



**Effect of deficit irrigation and training system on the
physiological, agronomic and quality parameters of
Touriga Franca grapevine (*Vitis vinifera* L.)**

Vitor Gonçalves Furline

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Supervised by

Antonio Castro Ribeiro

Américo Wágner Júnior

David Santos Barreales

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*"Our greatest glory is
not in never falling, but in
rising every time we fall"
(Confúcio 551 b.C).*

Dedication

I dedicate this work to my parents Antonio Claudio Furline and Denise Gonçalves Furline and my sister Leticia Gonçalves Furline, who have always supported me throughout my journey, for support, be it emotional or financial since I decided to follow my dream and leave home away to study, more than 1100 km from home, and supporting me since 2013.

In the middle of the way, I stayed away from my family and childhood friends and for several moments I thought about giving up, but they always supported me to continue and become a good person, I learned to manage by myself, cry alone, fight alone, make friends, lose friends, and mature a lot. I also ended up halfway through losing my grandmother Virginia Nogueira Furline, who I'm sure is seeing me from somewhere, and also dedicating my other grandmother Maria Luiza Rente de Lima for always remembering me and being happy when I come home for my courage and willingness to move on, despite the difficulties.

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Only I know how difficult it is to be away from home, breaking my face, and struggling every day, I often need to suffer in silence so as not to worry anyone, and I would also like to dedicate this to myself Vitor Gonçalves Furline, and say that yes, I am a winner for getting here, and many achievements will still come with a lot of work, I have no doubt about that.

To all these people I want to say thank you and wait for me, that soon I will be reaping the fruits of all this work and sacrifice, I love you very much.

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Abstract

The increase in the frequency and intensity of extreme weather events is one of the main effects of climate change, with particularly negative consequences for the development of agricultural activity. As water is one of the most limiting factors in yield, irrigation management is a critical factor for the future in a context of water scarcity. This study aims to implement three irrigation regimes (irrigation deficit with 25% and 50% of crop evapotranspiration (ET_c) and full irrigation (100% ET_c)) and two pruning systems (unilateral Cordon and Guyot) and evaluate their effects on physiological, agronomic and quality parameters in the Touriga Franca (*Vitis vinifera* L.) variety, in a commercial vineyard located in the Douro region. Field studies were carried out during 2019 in a commercial organic vineyard, located at Quinta do Romeu (41°53'02''N, 7°08'00''W, 326 m altitude) in the Northeast of Portugal. Agronomic, physiological and grape quality parameters were evaluated in the different treatments. The results showed that the plants that received the highest amount of irrigation were the ones that showed the best physiological and agronomic performance. In most of the quality parameters of the musts, no differences were observed between treatments, except for the total soluble solids (TSS) content. The deficit irrigation treatment (25% ET_c), pruned in Cordon, showed the lowest values, while the 100% ET_c full irrigation treatment pruned in Guyot was the one with the highest values. The results presented are from the first year of trials in a study that will take place over several years. Thus, the conclusions presented are still preliminary.

Keywords: *viticulture; Vitis vinifera* L.; *water use efficiency; pruning system.*

Resumo

O aumento da frequência e intensidade dos eventos climáticos extremos é um dos principais efeitos das alterações climáticas, com consequências particularmente negativas no desenvolvimento da atividade agrícola. Sendo a água um dos fatores mais limitantes da produção, a gestão da irrigação é um fator crítico para o futuro num contexto de escassez de água. Este estudo tem como objetivo implementar três regimes de irrigação (deficit de irrigação com 25% e 50% de evapotranspiração da cultura (ETc) e plena irrigação (100% ETc)) e dois sistemas de poda (cordão unilateral e Guyot) e avaliar seus efeitos em parâmetros fisiológicos, agronômicos e de qualidade na variedade Touriga Franca (*Vitis vinifera* L.), numa vinha comercial localizada na região do Douro. Os estudos de campo foram realizados durante o ano de 2019 em uma vinha orgânica comercial, localizada na Quinta do Romeu (41°53'02''N, 7°08'00''W, 326 m altitude) no Nordeste de Portugal. Foram avaliados parâmetros agronômicos, fisiológicos e de qualidade da uva nos diferentes tratamentos. Os resultados mostraram que as plantas que receberam maior dotação de rega foram as que apresentaram um melhor desempenho fisiológico e agronômico. Na maioria dos parâmetros de qualidade dos mostos não se observaram diferenças entre os tratamentos com exceção do teor em sólidos solúveis totais (TSS). O tratamento de rega mais deficitária (25%ETc) podado em cordão foi o que apresentou os menores valores, enquanto o tratamento de rega plena 100%ETc) podado em Guyot foi o que apresentou valores mais elevados. Os resultados apresentados dizem respeito ao primeiro ano de ensaios do estudo que vai decorrer ao longo de vários anos. Assim, as conclusões apresentadas são ainda de carácter preliminar.

Palavras-chave: *viticultura; Vitis vinifera* L.; *uso eficiente da água; sistema de poda.*

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Abbreviations

DDR – Demarcated Douro Region
ET_c - Cultivation evapotranspiration
ET₀ – Reference evapotranspiration
ha – Hectare
RDI – Regulated deficit irrigation
PRD – Partial Rootzone Drying
FAO – Food and Agricultural Organization
°C – Degree Celsius
CCI - Chlorophyll Concentration Index
SPAD - Soil and Plant Analysis Development
IRGA - Infrared Gas Analyzer
E – Transpiration rate
g_s – Stomatal conductance
A – Net photosynthetic rate
A/g_s – Intrinsic efficiency of water use
PQA – Point Quadrat Analysis
NLS - Number of layers of the sheet
%IL - % of Inner Leaves
%IC - % of Inner Clusters
%H - % of Holes
TF – Touriga Franca
TFC – Touriga Franca Cordon Royat
TFG – Touriga Franca Guyot
TF25 – Touriga Franca 25% ET_c
TF50 – Touriga Franca 50% ET_c
TF100 – Touriga Franca 100% ET_c or ET₀
Ψ – Water potential
ton – Tonnes
TPI – Total Polyphenol Index
TSS – Total Soluble Solids

1. INTRODUCTION

Wine is a very old drink and it has a very important function in almost all civilizations, being a drink with symbologies, either mysticism or religious, so the cultural importance of viticulture makes it a very popular and cultivated drink all over the world because of tasty and nutritious fruit. In Portugal, the vine was brought by the Romans at the expansion of the Roman Empire and it was disseminate throughout Portugal. In 1865 the phylloxera plague caused major problems in production and then new technologies began to be developed (IVV, 2016).

Portuguese viticulture production has reached about 5,900,000 hectoliters, representing 53.2% (approximately 1,542 million of Euros) of the total sales value in the beverage industry (INE, 2019). The regions of Douro, Alentejo and Lisbon are those with the highest incomes in the country (Wines of Portugal, 2019).

The Douro Demarcated Region (DDR) is located in Northeastern Portugal, in the Douro River Basin, with a total area of 250,000 hectares. Of this total area, 45,000 hectares are vineyards that extend along the Douro River, it being the main economic activity. The DDR is divided into three sub-regions (Baixo Corgo, Cima Corgo, Douro Superior) (IVDP, 2010).

Climate change has been an important point, as the average annual temperature increased during the years, with warmer than normal days, and the precipitation rate lower, which it makes the drought more intense and frequent, with the increase in the desertification areas (IM, 2010). The increase of the frequency and intensity of extreme weather events is one of the main aspects of climate change, with particularly negative effects on the agricultural activity. As water is the main limiting factor of yield, the irrigation is seen as a strategic element for climate change adaptation (MAMAOT, 2013).

The climate has a strong influence on the vine, with effect on the potentialities of the regions for the crop. It interacts with other components of the natural environment, in particular with the soil, as well as with variety and vine cultivation techniques (EMBRAPA, 2003).

Observing and assessing the impacts of climate change can help the viticulture sector adapt to future scenarios, because there are companies that explore new areas in cooler locations to produce a more balanced wine. The choice for resistant grape varieties is also important because high temperatures affect the color of the wine, as well as yield (Lopes, 2019).

Irrigation is a method of water management to improve the productive capacity of grapevines. Plant reproductive growth is less sensitive than vegetative growth, but the impact of lack of water will depend on the water shortage (Keller, 2005).

The grapevine is a plant that needs support to obtain better yield, providing vigorous plants, better aerial distribution that facilitates the entry of light to perform photosynthesis and better aeration of the leaves. The driving systems facilitate adaptation to the topography, in addition to facilitating the locomotion of producers in the vineyard (EMBRAPA, 2003).

In this sense, the present work had as main focus to understand the effect the of pruning system and irrigation in the productivity vineyard and fruit quality. With this, it can be evaluated if there is influence of the pruning system and if the irrigation can improve the productivity and quality of the vines.

1.1. Objectives

This study aims to implement water management techniques to evaluate its influence on Touriga Franca (*Vitis vinifera* L.) varieties in productive, physiological, biochemical and viticultural aspects in a vineyard in the Douro Superior region, by:

- ✓ Stress management with the adoption of irrigation methods, through 25% and 50% of reference evapotranspiration (ET_c) and one with total irrigation (100% of ET_c).
- ✓ Comparison of two pruning systems (Guyot and Royat Cordon unilateral).

2. INFLUENCE OF CLIMATE CHANGE IN THE VITICULTURE OF MEDITERRANEAN AREAS

The Demarcated Douro Region (DDR) is important from the point of view of wine and tourism, it being situated in the Northeast of Portugal, in the Douro river basin. This region has particular climatic characteristics, with total area of about 250.000 hectares (Loureiro, 2017). The same author says that in the Douro Superior, the climate is typically Mediterranean, with arid characteristics, often leading to early maturation of the grapes.

The production of grapes in Douro region is subject to a strong water deficit with the difference reaching between 730 and 750 mm per year between evapotranspiration and precipitation during the sprouting and harvest (IVDP, 2010).

Douro Superior is the most important sub region, because it has area of 110 thousands of hectares and 10.190 hectares of vineyard. This area has less decline when compared to other sub regions in Douro, allowing use of irrigation decreasing plant stress to improve the yield (Loureiro, 2017).

The importance of Portuguese viticulture is a reality of the point of view of economic, social, environmental and cultural. The Douro region (includes the Denominations of Origin Douro and Porto) is the oldest demarcated region of the world to be regulated by law and it has been designated by UNESCO World Heritage Site since 2001. In addition, it is currently the main wine-growing region of the country in terms of wine production and vineyard area (150 million liters and 45 000 ha). Of the total volume of wine produced in the Douro Demarcated Region, about 50% of the wine is intended for the production of "Porto Wine", while the remainder is for production using the controlled designation of origin "Douro" or " Douro Wine " (IVV, 2015).

According to the Koppen classification, the Douro region belongs to the Csa climate (C - temperate climate, s - winter rain, a - hot summer). This region has a temperate climate, with rainy winters. Its average monthly temperatures in the coldest months vary between -3°C and 18°C and summers are hot and dry with average monthly temperatures that can be equal to or greater than 22°C (Kottek et al., 2006).

The climate is characterized by a difference of radiation, temperature, relative humidity, precipitation, wind, altitude throughout the years and that seasonal variation may develop changes in water balance (Malheiro et al. 2010). The climate conditions are an important factor to considering in the wine production causing physiology

disturbs from region to region and from year to year. These differences may lead highs and lows in wine prices (Van Leeuwen et al. 2004; 2007). Temperature is the fundamental factor to control the phenological development phases of the vines, what it may to control the ripeness (Van Leeuwen et al., 2008).

The global warming predictions suggested a reduction in rainfall and increase of evapotranspiration for the Europe in throughout years (IPCC, 2007). This may suggest that grape varieties are adopting to cool temperate regions and consequently, it may lose some specific qualities (Horacio García and Díaz Fierros, 2009). Also affecting grapevine development such as phenological modifications and harvest anticipation (Webb et al., 2007). Winegrowers has been concerned about the impact on production due to water deficit and the safety of yields (Gouveia et al., 2012).

The climatic conditions affect the yield and quality more than another environmental factors such as grape variety and soil type. The increase of temperature in Mediterranean regions and advanced phenology cause changes in grapes metabolite, such as lower acidity, increase of alcohol levels, greater sugar accumulation and variables effects in secondary metabolites (Van Leeuwen et al., 2016). The climatic data evaluation is very important for the choice of variety. This is how you can know which varieties are best suited to a particular region, thus improving the chances that the chosen variety has better yields and qualities (Van Leeuwen and Saguin, 2006).

The balance of the vines is given by the reproductive and vegetative growth under ideal conditions for the cultivate, adequate yield and desired quality, namely sugar levels, acid balance and flavor compounds. Environmental factors and vegetative material may influence the potential growth of the vineyard. Management methods are a secondary factor for productivity in any area. Thus, these factors always considered with the irrigation and fertilization as determinants of growth and plant balance (Skinkis and Vance, 2013).

Climate change caused changes in production scenarios and it may cause production changes in colder areas with the use of new varieties and rootstocks adapted to drier and warmer conditions and changes in soil management (Lopes et al., 2011). Several companies around the world recognize the lower water use is part of the new corporate environmental strategy (Clothier et al., 2010).

Temperature changes increase water vapor levels in the atmosphere to make water availability less predictable. In dry regions, it may have a drought aggravation, especially in the summer months. According to a report by the European Environment

Agency, humid regions tend to be more frequent and intense, while dry regions extremely dry. Some adaptation measures target efficient water management in certain water-intensive sectors, such as agriculture by implementing a range of sustainable farming techniques such as drip irrigation to reduce water consumption (EEA, 2018).

Portugal consumes more water than its availability, therefore, they should not use irrigation in areas of water scarcity and where efficient water use systems are not guaranteed. “Drought” is continually confused with “scarcity” and the government decisions that have been taken are proof of this, repeating the same drought response measures year after year when preventive measures should be in place (WWF, 2019).

Since the 1950s, the economic costs caused by natural catastrophes such as excessive temperature changes. This is a factor that needs a lot of attention because agriculture plays an important role in people's diet (fiber and energy) as it is the main livelihood of 70% of the world's poor. (UNRIC, 2009).

The climate forecast for 2050 say the temperature will rise between 1 and 2 degrees Celsius depending on the amount of greenhouse gases that will be released. As a result, rainfall and temperature distribution will differ between wetlands and drylands, with the duration and intensity of droughts likely to increase in regions such as the Mediterranean basin, Central Europe, Central America, northeastern Brazil and South Africa (De Castro and Riviera, 2014).

Future projections for the viticulture regions have shown that changes will continue to appear and it may compromise the cultivation of vineyards without adaptation measures being made. The particular attention of these changes will be on issues of adapting irrigation systems, selecting rootstocks and grape varieties. It will be necessary to study which will be the most suitable for a given territory, as only those who adapt in advance to these climate changes will be able to profit from viticulture activities. (ADVID, 2019).

The main cause of reduced vine growth and productivity in the regions Mediterranean climate is water stress, more or less exacerbated by light and thermal stress due to the effect on the balance between the growth of vegetative and reproductive organs (Geerts and Raes, 2009; Chaves et al., 2010; Malheiro et al., 2011; Alves et al., 2011).

The use of water in the vineyard is a function, among other factors and usable water, rootstock/variety and selected conduction. In semi-arid or arid zones, moderate

to severe drought periods, may be controlled using high-efficiency practices such as regulated deficit irrigation (Intrigliolo and Castel, 2008).

Agronomic practices as well as weather conditions (light, rainfall and temperature) may significantly affect the biochemical composition of grape berries (Teixeira et al., 2013). Temperature is a factor that can drastically affect grape quality by affecting polyphenol metabolism, with high temperatures negatively affect berry coloration (De Rosas et al., 2017).

2.1. Irrigation strategies

Water utility may have different meaning to different people and within groups (agriculture water, forestry and fishery, industrial and domestic uses). (Dugan et al., 2006; Wesseling and Feddes, 2006). For the crop production the volume of water available interferes directly with respiration, transpiration and evaporation in the life cycle of plants (Ali et al. 2008; Barreales et al., 2018).

Water stress is a major problem due to the lack of growth of a vine, being a factor that alters the vegetative and physiological balance of plants (Malheiro et al., 2011). The scarcity of water in the soil can have negative impacts on the growth of plants, making irrigation practice to reduce water stress, especially in semi-arid regions (Teixeira et al., 2013). Total irrigation of areas is not recommended for vines, as it negatively affects the quality of the grape, increasing the berries and affecting the ratio between the pulp and the skin (Ruiz-Sanchez et al., 2010).

Irrigation is mainly used in Portugal in the Alentejo and Douro regions. This use increase year after year by rising global average temperatures, limiting the profitability of yield in more arid regions. Another fact that has led to increased use of irrigation is the competitiveness of the Portuguese market with other European markets, making producers produce more and a better quality wine (Loureiro, 2017).

Different strategies of water application through irrigation were developed with the common objective of controlling vegetative growth, fruit development and increase the efficiency of water use by the plant. In general terms, these strategies consist to reduce the availability of water in all or part of the production cycle, or all or part of the volume of soil explored by the root system (Rodrigues, 2011).

Chalmers et al. (1981) and Mitchell et al. (1982) proposed in their studies that regulated deficit irrigation (RDI) may be used to manage the vegetative vigor, in addition to improve the yield and prevent large amounts of water resources from being

wasted. RDI consists of applying certain amounts of water, which usually begins to flowering to supply the loss of water from the plant by evapotranspiration (Keller et al., 2008). RDI is a common strategy in dry regions with production goal to adjust vine development and improve fruit quality attributes according to wine style (Romero et al., 2013).

Shellie (2006) and Sousa et al. (2007) conducted studies in the vineyard with the RDI showed that direct benefits in fruit quality (mainly in coloration), airier vegetation wall, less susceptibility to pests and diseases and greater ease in phytosanitary treatments.

Partial Rootzone Drying (PRD), consists of alternating (every two weeks) watering the vine on one side and the other, to ensure that half of the root system is kept in contact with the dry soil and the other half with the moist soil. This technique seeks to stimulate the vine's physiological response to stress through "signals" produced by the roots in contact with the dry soil and transported to the leaves, which induce stoma closure (Stoll et al., 2000). The physiological and agronomic effects of this strategy of irrigation in the vineyard were studied by some authors and when compared to plants in hydrological comfort, it resulted reduced vigor of the plants (Souza et al., 2005) and with leaf reduction by inhibiting the growth of the lateral shoots, thus it give rise to vegetation walls more exposed to solar radiation (McCarthy et al., 2002; Chaves et al., 2007).

Contrary to what happens in RDI where there is only a reduction in production, PRD has no productivity losses. PRD may attribute benefits to fruit quality, especially in the concentration of total anthocyanins and polyphenols (McCarthy et al., 2002; Santos et al., 2005; Chaves et al., 2007). A greater exposure of the bunches positives its quality (McCarthy et al., 2002), due to the thermal and light changes in the fruiting zone (Santos et al., 2005). Other authors stated that the use of PRD in grapevines has little or no improvement in yield and fruit quality when compared to RDI (Lopes et al., 2011; Romero et al., 2014).

Fereres and Soriano (2007) reported that in many crops under RDI it not only significantly increases water productivity but also improves farmers' yield and its efficiency in the use of water showed that it is possible to maintain the yield of vineyards even reducing the use of water in plants and it to maintain or improve the quality of berries (Medrano et al., 2015). In the theory of PRD, the roots are irrigated and with this, it will be induced the hormonal production, called as abscisic acid (ABA), adjusting the plant to open its stomata (Costa et al., 2012). The water relation between water

and grapevine is compared by different irrigation strategies (RDI and PRD), on grape quality and consequently on wine quality, besides their effects on production (Medrano et al., 2015).

The water is important to all organisms in the world and the efficient use of resources such as rainwater for irrigation has become important to increase vineyard production and quality, especially in regions with low water availability throughout the year (Ali et al. 2008). The supply of water by irrigation can prevent overheating of the plants, thereby ensuring the longevity of plants (Chaves et al., 2010).

The water consumption of the vine varies throughout the growing season and it determined by environmental and physiological factors and for all cultural operations that condition the leaf surface size and roof architecture. Generally, in the first month after budging water consumption is low, increasing along the cycle accompanying the growth of leaf area. Since the moment the canopy reaches its maximum development, water consumption remains high during some time and then decreases, what reflect in decrease of evaporative demand atmosphere and the loss of leaf surface resulting from tissue aging and consequent senescence of the leaves (Williams, 2000).

Water is a fundamental element for the vegetative/reproductive growth of the vine and for its biochemical and physiological functioning, it being a determining factor for yield and quality of grapes (Ojeda et al., 2005).

The water regime throughout the production cycle should allow to verify the intensity or severity of the stress that the plant has been suffering, its duration and the moment in which it occurs. This regimen may be evaluated by checking evapotranspiration and physiological variables that are sensitive to water deficit and with these data make a comparison with plants in water status (Rodrigues, 2011).

Although it is widely recognized that water regime is a determinant of the composition of berries, in particular their content in sugars, anthocyanins, tannins, phenolics and acids, the results presented in the literature on the effects of irrigation on quality of musts do not always point in the same direction. It is possible that differences in the observed responses result from different mechanisms of response of the plant to the lack of water that is processed as a function of the moment and intensity of stress imposed (Chaves et al., 2010).

Total acidity at the end of maturity is higher under irrigated conditions (Lissarrague et al., 2007). The highest acid content observed in watered plants is due to a higher concentration of malic acid in the beginning of maturation and, on the other, its lower degradation during maturation (Garcia-Escudero, 2007).

In Europe, the grape production is still some suspicion that irrigation may prevent maturation resulting in less alcoholic, more acidic wines and poor in phenolic components. The truth is that in countries with a predominantly Mediterranean climate, the irrigated vineyard has been able to maintain a reasonable production with excellent quality wines (Loureiro, 2017).

Deficit irrigation has been studied as a valuable and sustainable strategy to use in dry regions to increase and maximize water productivity (Geerts and Raes, 2009). The efficient of the use of water in the wine grapevines to gain in many factors of water productivity, as berry composition, weight and wine quality (Girona et al., 2009; Ortega-Farias et al., 2012; Aragüés et al., 2014).

Vine reproductive growth is generally less sensitive to water deficit than vegetative growth (Williams, 2000). Rodrigues (2011) says that the plant's water regime is the balance between the need for water that grapevines needs and the amount of water available in the soil, which may be compared according to the amount of precipitation and the amount the soil might store of water. Depending on the stage at which this occurs, different consequences may be observed on the growth of the vine, such as it is productivity and fruit composition.

Water status in the plant throughout its cycle is a key factor for proper physiological and biochemical functioning. Under water restriction, the metabolism of the plant is affected and thus the development of the berries the type of wine produced (Deloire et al., 2005). Irrigation may reduce the negative effects of water shortages on a vineyard, preventing limited yield. It is important to note that in the future when there will be competition for water, consider to have an efficient use of those resource so that it is used at periods and levels that do not affect the quality of musts (Hernández, 2015).

Mediterranean zones often occur in summer water for a plant. These arise as a result of low soil water reserves due to low recorded rainfall and high water vapor pressure gradients between leaves and air (Malheiro et al., 2011). Irrigation may be used in these region with the aim to control the reproductive growth and controlling vigor (Chaves et al., 2010).

Prolonged drought during maturation period, as long as it doesn't affect photosynthesis, but it may favor the accumulation of sugars and phenolic compounds. As water availability decreases, there is an improvement in phenolic compounds, but the yield and size of the berries are reduced (Ojeda et al., 2005). High water stress tends to decrease plant vigor, sugar and acid content (De la Hera Orts et al., 2004).

Rankine et al. (1962) suggested that irrigation deficits might be good promote a ripeness uniformity. However, the berry composition and weight when compared to irrigate vineyards are unclear whether water stress may alter the uniformity.

The use of strategies against water deficit are important to supply water before evapotranspiration levels start to be higher than available water throughout the growing season (Fereres and Soriano, 2007). Long and severe water deficit may reduce vine vigor, yield and wine quality and it may have accumulated damage to plant growth and formation in subsequent years (Dayer et al., 2013). Large amounts of water can also have a negative impact on grape production, which it can lead to losses in productivity, color, acid balance and lower sugar content (Esteban et al., 2001). Studies have shown that late irrigation serves only to alleviate shrinkage of the berry and may not to increase in size (Castellarin et al., 2007; Keller et al., 2015).

Projections for the future in the Douro region are temperature rises more and more, which it will make the vines stress increase becoming a growing challenge for the production of quality wines. (Jones et al., 2012).

In warm regions, most grape varieties can reach sufficient levels of soluble solids, improve the quality wine, but it does not happen for anthocyanins and sugar in berries, what bring a negative balance between color and alcohol content in the wines (Sadras and Moran, 2012). Although water deficit changes the concentration of anthocyanins and tannins is unclear that affects biosynthesis in grapevine berries (Ojeda et al., 2002; Buchetti et al., 2011; Cáceres-Mella et al., 2017). Water deficit stimulates the secondary metabolism in berries and the concentration of phenolic compounds, protein and sugar accumulation (Pastenes et al., 2014; Villalobos-González et al., 2017) and its regulation is an important tool used for improving flavonoid content and to increase the quality in red wines (Kennedy et al., 2002).

In situations of extreme heat and drought, there are stops of maturation during the day due to water deficiency. Thus, watering when applied carefully is advantageous to the vine, as it favors the evolution of maturation, with adequate balance of sugars and acids, and a higher concentration in phenolic compounds (Loureiro, 2017).

To compare phenolic compounds with climatic conditions or chemical treatments, spectrophotometric analyzes have been used, as they are simple and low cost (Tabart et al., 2010). Sunlight intensity in the aerial part of the plant may be correlated with the amount of sugar, the variety of secondary metabolites and the amount of acid present in the wine's flavor, aroma and phenolic composition (Downey et al., 2006).

2.2. Training system

Training system is a set of decisions that determine the arrangement of aerial part tree in the available space, with direct influence on the yield and fruit quality. There must be a balance between the aerial part of the vineyards to allow light to enter the plant (Hidalgo, 2003).

The same author and Reynier (2005) observed that in viticulture two training systems are used and they are Cordon Royat and Guyot. The first is usually used for short pruning, it left only two nodes that will be the permanent cord. Guyot is used for long pruning where arms are left on one side or both sides of the plant.

The number of sticks and buds left after pruning is what will determine the leaf area of the plant and, consequently, the number of bunches, thus it define their yield. The plant vigor related to the intensity and type of pruning determine the final fruit composition for wine production (O'Daniel et al., 2012). Shooting management related to the type of pruning and the intensity will interfere in the next plant production cycles, affecting bud fertility and accumulated reserve (Pellegrino et al., 2014).

The most pruning system widely used in the Douro region is the unilateral or bilateral cord which is relatively simple to install, mechanized and wide range of grape varieties. Although apparent simplicity and adaptability of the cord, it is argued that a form of leading with a lower plant, as Guyot may be more appropriate, especially in the driest and most such as the Douro Superior, where it was traditionally used before participate in mechanization. In addition, vines in Guyot are less susceptible to exposition pruning leads increasing vines longevity (Magalhães, 2008).

The same author verified that the pruning in Cordon Royat is a short pruning, performing this pruning causes a large number of wounds that it can bring phytosanitary problems to the vineyard due to possible contamination with wood diseases. On the other hand, the performance of this pruning can cause problems in the aerial part, because the plants cause a high aerial part density. The main advantage of this type of training is the reduction of pruning time compared to other training systems such as Guyot.

Experiments conducted comparing plants in Cordon and plants in Guyot showed similarity in the results in relation to water deficit (Nader et al., 2019).

Training systems are very varied and differ according the number, position, length and shape they have. Pruning in double Guyot, also known as pruning on the stick and the branch (Zamboni, 2001), consists in remove two stems of the plant, and later, they

are tied to the trellis wire, and 2 thumbs. The type of pruning, we have all vegetation in a horizontal plane that allows to the clusters adequate lighting and aeration and beneficial for ripening. On the other hand, bilateral Cordon Royat pruning, also known as pruning cuts in Cordon (Hidalgo, 1999), where the buds are inserted along the Cordon.

For Ramos et al. (2016) the 2 pruning systems have similar way in terms of grape production and pruning firewood with a similar base load of buds in pruning. For the training system in Guyot, Draganov et al. (1975) observed that the plants conducted in this system have greater amount of heat on the leaves and berries, which it may impair their yield and cause greater phytosanitary problems, both in daily and nightly evaluations.

Although the temperatures are higher in plants conducted in Guyot, the plants do not show differences in productivity and composition of grapes and wine, when compared to other systems such as Cordon Royat (Peterlunger et al., 2002).

2.3. Grape Metabolism

Grape cultivation in dry and warm regions may affect wine production since berry growth, but in particular the amount of phenolic compounds. A loss of uniformity at the time of maturity can happen by high temperatures, making the appearance of flavor and aroma characteristics required for quality wines (Mira de Orduña, 2010).

In recent years some phenolic compounds have drawn attention for bringing benefits to human health (Thole et al., 2006). The quality of wines is related to the amount of phenolic compounds associated with taste, color and health benefits (Keller, 2010). It may directly prevent cardiovascular problems against the effect of free radicals against human metabolism (Virgili and Contestabile, 2000). Antioxidant compounds can reduce the effect of neurodegenerative diseases by protecting cells from oxidative stress, an example of disease that may be prevented with these compounds is Alzheimer's (Dixon and Pasinetti, 2010).

Anthocyanins (proanthocyanidins) and flavonoids are the most important groups of compounds present in grape berries (Castellarin et al., 2007). Phenolic compounds have great relevance in vegetables and viticulture have a great importance in berry coloration and taste properties. In red wines, tannins and anthocyanins are the most important compounds (Champagnol, 1984).

In the grapevine berry the strongly quality impact in colour and astringency are caused principally for flavonoids, anthocyanins and tannins synthesized (Cheynier et al., 2006; Kennedy et al., 2006). Phenolic compounds and anthocyanins are responsible for coloring in grapes and red wines. Their amount in grapes depend on production practices such as weather conditions and the variety used (Esteban et al., 2001).

The flavonoids stabilize by co-pigmentation the anthocyanin molecule. Thus, they bring benefits to human health by its antioxidant capacity, belonging to the group of phenolic compounds. (Singh Brar et al., 2008). Also, it may have effects against inflammatory issues and anticarcinogenic effects. With others flavonoids may still have antidiabetic activity (Doshi et al., 2015). One of the most important phenolic compounds present in grapes is resveratrol for its anticancer properties, which makes it an ingredient of interest to human health (Flamini et al., 2013). Little research has been conducted to compare field practices with the concentration of total phenolic compounds and antioxidant capacity (Coletta et al., 2013).

The wine quality is affected to the grape characteristics it being directly associated to sugar accumulation, total concentration of polyphenols and pigments, anthocyanins, tannins and total acidity (Peterlunger et al., 2000; Thole et al., 2006). From sensory and chemical point, the two most important phenolic classes in the grapevines are tannins and anthocyanins (Chassy et al., 2012).

Anthocyanin accumulation in berries is determined by climate factors, temperature, radiation, soil characteristics, nitrogen availability and vegetation wall management operations, such as driving and defoliation (Ojeda et al., 2002; Flamini et al., 2013). A fruitier aroma with lower vegetable aroma, higher amount of anthocyanins and lower astringency can be found in red wines correcting the water deficit (Castellarin et al., 2007).

When the berries begin to acquire color, this period should coincide with the highest temperatures of the year and may have an increase in amino acids, sugars and phenolic compounds (anthocyanins in red grapes), while the amounts of malic acid decrease and pH increase (Sadras and Moran, 2012).

Water use efficiency, stomatal conductance and photosynthesis levels change according to the variety used (Chaves et al., 2010). The photosynthesis pigments are responsible for the energy absorption (luminosity) by the plants, therefore, the chlorophyll content present is responsible for the photosynthetic performance of the plants. With water stress significantly decreasing chlorophyll content in plants, harming

the growth of several crops (Mafakheri et al., 2010; Gholamin and Khatnezhad, 2011). Chaves et al. (2007) shows that in different experimental conditions, various varieties showed that photosynthesis is enough resistant to hydric stress.

3. MATERIALS AND METHODS

3.1. Vineyard location

Field studies were carried out during cycle productive (2019) in a commercial organic vineyard, located at Quinta do Romeu (41°53'02'' North, 7°08'00'' West, 326 m altitude) in the NE Portugal (Figure 1). Touriga Franca (*Vitis vinifera* L.) vines were pruned in two different systems: Guyot and Cordon Royat. The vineyard was planted in 2011 with a vine density of 2.2 m x 1.0 m (4.545 vines/ha). The vines were grafted on 1013 Paulsen rootstock.

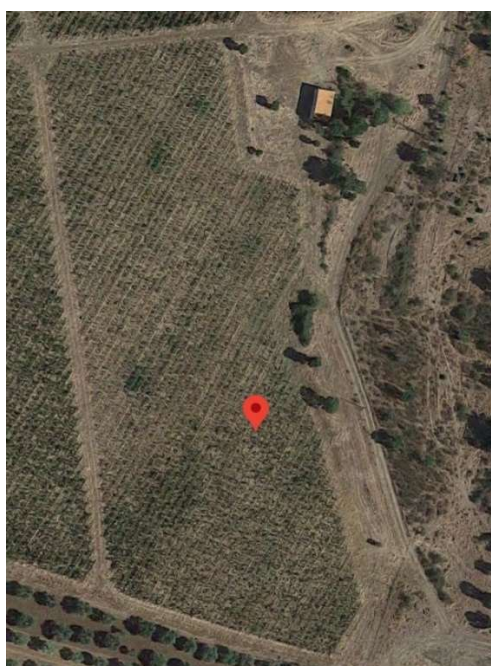


Figure 1 - Location of experimental vineyard. Font: Google Images (2020).

3.2. Experimental design

The experimental design consisted in two training system and three irrigation treatments in a randomized complete block design with three replications of five vines (Figure 2).

Vines were irrigated from pre-veraison until one week before harvest and the treatments consisted of two deficit irrigation regimes: 25 and 50% of the crop evapotranspiration (ET_c) and a full irrigation treatment (100% ET_c). The reference evapotranspiration was calculated using the FAO-Penman-Monteith method (Allen et al., 1998). Meteorological data were collected from an automatic weather station

(Campbell Scientific) located near the vineyard. The AWS is equipped with sensors to evaluate air temperature, relative humidity (Vaisala HMP155A-L, Campbell Sci), solar radiation (Skye SP1110, Apogee Instruments), wind speed and direction (03,002 Wind Sentry, R.M.Young), and, precipitation (Rain-O-Matic Small, Pronamic). Data was stored on a CR800 datalogger (Campbell Sci). The crop coefficient was calculated according to Allen et al. (1998). The beginning of water supplied was determined by the threshold value (-0.4 MPa) of pre-dawn leaf water potential (Malheiro, 2005). Irrigation water was applied through a drip irrigation system with pressure-compensated emitters of 2.2 Lh⁻¹ with one-meter line spacing (1 emitter per plant). The frequency of water applications was the same for all treatments and it varied from 2 to 3 days per week applied continuously until harvest. Emitters flow and water uniformity distribution were evaluated during the experiment. The frequency of water applications was the same for all treatments and varied from 2 to 3 days per week applied continuously until harvest. The water applied in each treatment, was shown in Table 1.

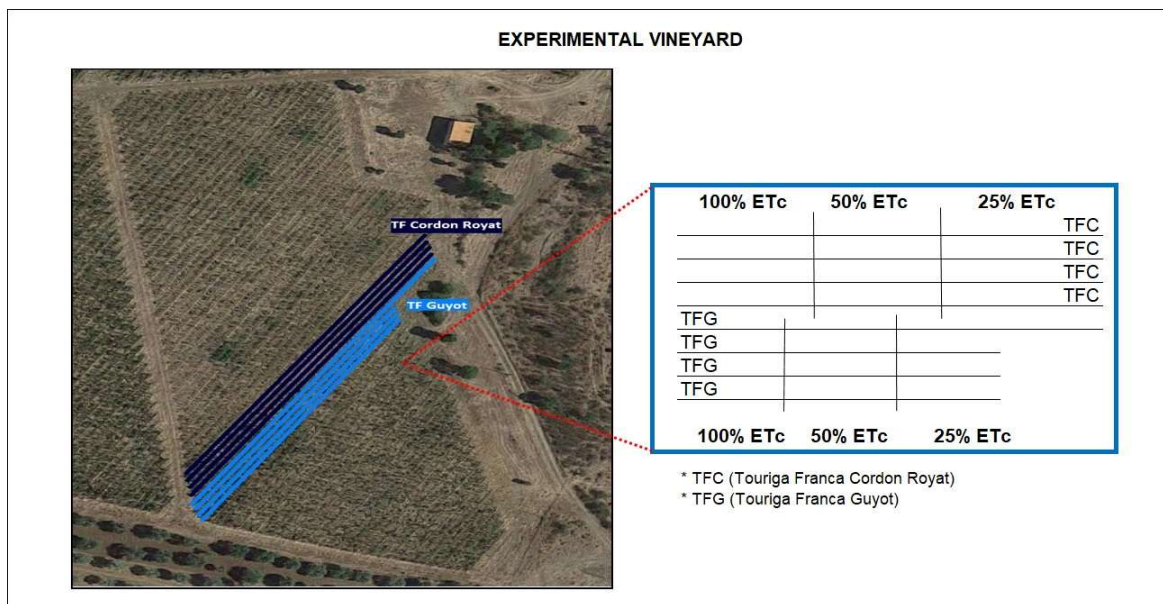


Figure 2 - Experimental design

Table 1 - Total amount of irrigation in the treatments.

Treatment	Code	% ETC	Water applied (mm)
TF Guyot	TFG25	25	34.5
TF Guyot	TFG50	50	68.9
TF Guyot	TFG100	100	137.8
TF Cordon Royat	TFC25	25	34.5
TF Cordon Royat	TFC50	50	68.9
TF Cordon Royat	TFC100	100	137.8

3.3. Climate

The Douro Region has a Mediterranean climate with rainy winters and dry summers with high temperatures (Malheiro et al., 2011). Average annual temperatures range from 11.8° C to 16.5° C. Average rainfall in the region ranges from 320 mm to 1200 mm with winds prevailing from the Northwest. In the experimental field there is a weather station that was used for data collection from January 1st to December 31st of 2019. According to the Koppen classification, the Douro region belongs to the Csa climate (C - temperate climate, s - winter rain, a - hot summer), it is a temperate climate, with rainy winters and monthly average temperatures, with average temperatures between -3°C and 18°C and hot and dry summers with monthly average temperatures of the hottest months which may equal or exceed 22°C (Kottek et al., 2006).

3.4. Phenological stages

The phenological stages are described by the BBCH scale with a description and coding system (Lancashite et al., 1991; Hack et al., 1992). The BBCH scale has been widely used to describe the phenological states of various crops, among which we can mention the vines (Lorenz et al., 1995). The BBCH scale is represented by two digits representing the growth stages, in the first digit represents the 10 major stages ranging from 0 to 9, and the secondary stages that they are within the primary stages divided into 10 secondary stages of growth, ranging from 0 to 9. The evaluation of the phenological stages in the present study began after the winter. The evaluations were carried out weekly until the plant reached the veraison stage and the beginning of berry maturation (81-83 on BBCH scale).

3.5 Physiological measurements

3.5.1 Water potential

The water potential (ψ_w) represents the free energy associated with the water molecules. In short-range movements such as osmotic, water travels from regions of greater water potential (higher free energy) for regions lower water potential (lower energy free) (Correia, 2014).

Predawn leaf water potential was measured with a pressure chamber before dawn. The pressure chamber model used is Model 1000, PMS Instrument Company, Albany, USA (Figure 3).



Figure 3 - Pressure chamber used in the experiment to measure leaf water potential. **Font:** PMS Instrument Company.

3.5.2 Chlorophyll Concentration Index (CCI)

Chlorophyll Concentration Index (CCI) were recorded non-destructively by using a portable SPAD (Soil and Plant Analysis Development) chlorophyll meter (Model, SPAD-502 plus, Konica Minolta Sensing Inc., Tokyo, Japan) based on measurements of leaf transmittance at 650 nm (red) and 940 nm (IR) wavelengths. For the SPAD evaluation, 30 leaves were measured within each irrigation block, with three replications with 10 leaves in each. Evaluations were made on August 14 and September 11, 2019.

3.5.3 Leaf gas measurements

Leaf gas-exchange rates were measured using a portable gas exchange system (LCA-4, Analytical Development Co., Hoddesdon, England). Measurements were performed in six fully expanded leaves per treatment on August 19 and September 9 of 2019.

3.5.4 Leaf area

Point quadrat analysis is performed to measure the aerial part of each plant (biomass), passing a stick through the production lines, measuring the contacts with a biomass, whether they are leafing, fruits or holes and recording the data found (Smart and Robinson, 1991).

For the Point Quadrat (PQA) evaluation, the hedge porosity was calculated according to the following parameters:

Number of layers of the sheet (NLS):

$$\frac{\text{Total number of sheets collected}}{\text{Total number of inserts}}$$

% of Inner Leaves (%IL):

$$\frac{\text{Number of inner leaves}}{\text{Number of leaves contacted}} \times 100$$

% of Inner Clusters (%IC):

$$\frac{\text{Total number of indoor clusters}}{\text{Total number of clusters}} \times 100$$

% of Holes (%H):

$$\frac{\text{Total number of holes}}{\text{Total number of inserts}} \times 100$$

Those analyses are usually performed in vines at the height of the grape bunches and thus evaluations of 0.2 m over a length of 2.0 m were realized, totaling ten evaluations per replication, with a total of 3 per treatment. At the end of the procedure, spreadsheets were created to standardize the data and thus make the appropriate evaluations.

3.6 Yield and berry characteristics

3.6.1 Biometric characteristics

To characterize the grape berries and verify their productivity and quality, biweekly evaluations were made using as parameter the longitudinal and equatorial diameter of the berries in millimeter, as well as the weight of the berries. For this evaluation, 100 berries were collected throughout each treatment. Berry evaluations were carried out from veraison to harvest.

3.6.2. Control of maturation and must quality at harvest

In order to analyze the composition of the musts, three samples of 100 berries per treatment were collected during the maturation period. The grape composition (pH, amount of total sugars, the total acidity and the total phenolic compounds) was evaluated through the official methods of the Organisation Internationale de la Vigne et du Vin (OIV, 2016). At harvest the samples were analyzed, through FTIR technology, Fourier-transform infrared spectroscopy (WineScan FT 120; FOSS, Hillerod, Denmark). The Total Soluble Solids (TSS), pH, total acidity, tartaric acid, malic acid, Total Polyphenol Index (TPI), Folin index, anthocyanins, color intensity were evaluated.

3.6.3 Yield components

At harvest, on September 16th, the number of clusters, yield per plant and cluster weight were determined on 15 plants per treatment. The number of berries per bunch was calculated, taking into account both the bunch mass and berry mass.

Same vines were pruned in the winter, and pruning mass was measured for each vine of each replication of each treatment. The Ravaz index (Ravaz, 1903) was calculated by dividing total yield per vine by the pruning weight.

3.7 Statistical analysis

Statistical analysis of the data was performed in program Statgraphics Centurión XVI Software. Analysis of variance was performed for each of the variables

dependents studied, with a variety (Touriga Franca), two conduction systems (Guyot and Royat Cordon) of variation the three availability water regimes (RDI25, RDI50 and RDI100). Whenever the result of the analysis of variance was significant ($P < 0.05$), we performed to the multiple comparison of means using the Tukey HSD test and the level of significance of 5%. The experimental design used was that of randomized blocks, which were chosen from the determined treatments.

4 RESULTS AND DISCUSSION

4.1 Climate conditions

Throughout the year of 2019, meteorological measurements were made and the climatic conditions of the experimental design showed that in the months in which the plant was in a vegetative cycle (months from January to April and post-harvest from September to December) (Figure 4) were the ones that presented a greater amount of precipitation, with an enhance on December with precipitation of 147 mm over the month, higher number when compared to the summation of the months of May to September with 90.2 mm, which it shows that the vineyard is located in a Mediterranean climate region with cold and humid winter and hot and dry summer, except for the month of February, which presented only 7.6 mm throughout the month.

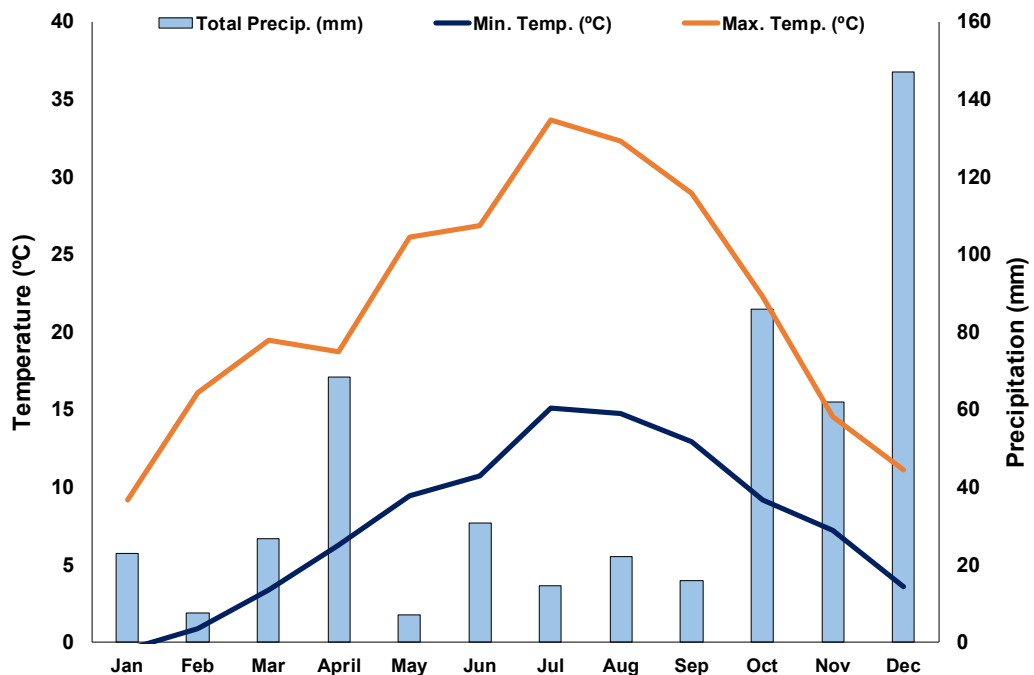


Figure 4 - Distribution of maximum temperature (°C), minimum temperature (°C) and total precipitation (mm) in 2019 at experimental vineyard.

The average temperatures of the months were not so different from other years, for the same region, showing that the plants did not suffer as much stress due to the temperature in the year 2019, even in the warmer months. The month with the highest average temperature (34°C) and the highest average minimum temperature (15.1°C) was the month of July.

With climate change, the impacts of temperature variations on agriculture have become an important concern over the last few years, with viticulture as an agricultural

activity influenced by temperature, what it can affect the composition of the vines and the quality of their final product, which is wine (Ribeiro, 2013).

Comparing the data of the meteorological station of the experimental area and the data of the IPMA (2020) among the years 1971 and 2000, it showed that in all months of the year 2019, the maximum temperatures in the experiment were higher when compared to the temperatures of previous years. Regarding the minimum temperatures, the months of January (-0.5 °C) and February (0.9 °C) were the months that presented average temperatures below the average, while the other temperatures were always higher, with the average annual temperatures higher than in the past, and it may cause damage to yield and also to the cultivation of vines, because with more heat it will be necessary to use a greater demand for water to keep the plants hydrated so that there are no problems in the development of the plants.

Observing the precipitation values of the IPMA data (2020) and comparing it with the data of the weather station, it may observe that in almost all months of 2019, the average precipitation of the months was lower than the average values among the years 1971 and 2000, with the months of April (68.4 mm), August (22.0 mm) October (86 mm) and December (147.0 mm) higher than that of the Climatological Norms it demonstrated that the annual precipitation was lower in the wine year. The decrease amount of rainfall in the summer months and with higher temperatures, it will be necessary to demand more water for irrigation and this increases yield costs, which can be a limiting factor.

4.2 Phenological stages

Table 2 – Phenology by BBCH codes for Touriga Franca in Cordon Royat (TFC) and Guyot (TFG) systems, in the 2019 cycle.

BBCH Code	Growth Stage	TFC	TFG
		Date	Date
1	Beginning of bud swelling: buds begin to expand inside the bud scales	3/11/2019	3/11/2019
14	Fourth leaves unfolded	4/15/2019	4/15/2019
60	First flowerhoods detached from the receptacle	5/21/2019	5/21/2019
66	60% of flowerhoods fallen	5/28/2019	5/28/2019
69	End of flowering	6/4/2019	6/4/2019
71	Fruit set: young fruits begin to swell. remains of flowers lost	6/4/2019	6/4/2019
81	Beginning of ripening: berries begin to develop variety-specific colour	7/31/2019	7/31/2019
89	Berries ripe for harvest	8/30/2019	8/30/2019

Considering the days of the year as calendar days and the BBCH scale, it was possible to verify that on day 105 was the day when the plants presented four unfolded leaves for Guyot (14) and five unfolded leaves for Cordon Royat (15) and inflorescences clearly visible according to the BBCH scale also start to appear for both pruning systems (53 inflorescences clearly visible). Information is present in Figure 5 and complete BBCH scale in Attachment 1.

In the Table 2 and Figure 5 showed the occurrence date of main phenological stages. Both tree in the Cordon Royat and the Guyot system showed similar development. On day 141 of the year the trees in the two systems begin of their flowering (61 beginning of flowering: 10% of flowerhoods fallen). On day 148, the vines in Guyot were 70% of flowerhoods fallen (67), while and the vines in Cordon Royat with only 50 % of flowerhoods fallen (65).

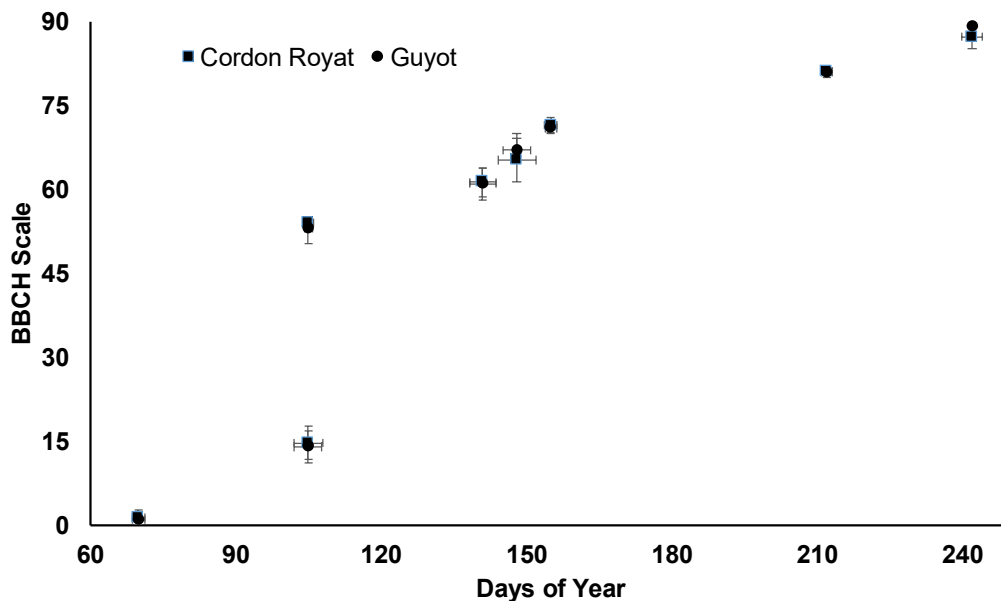


Figure 5 - Stage of development according to the BBCH scale for Touriga Franca at training systems in Cordon Royat and Guyot throughout the 2019 reproductive cycle on days of the year. Each training system was raised with an average of nine plants. Error bars represent the standard error of the mean.

The beginning of fruit development is a very important stage for the vineyard, as it is at this stage that the plants will determine the quantity of grapes that will be produced (71 fruit set: young fruits begin to swell. remains of flowers lost) and for both pruning systems, it occurred on day 155.

The stage 81 (beginning of ripening: berries begin to develop variety-specific colour) on the BBCH scale is the painter stage, where the berries begin to define the characteristics of the variety and both pruning systems presented this stage on day 212.

The harvest took place on September 16 (day 259), but in reality Guyot's berries were ready for harvest (89 berries ripe for harvest) on day 242, while on the same day for Cordon Royat they were still in the transition from 85 (softening of berries) and 89 (berries ripe for harvest). The system in Guyot by its design, favors for the fruits to have a faster maturation, due to the fact of it has not a barrier in the distribution of metabolites for the branches of the year, as only a branch and a stick are left, the distribution is makes it simple. In the case of plants in Cordon Royat, it has a natural barrier that is the firewood from the previous year.

4.3 Physiological measurements

The transpiration rate in the first assessment (Figure 6A) it showed that the vines that received more water (TFC100 and TFG100) were the ones that showed the higher rates during the morning time (3.66 mmol H₂O m⁻² s⁻¹ and 2.88 mmol H₂O m⁻² s⁻¹, respectively). They lost more water than the others due the greater amount of water available to the vines. The TFC50 treatment showed a statistically high value, but it was not as significant as the previous ones (2.58 mmol H₂O m⁻² s⁻¹). And the least transpiration rate treatments were TFC25 (1.01 mmol H₂O m⁻² s⁻¹), TFG25 (1.07 mmol H₂O m⁻² s⁻¹) and TFG50 (1.36 mmol H₂O m⁻² s⁻¹). The higher transpiration is caused by the greater amount of water available, which it helps in a better development of the plant, with better distribution of metabolites, that will serve for vegetative growth, in addition, it facilitates gas exchange between the plants and the environment allowing more stomata open and more water is exchanged with the environment, also facilitating the entry of CO₂ which will also allow the photosynthetic rate to be higher.

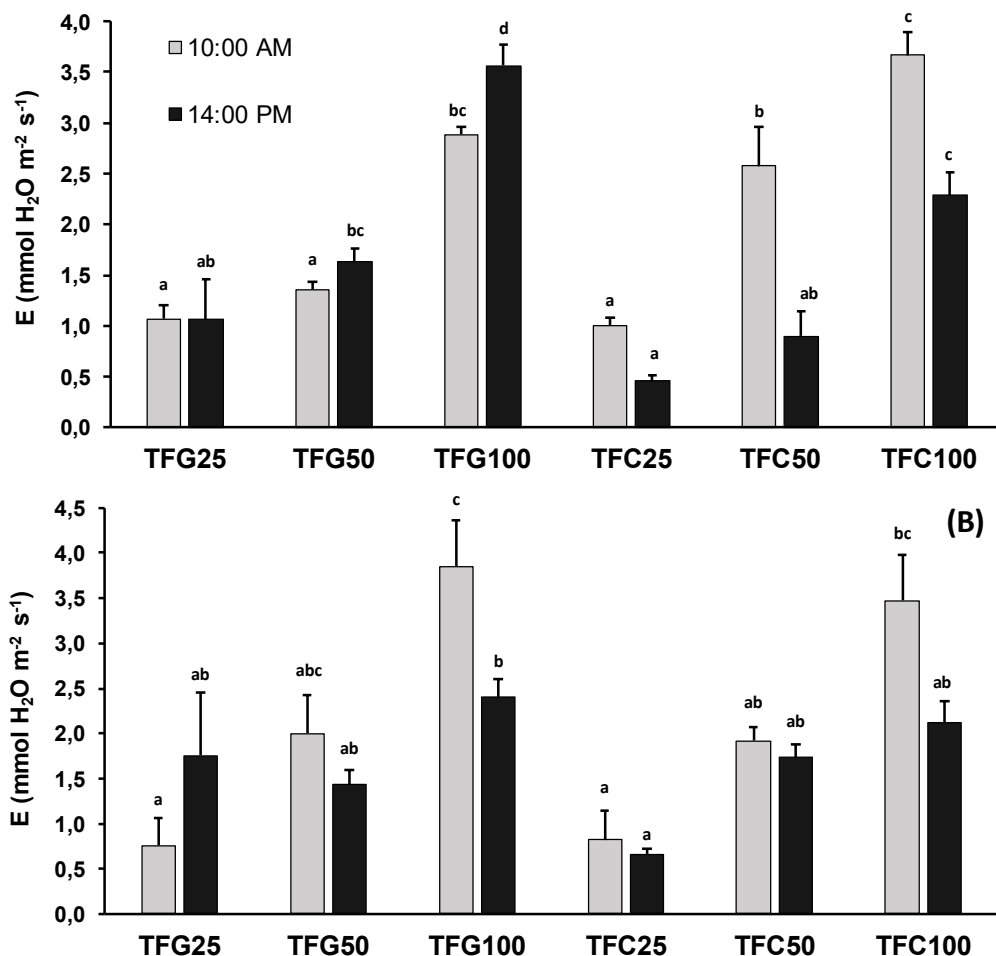


Figure 6 - Transpiration rate (E) measured in 19th of August (A) and 11th of September (B), 2019. The treatments followed by different letters are significantly different at P<0.05, according to Tukey HSD test. Error bars represent the standard error of the mean.

For the afternoon (Figure 6A), it may be seen that as the temperatures were warmer than in the morning, the plants showed less transpiration, due to the stomatal closure to avoid excessive water losses, but still, treatments that received more water through irrigation showed a transpiration rate when compared to the others (TFG100 with $3.56 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and TFC100 with $2.29 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$). The treatments in which the plants transpired less were the treatments TFC25 ($0.46 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$), TFC50 ($0.90 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) and TFG25 ($1.06 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$), this is because the treatments that received less water need to maintain their water reserves in order not to suffer even greater stress.

In the final stage of growth of the berries, the transpiration rate was better defined and with that, it was possible to really see in which treatments the plants were showing the highest transpiration rate, being the treatments TFG100 ($3.85 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) and TFC100 ($3.47 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$), while the treatments that showed statistically lower values were TFG25 ($0.76 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) and TFC25 ($0.82 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) values for the morning period (Figure 6B).

When it talks about the transpiration rate in the afternoon (Figure 6B), the values decreased because of the more intense heat, with that, the treatment that presented the highest rate of transpiration was TFG100 with $2.40 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ being statistically superior to the other treatments, while the TFC25 with $0.67 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ was the one that presented lower values than the others.

The stomatal conductance in the August evaluation in the morning (Figure 7A) showed that the treatments that received the most TFG100 and TFC100 water had the highest results, with $0.13 \text{ mmol m}^{-2} \text{ s}^{-1}$ and $0.13 \text{ mmol m}^{-2} \text{ s}^{-1}$, respectively. This showed that the stomata were open longer compared to other treatments, to help transpiration, facilitating the escape of water through transpiration and gas exchange, it was a parameter highly related to the rate of transpiration. As plants sweat more, stomatal conductance is also greater. Stomatal conductance is influenced by the vapor pressure deficit and the leaf's water potential, without excluding the effect of solar irradiance, so the plants that received the most watering will have the highest stomatal conductance. The treatments that received the lowest watering (TFC25 and TFG25) were those with the lowest stomatal conductance.

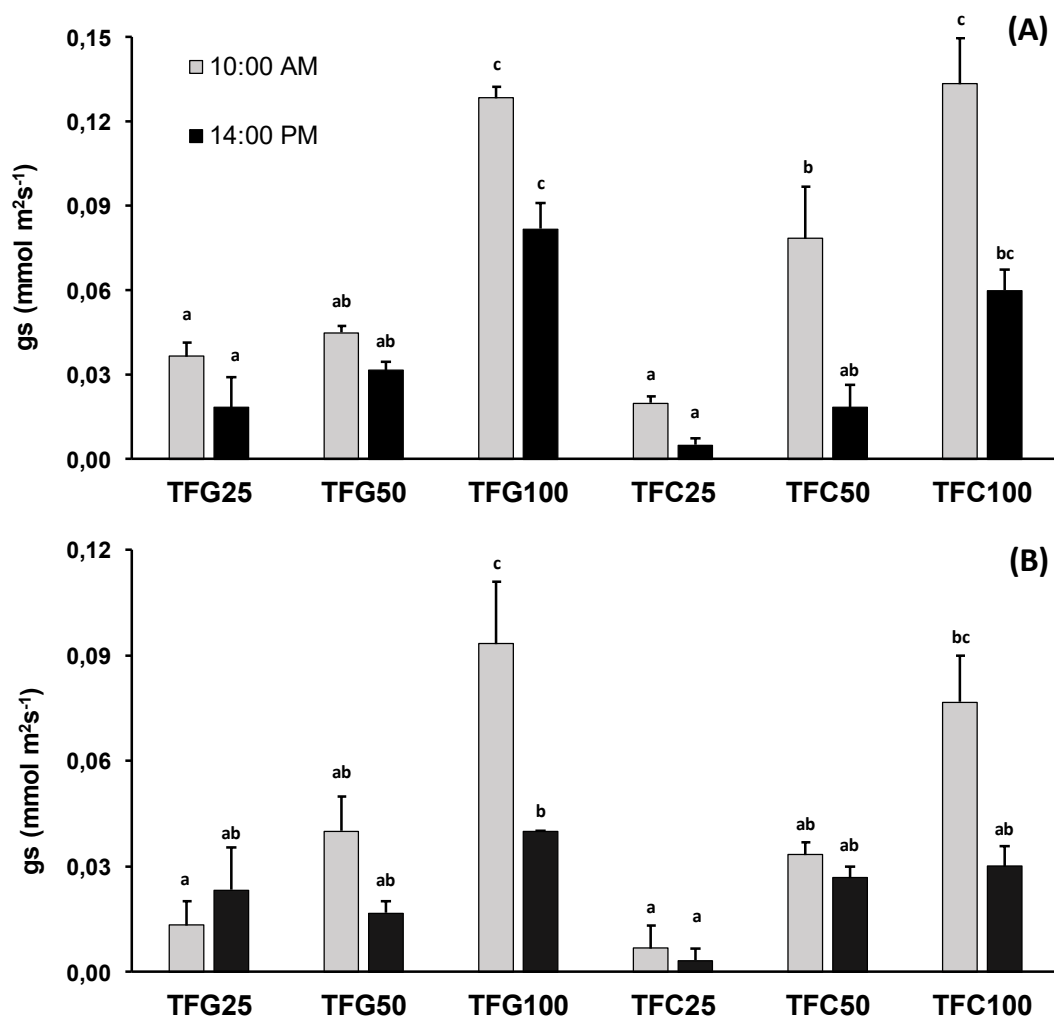


Figure 7 - Stomatal conductance (gs) measured in 19th of August (A) and 11th of September (B), 2019. The treatments followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test. Error bars represent the standard error of the mean.

For the afternoon evaluation on the same date, it may be observed that the control treatments TFG100 and TFC100 were the ones that presented the highest values. The treatments that received less allocation, were those with the lowest results (TFG25 with $0,02 \text{ mmol m}^{-2} \text{ s}^{-1}$ and TFC25 with $0.01 \text{ mmol m}^{-2} \text{ s}^{-1}$).

For the second evaluation that was carried out on September 11, in the morning (Figure 7B) the treatments that presented high values of stomatal conductance were those that received the treatments TFG100 and TFC100, while the ones that presented lower values were the treatments TFG25 and TFC25 with values of $0.01 \text{ mmol m}^{-2} \text{ s}^{-1}$ for both treatments. In the afternoon, TFG100 treatment showed the most significant result when compared to the others, with $0.04 \text{ mmol m}^{-2} \text{ s}^{-1}$, while the TFC25 showed lower results.

The plants of the treatments that received the greatest amount of irrigation water TFG100 and TFC100 were those that presented the highest stomatal conductance, while the treatments that received lower quantity of water oscillated in average values,

due to their water stress, however, the plants of the TFG25 and TFC25 treatments were the ones that presented the greatest stomatal closure, mainly due to the water stress of the plants and the heat of the place, causing the stomata to be more closed and lose less water. This greater stomatal opening in plants that receives a greater amount of water shows no difference in phenology, Guyot and Cordon Royat with similar growth, and the quality of the fruits did not show significant differences between treatments.

The decrease or increase in the net photosynthetic rate is directly related to stomatal conductance, since in the hottest hours of the day the stomata close, decreasing photosynthesis, absorption of water and nutrients by the roots. The results showed that for the first evaluation in the morning (Figure 8A) that the TFG100 and TFC100 treatments had a higher photosynthetic rate $13.39 \mu\text{mol m}^{-2}\text{s}^{-1}$ and $12.54 \mu\text{mol m}^{-2}\text{s}^{-1}$, respectively. The treatments that received the least amount of water were those that showed a net photosynthetic rate, being TFG25 and TFC25 with $5.75 \mu\text{mol m}^{-2}\text{s}^{-1}$ and $3.32 \mu\text{mol m}^{-2}\text{s}^{-1}$, respectively.

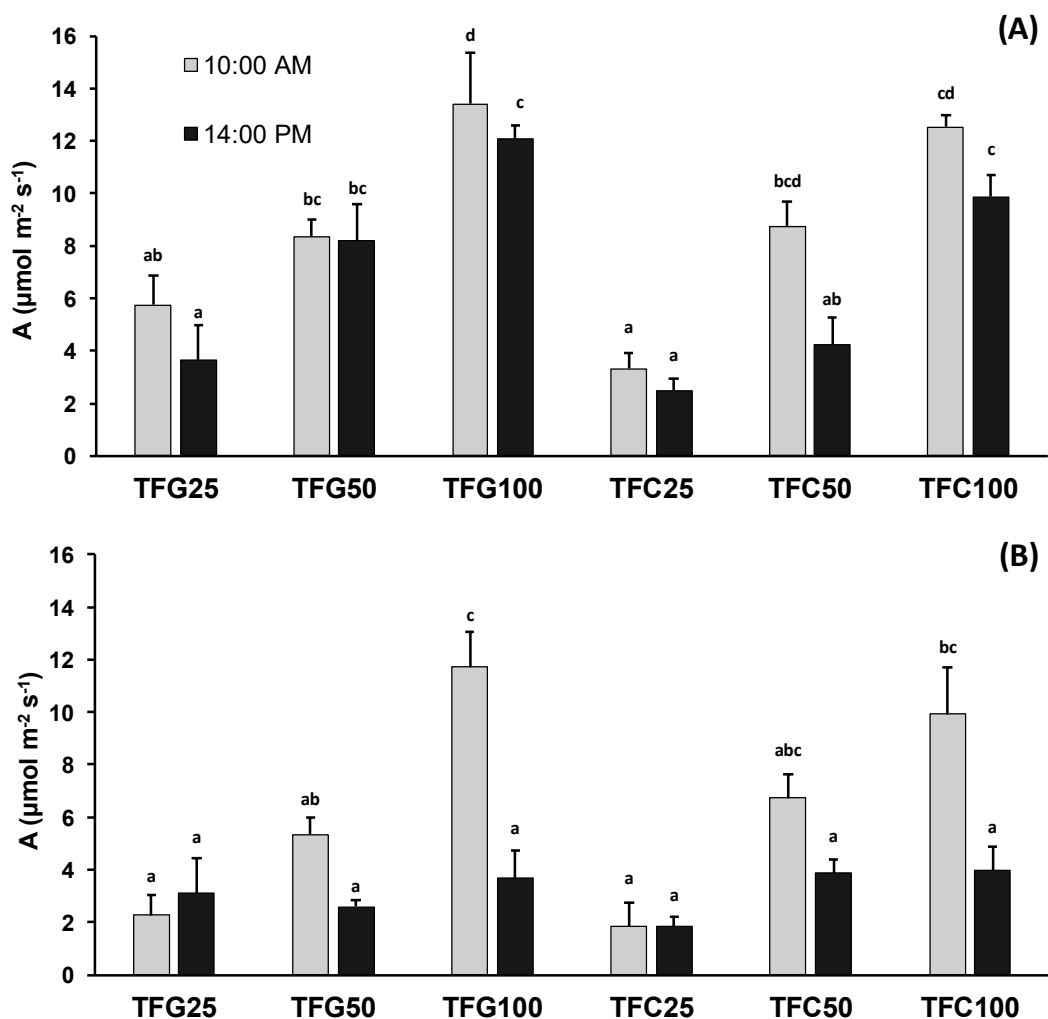


Figure 8 - Net photosynthetic rate (A) measured in 19th of August (A) and 11th of September (B), 2019. The treatments followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test. Error bars represent the standard error of the mean.

In the afternoon evaluation, the TFG100 and TFC100 treatments with the highest watering were the ones with the most significant values, while the TFG50 and TFC50 treatments were the intermediate treatments, and the TFG25 and TFC25 were the ones with the lowest values for this parameter. By the end of maturation, one week before harvest, the net photosynthetic rate in the morning (Figure 8B) showed results in which the treatments that received the most water through irrigation were those that presented the highest net photosynthetic rate (TFG100 with $11.71 \mu\text{mol m}^{-2} \text{s}^{-1}$ and TFC100 with $9.93 \mu\text{mol m}^{-2} \text{s}^{-1}$), while treatments with less watering, decreased this values. In the results of the afternoon there are no significant differences between treatments. With more heat in the afternoon than in the morning, the photosynthetic rate is lower due to stomatal closure, and with this lesser amount of open stomata, the assimilation of CO_2 by gas exchange ends up being lower as well, because the mechanism of gas exchange in a plant is due to its stomatic opening.

The intrinsic efficiency of water use is the ratio between the net photosynthetic rate and stomatal conductance, with that, it had for the first evaluation of August 19 in the morning (Figure 9A) with the results that the TFG25 (155.07) and TFG50 (185.48) were the treatments that showed a higher result compared to the others statistically, while the treatments TFG100 and TFC100 were that presented the lowest statistical results. The treatments with greater irrigation allocation have a greater net photosynthetic rate and greater stomatal conductance when compared to the others.

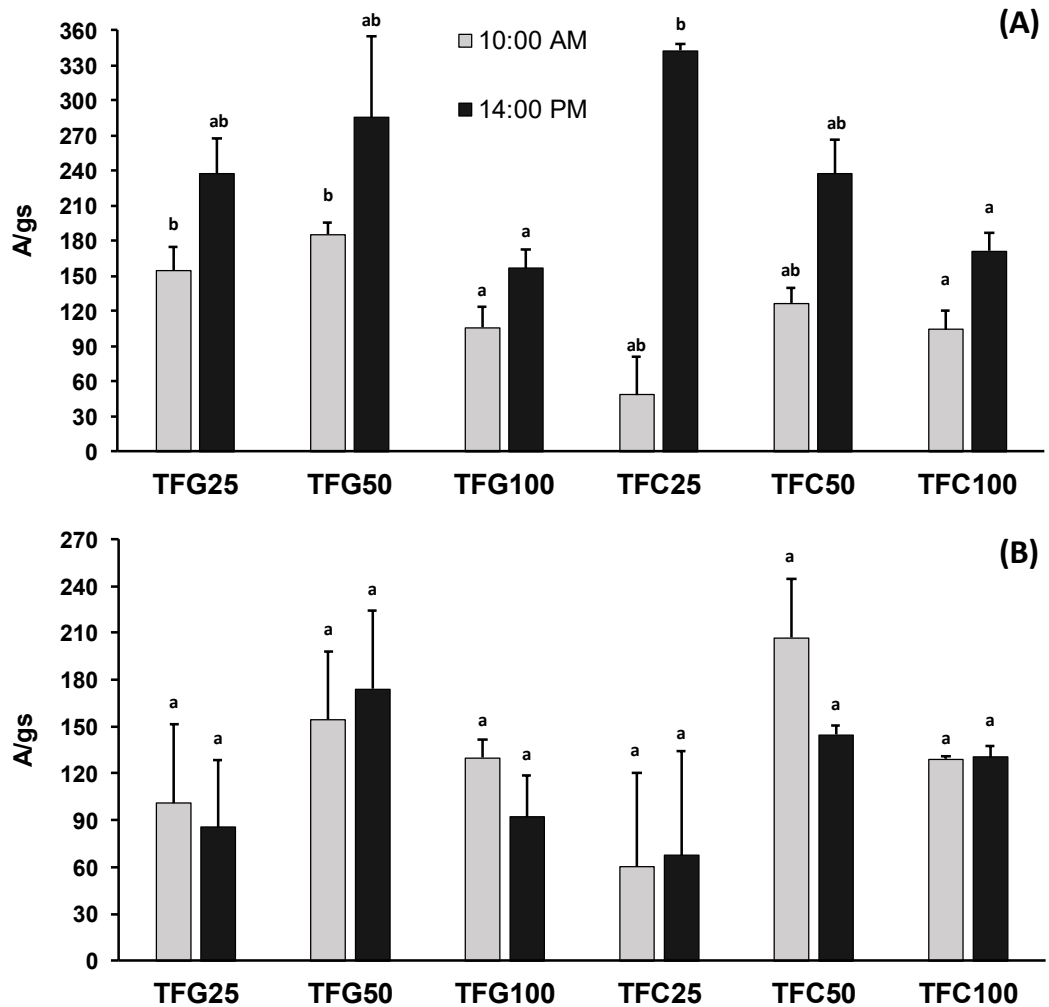


Figure 9 - Intrinsic water use efficiency (A/g) ($\text{mmol}\cdot\text{mol}^{-1}$) measured in 19th of August (A) and 11th of September (B), 2019. The treatments followed by different letters are significantly different at $P\leq 0.05$, according to Tukey HSD test. Error bars represent the standard error of the mean.

For afternoon measurements, TFC25 (237.32) is the one with the highest ratio when compared to the other treatments, while the treatments with the highest watering allocation were the ones with the lowest ratio (TFG100 with 156.49 and TFC100 with 171.39). For the second measurement date (Figure 9B) both the results in the morning and in the afternoon did not show significant differences among treatments.

The water deficit causes stomatal closure as the first physiological responses of plants, subsequently decreasing the photosynthetic rate and the rate of transpiration (Loveys et al., 2004; Chaves et al., 2010). The results of August 19, showed that in the morning the values of the results are higher than the results found in the afternoon, for the three variables (Figure 9B).

Regardless of water stress and water availability, the vine always activates its mechanisms to combat the high temperatures that are felt on its solar surface, although it is not always able to achieve this goal. According to Loureiro (2017) and Cifre et al. (2005) the recorded stomatal conductance also shows very low values in all

measurements, which it is an indicator of the grapevine's response to water stress, through stomatal closure to prevent water loss in periods with very high temperatures.

The link between stomatal conductance and photosynthetic rate may be used to monitor the water stress of plants (Cifre et al., 2005; Flexas et al., 2004). The response of photosynthesis to water stress is not linked to stomata, and it must be evaluated with the liquid photosynthetic rate, while the intrinsic efficiency of water use allows to assess the limitations of photosynthesis to water stress (Flexas et al., 2004). Comparing this, to our results we can see that stomatal conductance is quite low, being synonymous with the fact that plants have closed their stomata.

The predawn water potential in early summer (July) showed no difference between treatments, as solar irradiance began to increase this month (Figure 10A). Regarding the month of August, the treatment with the lowest water potential was TFG25 with -0.83 MPa, followed by the treatments TFC50 (-0.55 MPa) and TFC25 (-0.47 MPa) (Figure 10B).

The first treatment (TFG25) showed that the plants are close to drought, because this treatment showed greater water stress compared to the others, low humidity and excessive transpiration cause a decrease in leaf water potential, which may lead to the closure of stomata.

In September, the TFG25 plants managed to recover some of the initial water stress, while the TFC25 plants decreased in leaf water potential (Figure 10C). Regarding the total water potential, the treatments that received less water TFG25 and TFC25 presented lower values of leaf water potential, -0.58 MPa and -0.78 MPa, respectively. It showed that the plants submitted to these treatments underwent greater water stress, with that, their stomatal conductance, transpiration rate and photosynthetic rate were lower, because the vines needed to maintain their balance to avoid drought and thus senescence.

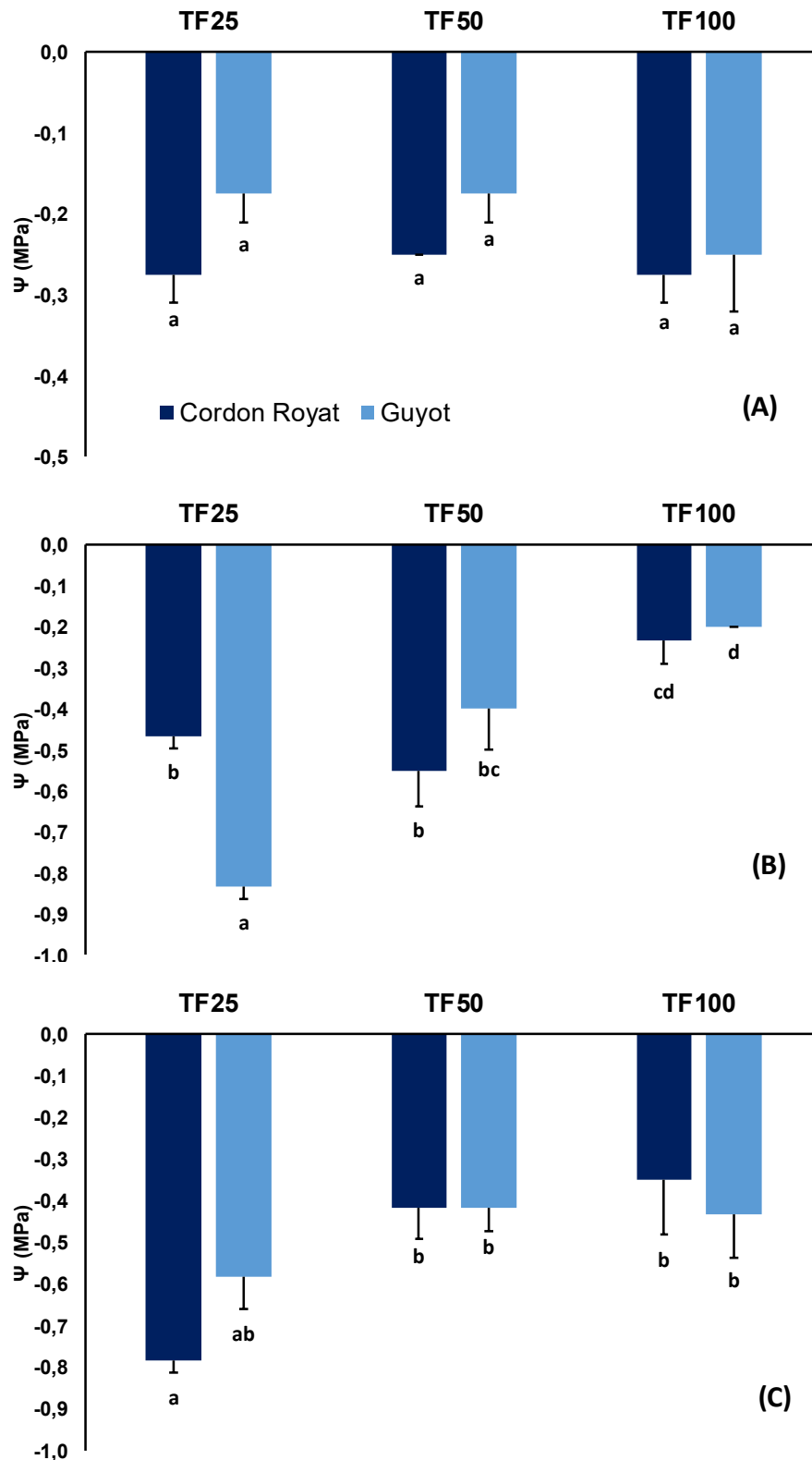


Figure 10 - Predawn stem water potential measured in 2nd of July (A) 19th of August (B) and in 11th of September (C). Each treatment had an average of three repetitions each. The treatments followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test. Error bars represent the standard error of the mean.

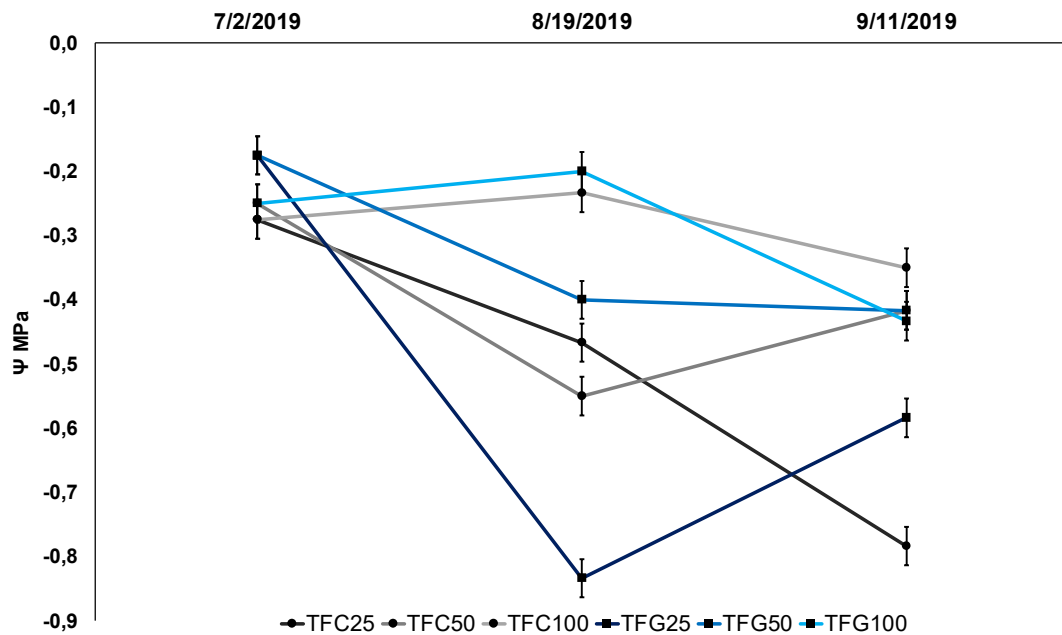


Figure 11 - Evolution of the predawn stem water potential along the cycle. Guyot treatments are shown with line and squares and Cordon Royat treatments are shown with line and circles. Error bars represent the standard error of the mean.

The measurement of the predawn stem water potential is performed before dawn, time of day when it is considered that there is a balance between the water potentials of the leaves and the soil, in the absorption zones without solar radiation, maintaining the absence of transpiration, and consequently, the amount of water present in the vine is equivalent to the water available in the soil (Lopes et al., 1998).

Observing the evolution of predawn water potential, we may see that in the first measurement (July 7), the treatments showed a good water potential and with plants had more humid than dry. However, this measurement was made in early summer and the temperatures were rising (Figure 11).

In the second evaluation (August 19), the TFG25 plants were very susceptible to lack of water, as it was the treatment that received the least amount of water during irrigation, while the TFG100 and TFC100 treatments were the most comfortable, because they are the treatments that received more water for irrigation (control).

In the final evaluation (September 11) it may see that the treatments that received the most water were the ones that became more humid at the end of the cycle, with that, the plants presented less water stress, with TFC100 presented the best result, followed by the TFG50 treatments, TFC50 and TFG100, with that, we may see that even with deficient irrigation being half of the control, the plants of these treatments (TF50) did not suffer much water stress. The TFC25 and TFG25 were the ones that

came closest to drought, having the first with greater drought problems over the summer, which may show that water stress on plants in Cordon Royat is greater with little water availability when compared to Guyot.

Even with water stress affecting more plants in Guyot, in the short term, there was no difference in the results between treatments, as it is an experiment in the first year of evaluation, it has to be observed whether in the next ones this stress will cause damage to production and in the quality of the berries.

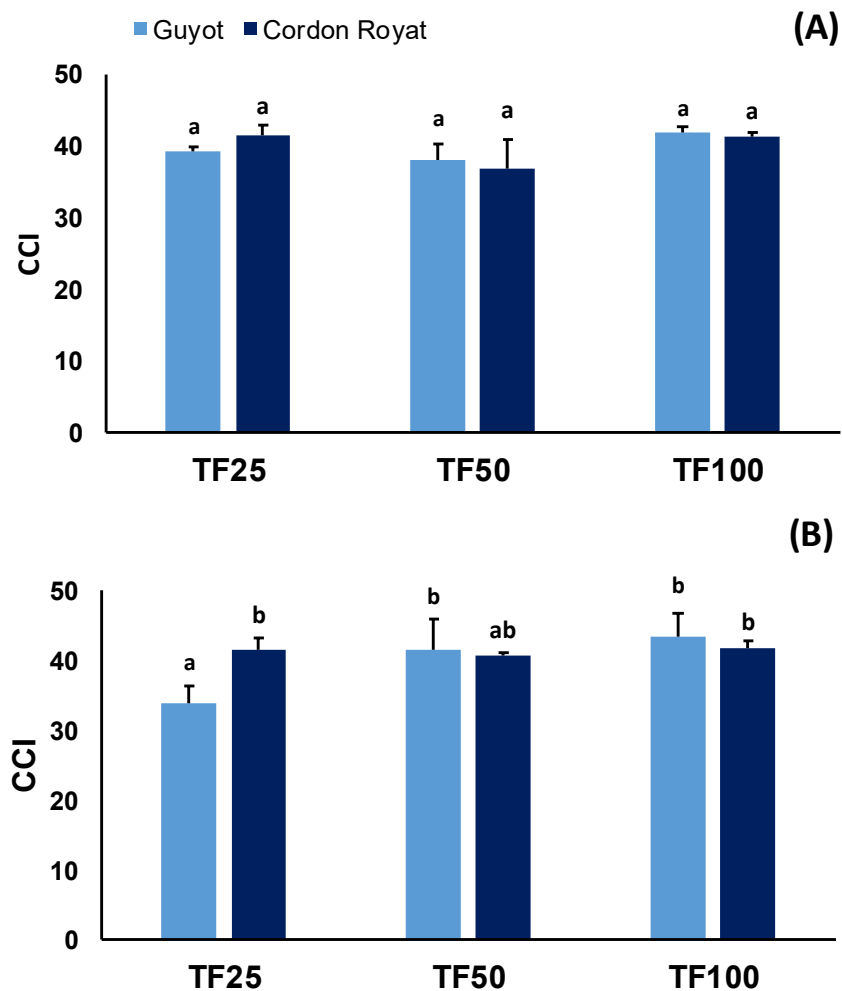


Figure 12 - Chlorophyll Content Index measured in 14th of August (A) and 11th of September (B). The treatments followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test. Error bars represent the standard error of the mean.

The SPAD is used to measure the levels of chlorophyll in the leaves. The results obtained in the month of August there was no significant difference in the treatments (TFG25 with 39.30; TFG50 with 41.63; TFG100 with 41.83; TFC25 with 41.53; TFC50 with 36.90 and TFC100 with 41.23), what it can show that the plants had good levels of total chlorophyll (Figure 12A).

In September, the plants ended up changing this stability, with a non-significant increase among treatments TFG50, TFG100, TFC25 and TFC100. However in the treatment TFC50 (40.67) the plants showed a slightly significant change when compared to the previous ones. The TFG25 treatment (33.90) presented the most significant difference, decreasing the levels of chlorophyll, a fact that it may be explained by the stress that the plants suffered, with more water losses, what it can decrease the magnesium rate that is responsible for the absorption chlorophyll, disrupting its main functions, such as breathing, sweating and photosynthesis (Figure 12B).

Comparing the months for the SPAD, the treatments in general increased, even if not significantly their average values. However, the TFC25 treatment showed a slight decrease in their values, while the TFG25 showed a significant decrease of values (39.30 to 33.90). The treatments that received less water through irrigation were those with lower levels of chlorophyll, since the plants underwent greater stress.

The level of chlorophyll was lower in TFG25, which received less water than the others, which leads us to say that the plants subjected to conduction in Guyot are more demanding for the same water treatment than Cordon Royat, and with that, presented a lower chlorophyll level than the others.

4.4 Point quadrat analysis and foliar area

The results for the Point Quadrat Analysis showed that for number of layers of the sheet (NLS) the TFG25 results (2.80) as being the highest of the values, while the TFC25 with 1.70. The treatments in Guyot were the ones with the highest results for this parameter, while Cordon Royat was the training system that presented the lowest results (Table 3).

It is possible to observe that the average NSL values were higher than the values suggested by Smart and Robinson (1991). However, the values may be justified by Queiroz (2002), since in regions with high temperatures and low rainfall in summer. It had a larger layer of leaves is advantageous and it may create a microclimate favorable to the clusters, being less exposed to sun exposure. The TFC25 (1.70) was the treatment that most closely matches the expected values, which it was between 1.0 and 1.5. The other treatments of Cordon Royat, were the ones that most closely approximated the values expected, with best results if compared to the results of the pruning system in Guyot, which presented the worst results, with TFG25 (2.80) being the lower among them.

Table 3 - PQA metrics for three sample inserts, with leaves (L), clusters (C) and holes (B). Mean values (\pm standard deviation) followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test.

Treatment	NLS	%IL	%IC	%H
TFG25	2.80 \pm 1.32 c	38.10 \pm 0.22 b	78.95 \pm 0.46 a	0.00 \pm 0.00 a
TFG50	2.47 \pm 1.19 abc	31.08 \pm 0.22 ab	86.36 \pm 0.67 a	6.67 \pm 0.25 ab
TFG100	2.57 \pm 1.07 bc	29.87 \pm 0.22 ab	88.24 \pm 0.73 a	3.33 \pm 0.18 a
TFC25	1.70 \pm 1.23 a	25.49 \pm 0.20 ab	50.00 \pm 0.47 a	23.33 \pm 0.43 b
TFC50	1.73 \pm 1.08 ab	21.15 \pm 0.21 a	65.00 \pm 0.66 a	10.00 \pm 0.30 ab
TFC100	1.90 \pm 0.88 ab	14.04 \pm 0.17 a	56.52 \pm 0.60 a	3.33 \pm 0.18 a

NLS – Number of layers of the sheet.

%IL - % of Inner Leaves.

%IC - % of Inner Clusters.

%H - % of Holes.

For the results of % of Inner Leaves, the treatments in Guyot presented a higher percentage with 38.10% (TFG25), 31.08% (TFG50) and 29.89% (TFG100) when compared with the treatments in Cordon Royat (TFC25 with 25.49%, TFC50 with 21.15% and TFC100 with 14.04%). This shows that the number of leaves inside the plants is higher in Guyot, with that, it is possible that the clusters are better protected against sunlight and heat than in plants in Cordon Royat. However, it is possible that the leaves are also more overlapping.

The treatments in Cordon Royat were the ones that presented the average with higher significant results for the %IF, with TFC100 (14.04%) and TFC50 (21.15%) as the closest to what it is expected, equal or less than 10%. The treatments in Guyot showed averages the lowest results, with TFG25 as the lowest average.

Regarding %FI, in general, values higher than those recommended by Smart and Robinson (1991) were found. They recommend a percentage equal or less than 10%. This difference is again justified by Queiroz (2002), because to be a region with high temperature and low values of precipitation in the ripening phase of the clusters.

The % of Inner Clusters data showed that the treatments in Guyot TFG25, TFG50 and TFG100 had the largest amount of indoor clusters, with values of 78.95%, 86.36% and 88.24%, respectively. It showed that the clusters are in adequate conditions avoiding the exposure of the berries to the sun, thus being able to improve the quality of the grapes and with less water losses.

Comparing with the previous result with the data in Cordon Royat in the treatments TFC25, TFC50 and TFC100, the percentages were of 50%, 65% and 56.52%, respectively. These average values, may be explained due to a smaller number of leaves or the presence of more holes when compared to the pruning system

in Guyot. For the statistical assessment of %CI, there were no significant differences between treatments, with all values above expectations (higher than 40%).

The % of Holes is a clearly important factor when evaluating the PQA, because the more holes there are in the vegetation, the fewer leaves protecting the clusters and and more clusters are exposed. This may end up bringing a non-uniformity to the vine due to poor conduction of the shoots or by the pruning system in Cordon Royat, that does not present a uniformity as good as the plants in Guyot. The treatments TFC25 (23.33%) and TFC50 (10%) were the ones with the highest percentage of holes, while the TFC100 had a value closer to the treatments in Guyot.

The %H showed values below expectations, which it would be between 20% and 40%, with the TFG25, TFG100 and TFC100 were the best treatments compared statistically with the others, with 0.00%, 3.33% and 3,33%, respectively (Table 3).

Analyzing all the results for the PQA, it was possible to observe that the pruning of the training system in Guyot was better, with more interior leaves and clusters than the treatments in Cordon Royat and less hole. However, if it evaluated that more interior leaves it may be a sign of overlapping shoots, the plants in Guyot were worse, as there are more interior leaves that may be interfering with the photosynthetic assimilation of plants in general, reducing the quality of the clusters. On the other hand, a smaller number of holes, may present a greater uniformity of plants, which leads to believe that the plants in Guyot were better in this parameter than the plants in Cordon Royat (Table 3).

Meyers et al. (2015) described that the accuracy of PQA analyzes restricts its use by make a simple diagnosis in the canopy of the plants, providing coarse information about the treatments. Improving the analysis increases the usefulness of this type of parameter, providing researchers with more accurate information, thus making direct quantitative comparisons between canopies and other studies.

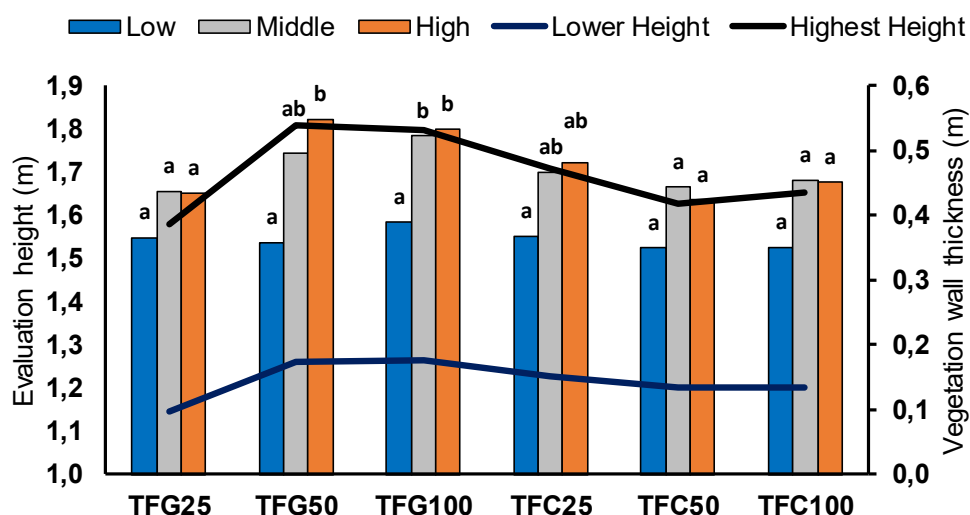


Figure 13 - Height of the vegetation walls and wall thickness. Low is the thickness of the underside of the vegetation wall between the two production lines. Middle is the thickness of the middle part of the wall. High is the thickness of the top of the wall. Lower Height is the minimum height of the vegetation walls and Highest Height is the maximum height of the walls. The treatments followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test.

The height of the walls is important to maximize the capture of light through the creation of a large vegetative wall, to reduce the density of the foliage and to increase the channeling of sugar to the clusters and thereby produce high quality wines. Usually for systems conducted in Guyot and Cordon Royat, the height of the plants should be around 1.80 meters.

The plants pruning in Guyot (1.58 m for TFG25; 1.81 m for TFG50 and 1.80 m for TFG100) were higher than the plants in Cordon Royat (1.71 m for TFC25; 1.63 m for TFC50 and 1.65 m for TFC100). It showed that the plants with greater height, had a better distribution in the lines when compared to the lower plants, which showed more density. However, the TFG25 treatment was the lowest of all, even in Guyot, which may indicate that the treatment that received the least amount of water in this pruning system - was the least developed (Figure 13).

According to the thickness of the vegetation walls, it is possible to see that at the bottom of the plants, which is the part where the clusters are located, the treatments did not show significant differences between them, due to all being at the height of the first wire of the bards. The middle part of the bards showed a statistically greater thickness in the TFG100 treatment (0.52 m) when compared to the others, with the TFG50 and TFC25 with a statistic very close to that of the best treatment.

In general, the treatments that presented an average of larger plants (TFG50 with 1.81 m, TFG100 with 1.80 m and TFC25 with 1.71 m) were the ones that presented the largest wall thicknesses in the middle and upper part of the bards, it showed that

the greater the vigor of the plants, the greater will be the height of the vegetation wall and the thickness of these walls (Figure 13).

4.5 Yield and berry growth

The yield of the vineyard is one of the factors that is expected when choosing a conduction system and the cultural treatments and this parameter that will define the quantity and quality of the wine to be produced, it being a comparative of performance, to improve the production process more and more, since choosing the training system until the efficiency of water use, to achieve the desired.

The treatments more productive were TFC100, reaching almost 14 ton/ha (13.93 ton/ha), TFG50 with 13.85 ton/ha and TFG100 with 12.38 ton/ha of grapes. These results would be a good quantitative value when compared to the other treatments, while the TFG25 treatment with 6.42 ton/ha was the one with the worst productivity. A fact that may be explained by the fact that the plants had greater hydric stress because they received less water in irrigation (Figure 14). It is important to say that the TFC25 treatment, which received a small dosage of water, showed good productivity (12.74 ton/ha) approaching the best treatments statistically, which may show that the productivity of the plants in Cordon Royat with less water are more productive even under water stress than the vines in Guyot.

Observing the results and comparing with the bibliography, the greater application of irrigation favored the increase in yield, it shows significant differences between treatments (Dos Santos et al. 2003; Serna, 2018). With a greater amount of water, plant roots have greater availability of nutrients, allowing for greater production (Van Leeuwen and Seguin, 1994).

Keller et al. (2016) observed that when the water regime of plants was 25% ETc reduces sprout growth and fruit development, making productivity lower. Studies with the Tempranillo grape variety in Spain (Uriarte et al., 2015) and with the Malbec grape variety in Argentina (Dayer et al., 2013) have shown the effects of water deficit on the growth and yield of vines. The bibliography shows that values close to 25 % of ETc as 30% to 35% of irrigation also limits the vines, but not as severely over the years (Casassa et al., 2015).

The average number of clusters per plant in Guyot (TFG25 with 10.27; TFG50 with 12.60 and TFG100 with 14.27) showed a smaller and less statistically significant number of clusters when compared with the plants in Cordon Royat (TFC25 with 15.58;

TFC50 with 14.60 and TFC100 with 16.67) (Figure 14B). It is possible to notice that the plants in Guyot, even having a good productivity for the treatments TFG50 and TFG100, that the plants had less clusters than the plants in Cordon Royat. It showed that the quantity of the berries must have been greater for this training system.

Regarding water productivity, there was a significant difference between the TFC25 treatment (32.55 kg/m³) to the other treatments, which they were statistically similar to TFG25 with 13.17 kg/m³ TFG50 with 16.64 kg/m³, TFG100 with 8.14 kg/m³, TFC50 with 11.23 kg/m³ and TFC100 with 9.16 kg/m³ (Figure 14C). A fact that it may be explained by the better water absorption and conversion into weight and productivity by TFC25, which it was a treatment that received less water in the training system. It showed that it was the one with the best use of the water, it being more efficient than the other treatments (Figure 14C).

Exploring a sustainable way of conducting a vineyard, in a scenario of low rainfall and high temperature, a treatment that receives less water and makes better use of this resource, ends up standing out above the rest, even with that, its productivity a little less than the best treatments.

Hernández (2015) verified with Tempranillo in different irrigation treatments it is possible to verify the effects on the intensity of water stress on the quality of wines and efficient use of water. Thus, treatments that receive the least amount of water are those that may be more efficient (25% ET_c), as long as this treatment results in greater water use efficiency. The severity of water deficits is linear with water productivity in the vineyard, as described by Fereres and Soriano (2007), yield and pruning increase as the amount of water for irrigation increases (Shellie, 2014).

Romero et al. (2010) and Chaves et al. (2007) reported a decrease in pruning weight, between the control treatment and the average treatment, which they were similar to the TF100 (control) and average TF50 treatments. However, in this study, only the treatment in Guyot showed a decrease, whereas Cordon Royat had an increase in weight.

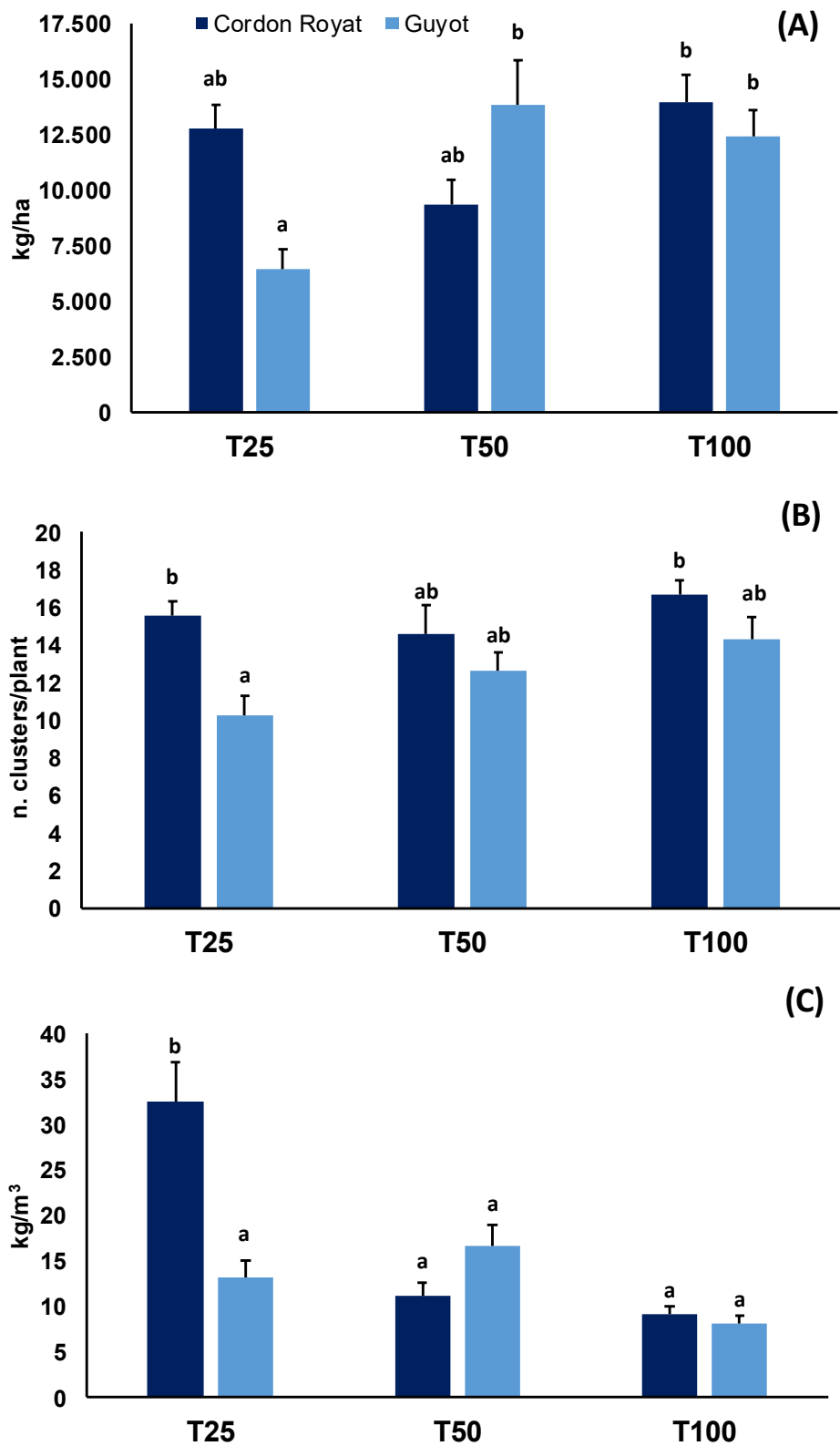


Figure 14 – Yield (A), number of clusters per vine (B) and water productivity (C). The treatments followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test. Error bars represent the standard error of the mean.

Observing the results of Table 4, the plants that presented a larger number of branches was the TFG50 when compared to the others statistically, with an average of 9.20 shoots per plant. The TFC25 was the one that obtained lower results when

compared to the others (6.87 shoots per plant). In general, the training system in Guyot was the one with the highest number of branches when compared to Cordon Royat, a fact that is explained by the system in Guyot at the time of the previous pruning, have been left a higher load than Cordon Royat.

Comparing individually according to the irrigation allocations applied, it is found that the only treatment that showed significant differences was the TFG50 when compared to the TFC50, while the others, even though the plants in Guyot had higher values when compared to the plants in Cordon Royat, did not present significant values higher for treatments TF25 and TF100.

Regarding pruning weight (Table 4), the treatment that produced the lower weight was TFC25 (0.27 kg/plant). Comparing the treatments individually within the same irrigation allocation, the TFG25 treatment presented a higher pruning weight when compared to the TFC25. The TFC50 treatment showed a statistically better result than the TFG50, while the TFG100 treatment presented a better statistical result when compared to the TFC100. Observing these results, it is difficult to say which training system presented the best results, but it is possible verified that the best individual treatment in the water supply and compared with all treatments was TFC50 (0.64 kg/plant). The treatment with the worst results in both variables was TFC25 (0.27 kg/plant).

Table 4 - Number of shoots, pruning weight and Ravaz index. Mean values (\pm standard deviation) followed by different letters (with the lower case letters representing the comparison between all treatments and the upper case letters comparing between the same irrigation allocations) are significantly different at $P \leq 0.05$, according to Tukey HSD test.

Treatment	Number of Shoots		Pruning weight (kg/plant)		Ravaz Index	
	TFC	TFG	TFC	TFG	TFC	TFG
25% ETc	6.87 \pm 1.41 aA	7.80 \pm 1.15 abA	0.27 \pm 0.14 aA	0.51 \pm 0.25 bB	12.85 \pm 7.42 bB	3.22 \pm 2.00 aA
50 % ETc	8.00 \pm 1.60 abcA	9.20 \pm 0.77 cB	0.64 \pm 0.19 bB	0.48 \pm 0.12 bA	3.50 \pm 1.85 aA	6.76 \pm 4.27 aB
100 % ETc	7.87 \pm 0.83 abA	8.47 \pm 1.41 bcA	0.55 \pm 0.15 bA	0.53 \pm 0.24 bA	5.86 \pm 2.08 aA	7.55 \pm 8.05 abA

According to Ravaz index, the treatment that presented the highest values, with a negative result was TFC25, which showed good productivity. However, it presented low pruning weight, showing that the plants aimed at the yield, then the vegetative production, bringing weaker plants, however, with good yield. The other treatments did not show significant differences between them when comparing all treatments together (Table 4).

When compared within the same irrigation allocation, TFC25 was the one that showed significant values when compared to TFG25, which it showed low yield, however, high pruning weight. The treatments in TF50 showed that TFG50 was statistically higher than TFC50, because the second, presented lower yield when compared to the first, due to its lower yield, and higher pruning weight. The control irrigation supply (TF100) did not show results that differed significantly between treatments.

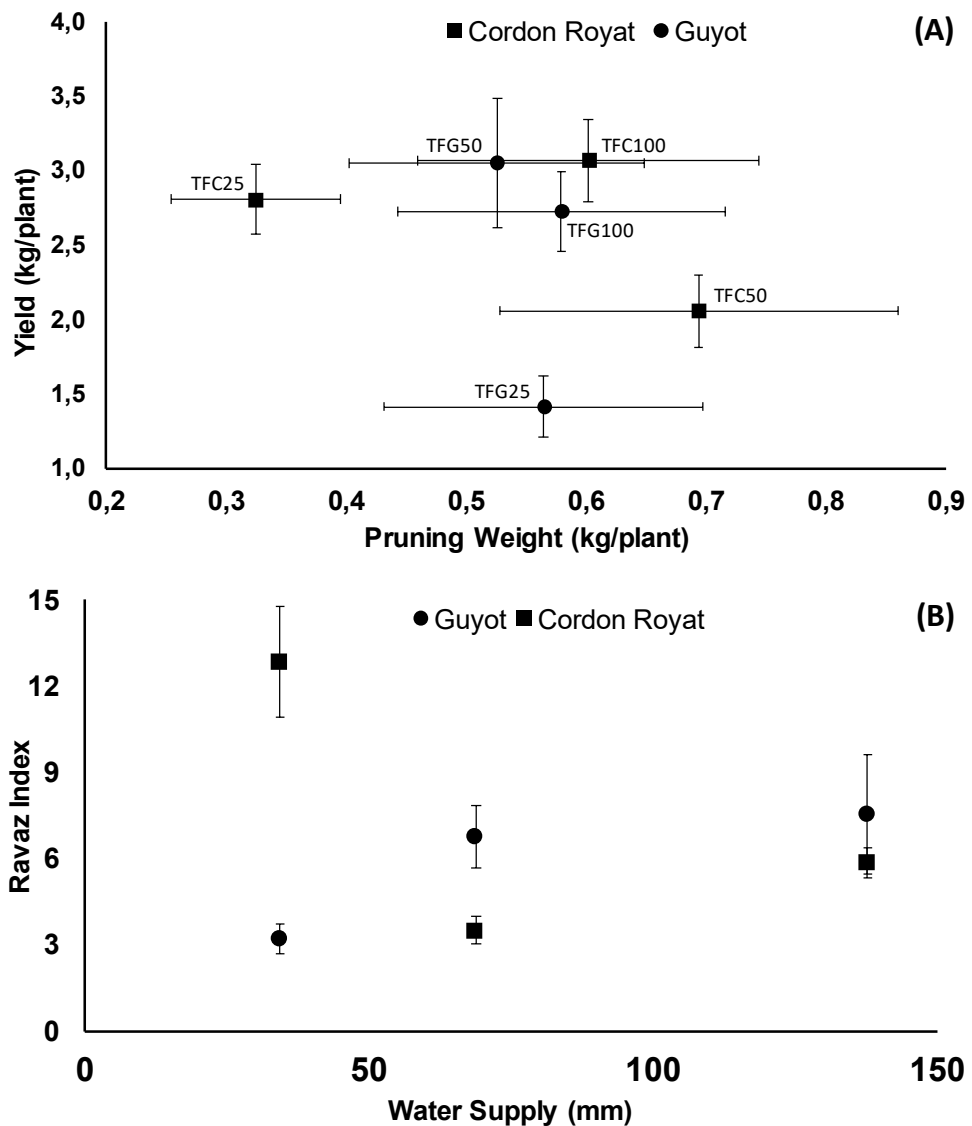


Figure 15 - Relationship between pruning weight and yield (A) and between Ravaz Index and water supply (B). Error bars represent the standard error of the mean.

In the relationship between productivity and pruning weight, it is expected that the most productive treatments are those with the greatest amount of firewood, due to the vigor of the plants being greater (Figure 15A). Of the treatments in Cordon Royat, the TFC25 showed high productivity. However, the weight of the pruning firewood was

quite low. It shows that the plants of this treatment with water stress were aimed at the production of bunches, as a consequence of high productivity.

The TFC50 treatment showed low productivity and high wood production, which leads us to believe that the vigor of the plants was greater and the plants intended for better vegetative growth. The treatment that received the largest amount of TFC100 water had good productivity and a good production of firewood, which it showed that the water received allowed the plants to grow and produce better.

For treatments in Guyot, TFG25 presented a low productivity and high production of pruning firewood, with that, the plants felt the lack of water, and leaned towards vegetative production. The TFG50 showed good productivity and production of firewood. It showed that even receiving half of water than the TFG100 treatment, it was ideal for the plant to develop and produce properly, the same result for TFG100 that showed a little more production of firewood. pruning, however, in productivity was similar to TFG50.

The Ravaz index showed that the treatment that showed the highest relationship between productivity and firewood production was TFC25 (12.85) when compared to the others. The treatments with the lowest Ravaz index were the TFG25, TFG50, TFC50 and TFC100 treatments with 3.22, 6.76, 3.50 and 5.86 respectively. The TFG100 treatment had a value of 7.55 and was intermediate (Figure 15B).

It is known that the Ravaz index should be between 4 and 10, although values above 7, what indicate a risk of depleting the plant's reserves, such as carbohydrates and nutrients, due to the excessive production of grapes (Kliwer and Dokoozlian, 2005). Already values less than 4 characterize the excess of vigor in the vines and/or the low grape production. Thus, the results obtained showed that the vines of the TFG50, TFC100 and TFG100 treatments were satisfactory according to the index, without risk of depletion or excess of vigor.

The TFG25 and TFC50 treatments showed values below 4, with the second had medium productivity and low vigor, while the first low productivity and good vigor. The TFC25 treatment showed values above 7 with good productivity and low production of pruning firewood, which it may mean depletion of plant reserves.

Shellie (2014), Williams et al. (2010) and the present study showed the highest Ravaz index for greater water deficit, what result in lower pruning weight, in the case of our study, only for Cordon Royat. This information is true, since plants in Guyot showed good pruning weight, however, with low productivity.

The increase in pruning weight showed that the water deficit inhibited reproductive growth for TFG25 and an increase in vegetative growth, while for TFC25 the water requirement prevented vegetative growth, but it did not interfere with reproductive growth. The reduction in irrigation reported by Romero et al. (2010) and Chaves et al. (2007) were the one that most influenced the fall in vigor of the vines.

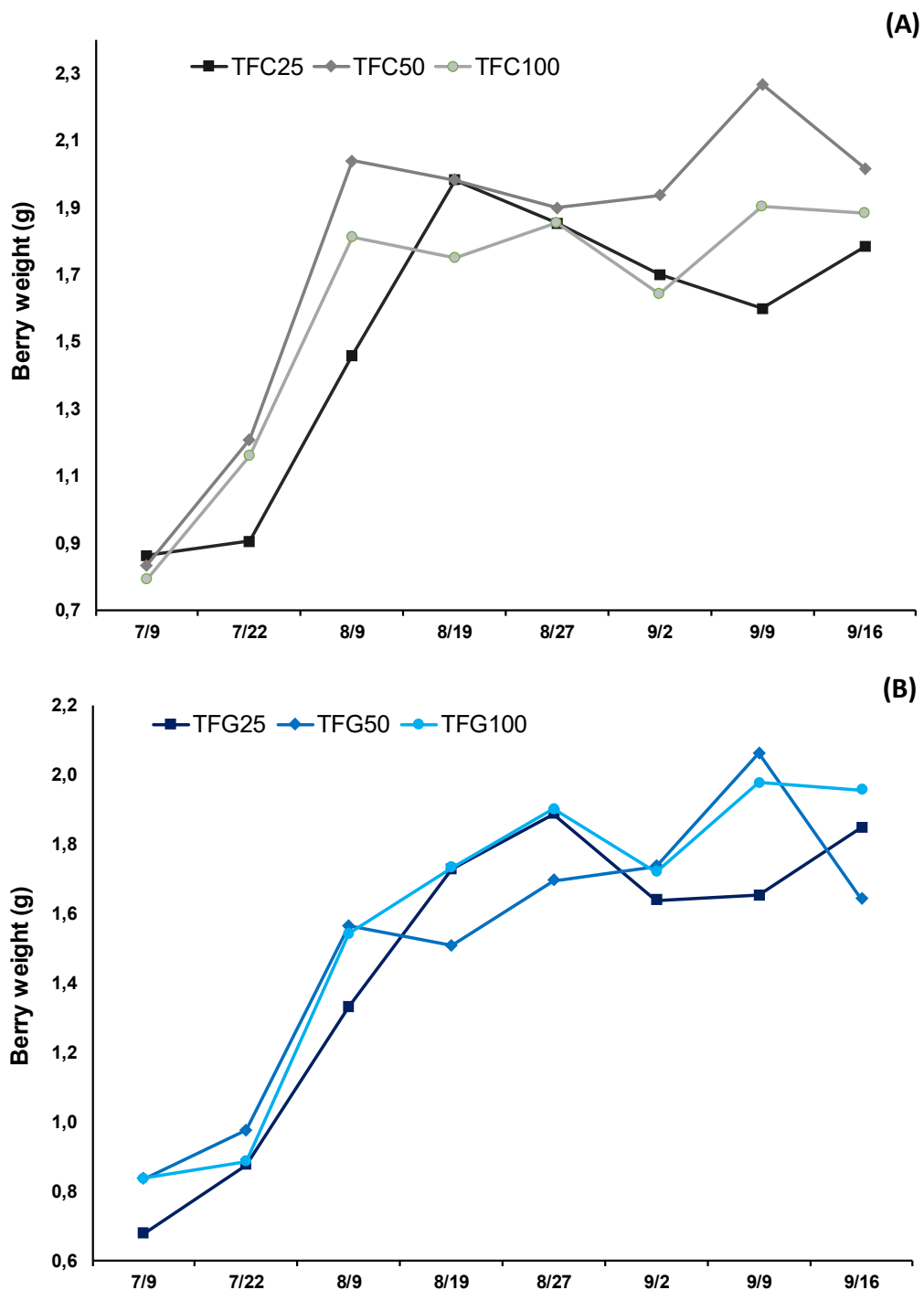


Figure 16 – Course of berry weight in Cordon Royat (A) and Guyot (B) pruning systems from pre-ripening to harvest. The treatments followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test.

Regarding the size of the berries, it is possible to say that the results had no significant differences between them, with the berries that received more water pulling out in initial growth when compared to those that received less water, and with the Cordon Royat berries (Figure 16A) standing out at the beginning of growth when compared to the berries in Guyot (Figure 16B). However, at the end, there was uniformity and all the berries showed results close to growth, regardless of the conduction system used, showing that regardless of receiving less water. All over the treatments showed a pattern of growth of the berries.

4.6 Quality of the musts at harvest

The treatment plants that produced the greatest amount of average SST were TFG100 (21.00 °Brix) followed by TFC100 (20.20 °Brix) in statistical values, while TFC25 (18.90 °Brix) was the one with the most average value low (Table 5). With this, the treatments that received the highest amount of water through irrigation, were the ones that presented the highest TSS values (TF100), followed by the TF50 treatments and with lower results the TF25, this within the same pruning systems.

Reference values for the pH of the berries varied according to the phase of the phenological cycle. They are between 2.3 and 2.9 in the painter's phase, and between 3.2 and 4.0 in the maturation phase (Champagnol, 1984). According to the results there were no significant differences between treatments. However, all over the treatments showed lower than expected values, with TFG100 getting closer to the expected values with 3.31 (Table 5).

It is known that the total acidity decreases during maturation. It is the sum of the volatile and fixed acidity of the must. Looking at the results, there are no significant differences between treatments. It shows that regardless of the irrigation allocation used, there were no significant differences. This parameter was not directly influenced by irrigation (Table 5).

Tartaric acid is the predominant acid in grapes and it decreases as the berries ripen. According to the results obtained in the harvest, the treatments did not present significant differences between them, with the TFG25 highest mean value (6.20 g L⁻¹), while TFC100 (5.33 g L⁻¹) was the one with the lowest average values (Table 5).

Regarding malic acid, it is known that its proportion decreases during maturation and that it is related to the increase in tartaric acid. The results showed that the lowest average value was observed in the TFG25 (0.80 g L⁻¹), in contrast, the TFC50 was the

one with the highest average value with 1.00 g L^{-1} . However, the treatments did not showed results significant, not varying statistically from each other (Table 5).

The similarity between the results is due to the conditions of the year and being the first year of research. However, as the pruning started in the year of research, there were net adaptation of the plants and they have not been subjected to years of stress. The metabolism of plants has not changed much this year. On the other hand, it is possible that over the years, the levels of malic and tartaric acid change according to the amount of water available for each treatment.

The Total Polyphenols Index (TPI) is based on the absorbance of the sample at a wavelength of 280 nm, whose maximum absorbance occurs for substances characteristic of the benzene cycle, which represent the majority of phenolic compounds in the grape. According to the treatment samples, it is possible to say that the treatments did not present significant differences among them, with little difference between average values for all treatments, with TFC50 being the treatment that presented the highest value with 57.43 mg/ml (Table 6).

The Folin C index is used to observe the absorbance of phenolic and polyphenolic antioxidants, reacting with the grape reducing substances, thus measuring the total reduction capacity of a sample. According to the results, there were no significant differences between treatments, despite the TFC25 (274.00 EAG) showing a higher value than the others (Table 6).

However, it is possible to observe that the treatments that received less water through irrigation (TF25) were the ones that presented the highest amount of compounds, and decreased successively according to the amount of water applied (TF50 presents higher values than the control treatment TF100).

The color intensity is defined as the sum of the optical densities, measuring the wavelengths, indicating the combination of the phenolic compounds present in the musts. The treatments did not present significant differences between them. TFC25 was the one with the highest mean value (3.50 mg/ml), while TFC100 was the treatment with the lowest mean value (1.97 mg/ml) (Table 6). It is important to note that like OD 520, TF25 treatments showed greater results, even if not significant, when compared to other treatments.

The amount of anthocyanins (red pigment) that is responsible for the color of the wines, did not showed significant differences among the treatments, regardless of the amount of water by the irrigation that was applied, and the training system. The treatment with the highest mean values was TFC25 with 69.67 mg L^{-1} , while the TFG50

was the one with the lowest mean values 42.67 mg L⁻¹ (Table 6). Observing values of the same training system for Cordon Royat, the plants that received less water, were the ones that produced the greatest amount of anthocyanins, while for Guyot, TFG25 was the best, followed by TFG100, and TFG50 was the one with the lowest average values (Table 6).

The accumulation of sugars in the berry is a very important phenomenon in the maturation phase, potentiating the subsequent formation of other compounds, such as the increase in anthocyanins, aromas and polyphenols. The increase in concentration is directly related to photosynthetic activity, which depends on the availability of water in the plant (Brandão, 2018).

The amount of water will influence the levels of sugars available in berries, since it is the first year of work and observing the results. It was possible to verify that the treatments that received the greatest amount of water were those that presented the greatest amount of SST. The accumulation of SST is important as shown by Brandão (2018). Therefore, it is necessary to observe long term for the treatments that receive less amount of water (TFG25 and TFC25), if they will show any sugar deficiency when compared to treatments that receive more water.

Table 5 - Must quality at harvest for the parameters of Total Soluble Solids - TSS (°Brix), pH, total acidity (g L⁻¹ of tartaric acid), tartaric acid (g L⁻¹), malic acid (g L⁻¹). Each treatment had an average of three repetitions of must. Mean values (\pm standard deviation) followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test.

Treatment	TSS (°Brix)	pH	Total Acid	Tartaric Acid	Malic Acid
TFC25	18,90 \pm 0.69 a	3.22 \pm 0.08 a	5.00 \pm 0.27 a	5.97 \pm 0.32 a	0.90 \pm 0.30 a
TFC50	20.00 \pm 1.25 ab	3.23 \pm 0.10 a	5.35 \pm 0.40 a	6.10 \pm 0.35 a	1.00 \pm 0.10 a
TFC100	20.20 \pm 0.36 ab	3.28 \pm 0.01 a	4.73 \pm 0.20 a	5.53 \pm 0.06 a	0.93 \pm 0.12 a
TFG25	19.50 \pm 0.20 ab	3.24 \pm 0.03 a	5.23 \pm 0.09 a	6.20 \pm 0.26 a	0.80 \pm 0.17 a
TFG50	19.83 \pm 0.12 ab	3.28 \pm 0.05 a	4.74 \pm 0.29 a	5.77 \pm 0.06 a	0.90 \pm 0.20 a
TFG100	21.00 \pm 0.96 b	3.31 \pm 0.07 a	5.05 \pm 0.57 a	5.97 \pm 0.38 a	0.83 \pm 0.32 a

Table 6 - Must quality at harvest for the parameters of TPI (mg/ml), Folin C Index, color intensity (mg/ml), anthocyanins (mg L⁻¹). Each treatment had an average of three repetitions of must. Mean values (\pm standard deviation) followed by different letters are significantly different at $P \leq 0.05$, according to Tukey HSD test.

Treatment	TPI	Folin C Index	Color Intensity	Anthocyanins
TFC25	54.13 \pm 8.09 a	274.00 \pm 59.07 a	3.50 \pm 1.31 a	69.67 \pm 36.17 a
TFC50	57.43 \pm 3.97 a	270.13 \pm 12.58 a	2.63 \pm 0.65 a	62.33 \pm 12.22 a
TFC100	56.93 \pm 0.67 a	174.40 \pm 41.40 a	1.97 \pm 0.25 a	48.33 \pm 21.55 a
TFG25	56.10 \pm 2.98 a	236.50 \pm 52.24 a	3.30 \pm 0.10 a	63.33 \pm 13.20 a
TFG50	56.77 \pm 2.41 a	212.07 \pm 27.37 a	2.63 \pm 0.57 a	42.67 \pm 10.07 a
TFG100	58.67 \pm 9.10 a	189.27 \pm 96.90 a	2.73 \pm 1.33 a	52.33 \pm 23.69 a

The acidity determines the organoleptic characteristics and ensures the protection of musts and wines against the occurrence of diverse disorders in its correct natural evolution (Curvelo-Garcia and Catarino, 2015). Variations in total acidity occur mainly due to the variation and degradation of malic and tartaric acids (Queiroz, 2002). The degradation of tartaric acid generally decreases steadily throughout maturation while the concentration of malic acid is somewhat conditioned by factors such as temperature and the availability of water and nitrogen in the soil (Magalhães, 2015).

5 CONCLUSIONS

The physiological parameters showed that the plants of the treatments that received the highest irrigation amounts were the ones that presented the highest water potential values, stomatal conductance, transpiration and net photosynthetic rates while the treatments that received the least irrigation allocation were the ones that presented the lower values. The differences between the Guyot and Cordon Royat training systems, were not statistically significant on these physiological parameters.

Regarding yield, the treatments that received the most water were the ones that showed the greatest results. The TFC25 was the one treatment that showed good results both in productivity and in bunches production and it was the one that best used the water received, while the TFG25 treatment showed the worst results for the same parameters.

Evaluating the size and quality of the berries, the treatments did not differentiate among themselves, Guyot and Cordon Royat presented similar values for the evaluated parameters, with the exception of TSS that the TFC25 with less sugar than the others, while TFG100 was the one with the highest values in this parameter.

In conclusion, it is possible to say that the evaluated training systems present some differences, but in general, they do not present significant differences between them. However, when we change the amount of water applied in irrigation, we can conclude that, quantitatively. The vines that receive more water showed a better performance, with the exception of vines under TFC25 treatment.

These results refer to the first year of the project and it is necessary to carry out the experiment over several years in order to consolidate the results.

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Annex

Phenological growth stages and BBCH-identification keys of grapevine (*Vitis vinifera* L. ssp. *vinifera*). Lorenz et al., 1995.

Code / Description

Principal growth stage 0: Sprouting/Bud development

00 Dormancy: winter buds pointed to rounded, light or dark brown according to cultivar;

Bud scales more or less closed according to cultivar;

01 Beginning of bud swelling: buds begin to expand inside the bud scales

03 End of bud swelling: buds swollen, but no green

05 "Wool stage": brown wool clearly visible

07 Beginning of bud burst: green shoot tips just visible

08 Bud burst: green shoot tips clearly visible

Principal growth stage 1: Leaf development

11 First leaf unfolded and spread away from shoot

12 Second leaves unfolded

13 Third leaves unfolded

1. Stages continuous until...

19 Nine or more leaves unfolded

Principal growth stage 2: ———

Principal growth stage 3: ———

Principal growth stage 4: ———

Principal growth stage 5: Inflorescence emergency

53 Inflorescences clearly visible

55 Inflorescences fully developed: flowers separating

Principal growth stage 6: flowering

60 First flowerhoods detached from the receptacle

61 Beginning of flowering: 10% of flowerhoods fallen

62 20% of flowerhoods fallen

63 Early flowering: 30% of flowerhoods fallen

65 Full flowering: 50% of flowerhoods fallen

66 60% of flowerhoods fallen

67 70% of flowerhoods fallen

68 80% of flowerhoods fallen

69 End of flowering

Principal growth stage 7: Development of fruits

71 Fruit set: young fruits begin to swell. remains of flowers lost

73 Berries goat-sized, bunches begin to hang

77 Berries beginning to touch

79 Majority of berries touching

Principal growth stage 8: Ripening of berries

81 Beginning of ripening: berries begin to develop variety-specific colour

83 Berries developing colour

85 Softning of berries

89 Berries ripe for harvest

Principal growth stage 9: Senescence

91 After harvest: end of wood maturation

92 Beginning of leaf discolouration

93 Beginning of leaf-fall

95 50% of leaves fallen

97 End of leaf-fall

99 Harvested product
