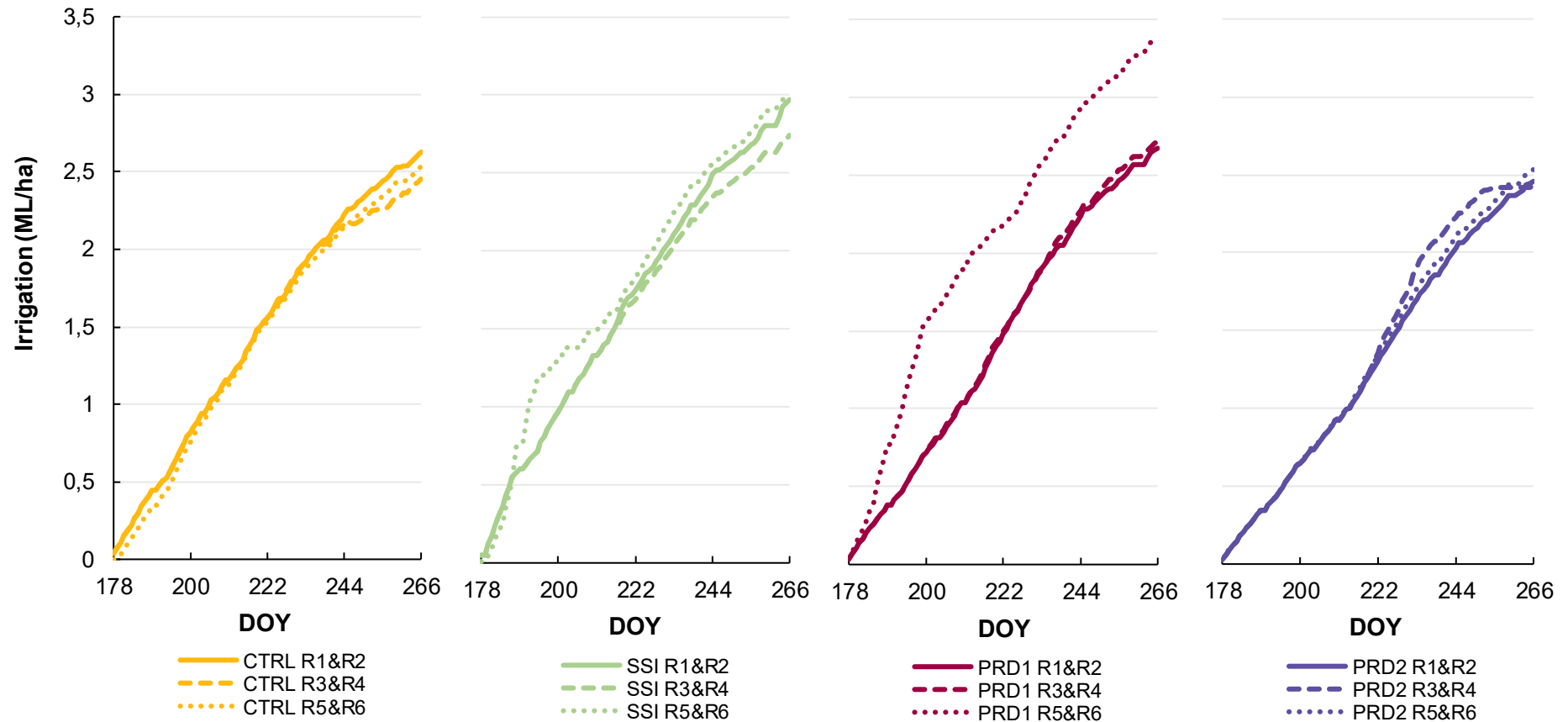


Figure 16 – Cumulative irrigation (ML/ha) of four different irrigation regimes applied throughout the 2023 season to Cabernet Sauvignon vines: CTRL (control irrigation); SSI (subsurface irrigation); PRD1 (partial rootzone drying alternated); PRD2 (partial rootzone drying one-side). Different line types represent different replicates. Irrigation amount was tracked through flowmeters and pooled in three groups of two replicates each. Irrigation started June 27 (DOY 178) and ended September 23 (DOY 266).

Appendix D

A. pH

	DOY					
	235	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>b</i>	<i>a</i>	<i>a</i>	<i>a</i>
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD1	<i>a</i>	<i>a</i>	<i>c</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>ab</i>	<i>b</i>	<i>a</i>	<i>a</i>

B. Titratable Acidity

	DOY					
	235	242	249	256	264	270
CTRL	<i>a</i>	<i>b</i>	<i>ab</i>	<i>a</i>	<i>b</i>	<i>a</i>
SSI	<i>a</i>	<i>a</i>	<i>b</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD1	<i>a</i>	<i>b</i>	<i>a</i>	<i>a</i>	<i>b</i>	<i>a</i>
PRD2	<i>a</i>	<i>b</i>	<i>b</i>	<i>a</i>	<i>b</i>	<i>a</i>

C. NOPA

	DOY					
	235	242	249	256	264	270
CTRL	<i>b</i>	<i>a</i>	<i>b</i>	<i>a</i>	<i>ab</i>	<i>a</i>
SSI	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>	<i>c</i>	<i>a</i>
PRD1	<i>ab</i>	<i>a</i>	<i>b</i>	<i>a</i>	<i>bc</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>

D. Malic Acid

	DOY					
	235	242	249	256	264	270
CTRL	<i>a</i>	<i>ab</i>	<i>a</i>	<i>a</i>	<i>b</i>	<i>b</i>
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD1	<i>a</i>	<i>b</i>	<i>a</i>	<i>a</i>	<i>b</i>	<i>b</i>
PRD2	<i>a</i>	<i>b</i>	<i>a</i>	<i>a</i>	<i>b</i>	<i>b</i>

E. YAN

	DOY					
	235	242	249	256	264	270
CTRL	<i>b</i>	<i>a</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>
SSI	<i>b</i>	<i>a</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>
PRD1	<i>b</i>	<i>a</i>	<i>a</i>	<i>b</i>	<i>b</i>	<i>ab</i>
PRD2	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>

F. Tartaric Acid

	DOY					
	235	242	249	256	264	270
CTRL	<i>a</i>	<i>ab</i>	<i>a</i>	<i>a</i>	<i>ab</i>	<i>a</i>
SSI	<i>a</i>	<i>a</i>	<i>b</i>	<i>b</i>	<i>a</i>	<i>a</i>
PRD1	<i>a</i>	<i>b</i>	<i>a</i>	<i>c</i>	<i>b</i>	<i>a</i>
PRD2	<i>a</i>	<i>b</i>	<i>ab</i>	<i>ab</i>	<i>b</i>	<i>a</i>

G. TSS

	DOY					
	235	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
SSI	<i>ab</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>
PRD1	<i>b</i>	<i>b</i>	<i>b</i>	<i>bc</i>	<i>b</i>	<i>ab</i>
PRD2	<i>b</i>	<i>b</i>	<i>b</i>	<i>c</i>	<i>b</i>	<i>b</i>

Figure 17 – Temporal progression of basic biochemical parameters (**A – G**) of Cabernet Sauvignon juice during ripening. Samples collected in the later stages of ripening (DOY 235 – 270) of the 2023 season. Tables indicate the significant differences among treatment means for each sampling date according to a linear mixed model with one fixed effect (treatment) and one random factor (replicate), followed by Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).

Appendix E

A. Moisture

	DOY				
	242	249	256	264	270
CTRL	<i>b</i>	<i>b</i>	<i>c</i>	<i>c</i>	-
SSI	<i>b</i>	<i>b</i>	<i>bc</i>	<i>bc</i>	-
PRD1	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>ab</i>	<i>ab</i>	<i>a</i>

B. pH

	DOY				
	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>

C. Malic Acid

	DOY				
	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>c</i>	<i>b</i>	-
SSI	<i>a</i>	<i>a</i>	<i>ab</i>	<i>a</i>	-
PRD1	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>bc</i>	<i>a</i>	<i>a</i>

D. Titratable Acidity

	DOY				
	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>c</i>	<i>a</i>	-
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>a</i>	<i>a</i>	<i>ab</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>bc</i>	<i>a</i>	<i>a</i>

E. YAN

	DOY				
	242	249	256	264	270
CTRL	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>	-
SSI	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>	-
PRD1	<i>a</i>	<i>a</i>	<i>ab</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>

F. β -damascenone

	DOY				
	242	249	256	264	270
CTRL	<i>a</i>	<i>c</i>	<i>a</i>	<i>a</i>	-
SSI	<i>a</i>	<i>bc</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>a</i>	<i>ab</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>

G. IBMP

	DOY				
	242	249	256	264	270
CTRL	<i>ab</i>	<i>b</i>	<i>a</i>	<i>a</i>	-
SSI	<i>b</i>	<i>a</i>	<i>a</i>	<i>b</i>	-
PRD1	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>

H. C₆ Compounds

	DOY				
	242	249	256	264	270
CTRL	<i>b</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
SSI	<i>b</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>
PRD2	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>

I. 1-octen-3-ol

	DOY				
	242	249	256	264	270
CTRL	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>	-
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>
PRD2	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>

J. Total anthocyanins

	DOY				
	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>b</i>	<i>b</i>	<i>a</i>	<i>b</i>	<i>a</i>
PRD2	<i>b</i>	<i>b</i>	<i>a</i>	<i>b</i>	<i>a</i>

K. Quercetin glycosides

	DOY				
	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>
PRD2	<i>b</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>

L. Polymeric tannins

	DOY				
	242	249	256	264	270
CTRL	<i>ab</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
SSI	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
PRD1	<i>bc</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>
PRD2	<i>c</i>	<i>b</i>	<i>b</i>	<i>b</i>	<i>a</i>

M. TSS

	DOY				
	242	249	256	264	270
CTRL	<i>a</i>	<i>a</i>	<i>a</i>	<i>a</i>	-
SSI	<i>a</i>	<i>a</i>	<i>ab</i>	<i>ab</i>	-
PRD1	<i>b</i>	<i>b</i>	<i>c</i>	<i>c</i>	<i>a</i>
PRD2	<i>b</i>	<i>b</i>	<i>bc</i>	<i>bc</i>	<i>a</i>

Figure 18 – Weekly maturity level chemical analysis (**A – M**) of Cabernet Sauvignon cluster samples during the later stages of ripening (DOY 242 – Week 1; DOY 249 – Week 2; DOY 256 – Week 3; DOY 264 – Week 4 and WL; DOY 270 – WL) of the 2023 season. Tables indicate the significant differences among treatment means for each sampling date according to a linear mixed model with one fixed effect (treatment) and one random factor (replicate), followed by Fisher's Least Significant Difference (LSD) test at the 95% confidence level ($p < 0,05$).