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Water stress, yield, and grape quality in a hilly rainfed "Aglianico" vineyard grown in two different soils along a slope

R. Albrizio [a], À Puig-Sirera [a,b,*], M.H. Sellami [a,c], G. Guida [a], A. Basile [a], A. Bonfante [a], A. Gambuti [d], P. Giorio [a]

[a] Institute for Mediterranean Agricultural and Forestry Systems, National Research Council of Italy, P.le Enrico Fermi 1, 80055 Portici, Naples, Italy
[b] Università di Pisa, Dipartimento Scienze Agrarie, Alimentari e Agro-Ambientali, Via del Borghetto 80, 56124 Pisa, Italy
[c] ARCA2010, Contrada Varignano, 81, 81100 Acerra, NA, Italy
[d] Department of Agricultural Sciences, Grape and Wine Science Division, University of Napoli 'Federico II', Viale Italia, 83100 Avellino, Italy

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ABSTRACT

Vineyards from hilly areas of the Mediterranean region are mostly grown in shallow soils that present low soil water holding capacity. These vineyards are prone to water stress conditions, which if well managed can raise the grape quality potential. This study aimed to investigate the water stress development in an "Aglianico" vineyard grown along a 90 m slope. The two-year (2011–2012) trial was conducted in two soils having different hydraulic properties, the Up-slope with lower soil water holding capacity than the Down-slope site. The results showed that grapevines were more stressed in the Up-slope soil than in the Down-slope soil, as reflected by the higher crop water stress index, lower leaf water potential and leaf gas exchanges values. Consequently, the yield was significantly lower by 40% in the Up-slope, which was determined by the lower weight and volume of berries. The smaller berries improved must quality parameters of total soluble solids, total polyphenols, total anthocyanins, and color intensity within a range of 4–25% higher in the Up-slope compared to the Down-slope site. Moreover, the pre-veraison stress experienced in 2012 reduced yield by 30% and depressed berry weight and volume, compared to 2011. The post-veraison stress induced the improvement of must quality, mainly in the Up-slope 2011. Interestingly, there was no significant difference in the pH and titratable acidity between both sites, which indicates the ability of Up-slope vines to make up for more stressful conditions, and, thus, their resilient behavior to maintain their high-quality wine. This study highlights that vineyards in hilly areas y benefit from a differentiated management between different viticulture zones to bring up their high-quality wine.

1. Introduction

Grapevine (*Vitis vinifera*, L. subsp. *Vinifera*) is a woody perennial crop that has been cultivated for more than 8000 years in the Mediterranean region, and still nowadays constitutes one of the main elements of its agricultural sector (Novara et al., 2021).

The planning and management of grapevine towards high quality wines are typically carried out using viticultural zoning procedures (Carey, 2001; Gladstones and Smart, 1997; Vaudour, 2003). These are an extension – at a finer scale – of the standard concept of terroir (Bonfante and Brillante, 2022), defined as "a spatial and temporal entity with homogeneous or outstanding grape and/or wine, soil landscape and climate characteristics within a territory, marked by socio-cultural technical choices" (Vaudour, 2003). Within each of these viticulture zones, it is expected that the specific pedoclimate and specific management affects grapevine physiology creating in turn unique wines in each territory (Van Leeuwen et al., 2004; García-Navarro et al., 2022). The environment of the Mediterranean region that fosters potential high-quality wine and grape production has been related to vineyards in hilly areas and rainfed farmed (Hofmann et al., 2014; Prosdocimi et al., 2016). Grapevine in this region is well adapted to marginal areas with sloping, shallow, well-drained, and poor fertile soils, and thus, with a higher risk to develop water deficit (Lazcano et al., 2020). Consequently, grapevines are suited to the lack of water and recurrent drought, and thus, it plays a role in the adaptation to climate change by agriculture (Santillán et al., 2020).

* Corresponding author at: Institute for Mediterranean Agricultural and Forestry Systems, National Research Council of Italy, P.le Enrico Fermi 1, 80055 Portici, Naples, Italy

E-mail address: angela.puig@unipi.it (À. Puig-Sirera).

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Previous research have strengthened the hypothesis that the physical soil properties and geomorphological features, affect the final yield and grape quality characteristics, even more than the soil chemical properties (Poni et al., 2018). There is a consensus that steep sloped, coarse textured, and stony soils confer the best wine quality due to their lower soil water holding capacity which increases the grapevine water deficit (Poni et al., 2018). Water deficit conditions generated in these soils would boost earlier shoot-growth slackening, smaller berry size, high grape sugar and anthocyanin concentrations, thus rising grape quality potential (Bonfante et al., 2017; Brillante et al., 2017; Lazcano et al., 2020; Van Leeuwen et al., 2004).

The impact of water stress on vine growth and yield fulfillment has been demonstrated as highly dependent on the intensity of the stress, the phenological stages, and greatly variable within short distances in the same field, especially in vineyards grown in sloped soils (Bonfante et al., 2017; Brillante et al., 2017; Wenter et al., 2018; Basile et a, 2020). This temporal and local soil variability has significant consequences not only in the physiological processes such as photosynthesis and stomatal conductance, but also, on the synthesis of grape compositional factors such as sugar, acids, anthocyanins, and tannins, and must composition (Pereyra et al., 2022).

Pre- and post-veraison stages in grapevines are the most sensitive phenological phases to the effects of water deficit on yield and grape quality. However, results are not consistent among studies, especially for post-veraison stress (Poni et al., 2018). The stress effects in these two phases are interrelated because deficit in pre-veraison could cause a greater post-veraison water deficit due to their cumulative effect (Van Leeuwen et al., 2004; Casassa et al., 2015). The same magnitude of water deficit has been reported to affect canopy growth, yield, and grape quality more when occurring in pre- than post-veraison (Girona et al., 2009; Palai et al., 2021; Wenter et al., 2018). This is mainly related to the increased berry sensitivity to the soil water deficit and, therefore to the vine water stress before veraison because the xylem is the main source of water for the berry instead of the phloem, which becomes the unique berry-stem hydraulic connection after veraison (Munitz et al., 2017). In addition, pre-veraison water stress can restrain cell division, and thus the total cell numbers, which irreversibly decreases berry growth, while late water stress can only produce a reversible negative effect on cell size (Girona et al., 2009; Ojeda at al, 2002; Wenter et al., 2018; Palai et al., 2021). Berry size diminution increases the skin-to-pulp weight ratio, which results in higher concentrations of phenolic and anthocyanins compounds in the berry skin, and lower concentration of total soluble solids (Ojeda et al., 2002). The aroma composition of berries is also affected in relation to the phenological stage as glycosilated VOCs were enhanced when water deficit occurred in pre-veraison, while no effect was detected for post-veraison stress (Palai et al., 2022).

The greater variability reported in the literature about the effects of post-veraison stress on the grapevine and wine performance can be attributed to the different intensity of the applied stress and the sensitivity of the cultivar. In this way, Girona et al. (2009) observed an improvement in the grape quality of *cv* Tempranillo only when post-veraison water deficit at midday did not decrease below − 1.12 MPa leaf water potential threshold. Similar results were reported in the same cultivar by Intrigliolo et al. (2012), who informed that an excessive water deficit in post-veraison reduced leaf photosynthesis and thus impaired sugar accumulation. In another research conducted in potted Shiraz vines in south of France, Ollé et al. (2011) found that post-veraison stress caused a decrease in berry weight, whilst no effects were found in pre-veraison stress as it was in the full irrigated vines. More recently, Geng et al. (2022) in Cabernet Sauvignon grown in semi-arid area of China, showed improved grape quality when water stress was mild from fruit setting to veraison or mild-severe during post-veraison. Consequently, comparing studies and drawing general conclusions about grapevine adaptation to water stress remains difficult to establish.

Even though vineyards in the Mediterranean region are characteristic of hilly areas, most of the conducted research dealing with water deficit reported the outcomes of experiments conducted in flat vineyards (Bota et al., 2016; Intrigliolo et al., 2012; Edwards and Clingeleffer, 2013), or compared the performance of vineyards grown in two separate sloped and flat fields (Zsófi et al., 2009).

In a Mediterranean hilly viticultural area, in a two-year experiment, the influence of different soil hydrological characteristics along a slope with the same textural class but different hydraulic properties on the vineyard performance was highlighted by our research group (Bonfante et al., 2015, 2017). Moreover, through a modeling approach, we confirmed that rather than the slope gradient, the different soil hydrological properties between Up- and Down-sloped soils were the main factors in driving the soil water potential and the vine water stress (Basile et al., 2020).

In the same experimental vineyard, we continued the previous research conducted by Basile et al. (2020) as there is an evident gap in knowledge on the performances of rainfed vineyards grown in sloped soils. Indeed, the data on the grape yield and must quality responses to different water stress conditions in vines grown at short distances along a slope are scarce. Therefore, the main aim of this paper was to study the differences associated with the development of water stress conditions between rainfed vines grown in two different soils along a slope, in terms of physiological responses, grape yield, and must quality. Moreover, the paper studied the effects that pre- and post-veraison stress could have on the above parameters of the two sites.

2. Materials and methods

2.1. Study area

The two-year research (2011 and 2012) was carried out at Quintodecimo farm, which produces high-quality wine and is located in a hilly environment of southern Italy (Mirabella Eclano – AV, Campania region: Lat. 41.047808°, Lon 14.991684°, Elev. 368 m a.s.l.).

The experimental area is characterized by a typical Mediterranean climate, with a mean daily temperature of 14.7 °C (± 0.9) and mean annual rainfall of 802 mm (± 129 mm) (averaged data from 2003 to 2013 recorded by the regional weather station of Mirabella Eclano at about 1 km distance from the farm).

The main weather parameters, including global radiation, air temperature, relative humidity, rainfall, and wind speed at 2 m above the ground were collected from an agrometeorological station installed on the farm. The daily reference evapotranspiration (ET_o) was calculated by the FAO-Penman Monteith equation (Allen et al., 1998). The daily weather data during two experimental seasons (reported as decadal data) are shown in Fig. 1 in terms of average air temperature (T_{avg}), average relative humidity (RH_{avg}), global radiation (R_g), rainfall, and reference evapotranspiration (ET_o). The 2012 season between May and the end of September was characterized by slightly warmer and drier weather than 2011: T_{avg}, ET_o and R_g on daily basis were respectively 0.5 °C, 7% and 14% higher, and the rainfall was 198 mm in 2011 and 144 mm in 2012.

The total rainfall in 2011 (550 mm) was practically the same as in 2012 (553 mm), quite lower than the ten-year average of 802 mm year^{-1} recorded from 2003 to 2013. In 2011 three spells of rainfall occurred in mid-September and mid-November of about 30 mm, while in 2012 the biggest spell of rainfall was 60 mm during mid-July (Basile et al., 2020).

The experiment was performed during 2011 and 2012 in a rainfed *Vitis vinifera* L. cv "Aglianico" vineyard, that was 10 years old, grafted on *V. berlandieri* Planch. x *V. rupestris* Scheele (1103 P) and trained with a spurred cordon. The rows were oriented NW-SE (320°) and the vines were spaced 2 m between rows and 1 m along the rows (i.e., 5000 plants per hectare).

The monitored vines were located along a slope (length=90 m, inclination β = 10°) in two different soils of the vineyard, named the Up-

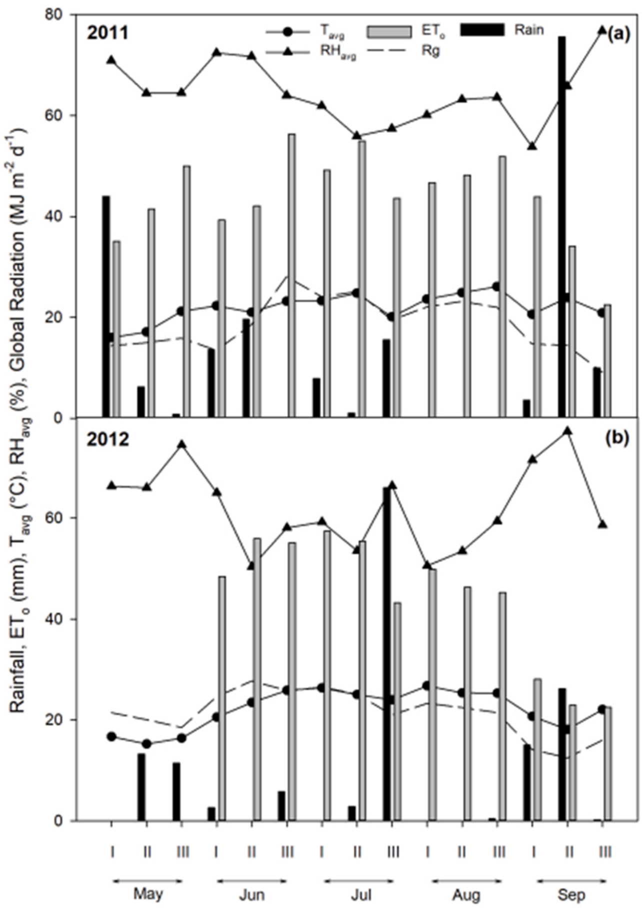

Fig. 1. 10-day average of air temperature (T_{avg}; °C), relative humidity (RH_{avg}; %) and global radiation (R_g; MJ m^{-2} day^{-1}), and 10-day total rainfall (mm), and reference evapotranspiration (ET_o; mm), during the experimental seasons (May - September) of 2011 (a) and 2012 (b).

slope and the Down-slope sites. These two experimental plots were chosen after identifying them in two functional homogeneous soils by Bonfante et al. (2015). Thus, the two soils are classified according to the IUSS Working Group WRB (2014) as Calcisol (Clayic, Aric) in the Up-slope site, and Cambisol (Clayic, Aric, Colluvic) in the Down-slope site. The two soils, despite their same textural class (clay loam), show different soil hydrological behavior due to the different water retention and hydraulic conductivity curves of their soil horizons. The Down-slope soil had 20% more water storage capacity (i.e., 145 mm of available water content, AWC) than the Up-slope soil (80 mm of AWC) (Bonfante et al., 2015, 2017; Basile et al., 2020). Parameters of soil hydraulic properties along the profile of the two soils are reported in Basile et al. (2020).

2.2. Field and laboratory measurements

2.2.1. Soil matric potential

The average soil matric potential in the rooting zone of the Up-slope and Down-slope sites was estimated based on the results of the calibrated and validated Hydrus 2D/3D model, as detailed in Basile et al. (2020).

2.2.2. Plant water status

Leaf water potential (Ψ_l, MPa) was measured during the season using a Scholander type pressure bomb (SAPS II, 3115, Soilmoisture Equipment Corp., Santa Barbara CA, USA). The measurements were taken on 10 well-expanded leaves (1 leaf/vine) for each site and started at 10:00 until 12:00 (solar time).

The potential Crop water stress index (CWSI) was derived from the outputs of the Hydrus 2D/3D model (Šimůnek et al., 2016), used to simulate two-dimensional water flow in Soil-Plant-Atmosphere continuum (Basile et al., 2020). The potential CWSI was calculated as relative transpiration deficit (Kozak et al., 2006) and used as a hydrological indicator (Alfieri et al., 2019), was given as follows (Basile et al., 2020):

$$CWSI = \left[1 - \frac{T_{cact}}{T_c}\right] \times 100 \tag{1}$$

Where $T_{c\ act}$ is the actual crop transpiration (mm) and T_c is the potential crop transpiration (mm).

2.2.3. Gas exchanges and fluorescence

The photosynthetic CO_2 assimilation rate (A, µmol m^{-2} s^{-1}) at saturating light, stomatal conductance to water vapor (g_s, mol m^{-2} s^{-1}) and actual quantum yield of PSII (ϕ_{PSII}, r.u.) were measured using a portable open-system gas-exchange Analyzer Li-6400XT (Li-Cor Biosciences, Lincoln, NE, USA) on fully expanded and light-exposed leaves of 10 vines on each site (1 leaf/vine). Intrinsic water use efficiency (iWUE) was calculated as A/g_s (µmol mol^{-1}). The CO_2 inside the leaf chamber was set to 400 µmol mol^{-1} by means of an external bottled CO_2 source. A LED light source with emission peaks at 630 and at 460 nm provided a photosynthetic photon flux density (PPFD) at 2000 µmol (photons) m^{-2} s^{-1} (90% red, 10% blue). The instrument software calculated the gas-exchange parameters based on the von Caemmerer and Farquhar (1981) model, and the actual quantum yield according to Genty et al. (1989). Measurements were taken between 10:00–12:00 (solar time).

2.2.4. Yield components, berries and must composition

In each experimental site, 27 vines were visually selected as being representative of the entire experimental plot, and at harvest (2nd October in both years) total yield, weight, and volume of 100 berries were determined on 12 of these selected plants. From the 15 vines left of each experimental site, a sample of 200 berries was used to determine berry total soluble solids (TSS) concentration, pH, titratable acidity, total anthocyanins, total polyphenols, and total tannins of berries during the ripening period (from berry color change to harvest).

The polyphenol extraction from grapes was done as follows: the extraction of berry components was carried out in duplicate while simulating the maceration process necessary to produce red wines (Mattivi et al., 2002; Vacca et al., 2009). Briefly, berries (200 g) were cut in two with a razor blade, and seeds and skins were carefully removed from each berry-half. The pulp on the inner face of the berry skin was removed using an end-flattened spatula trying to preserve the integrity of berry skin. Skins and seeds were immediately immersed in a 200 mL solution consisting of ethanol: water (12:88 v/v), 100 mg/L of SO_2, 5 g/L tartaric acid, and a pH value adjusted to 3.2 (with NaOH) and extracted for five days at 30 °C. The extracts were shaken by hand once a day. Skins and seeds were removed from the hydro-alcoholic solution after five days and the skin extract was centrifuged for 10 min at 3500g. Extracts were poured into dark glass bottles, flushed with nitrogen, and stored at 4 °C until spectrophotometric analyses.

The chemical analyses and spectrophotometric measurements of must, skin and seeds extracts, and wine were done as follows: Standard chemical analyses (soluble solids, total acidity, pH, total polyphenols (Folin-Ciocalteau Index) and Absorbances (Abs) were measured according to the OIV Compendium of International Methods of Analysis of Wine and Musts (OIV, 2016). Color intensity (CI) and hue were evaluated according to the Glories method (Vivas, 1998). Total anthocyanins were determined by the spectrophotometric method based on SO_2 bleaching (Ribéreau-Gayon and Stonestreet, 1965). Tannins were determined according to Ribéreau-Gayon and Stonestreet (1965). Analyses were performed in duplicate using basic analytical equipment and a Shimadzu UV-1800 (Kyoto, Japan) UV spectrophotometer.

2.3. Statistical analysis

Each dependent variable was preliminarily evaluated for normal distribution according to Shapiro–Wilk's test, if the normality assumption was violated, we transformed data into Box-Cox transformation. Combined analyses were run over 2011 and 2012, after verifying the homogeneity of error variances using Bartlett's chi-square test (Gomez and Gomez, 1984). The missMDA package (Josse and Husson, 2016) in R studio software (R Core Team, 2013) was used to pair an unbalanced dataset due to an unequal number of observations of the dependent variables.

Two-way non-parametric analysis by applying Scheirer–Ray–Hare test was carried out to evaluate the effect of the different treatments (i.e., slope position and year) and their interactions on yield and quality parameters of grapevine. This analysis was performed using the software package rcompanion in R studio software. When Scheirer–Ray–Hare test found significant differences, pairwise Dunn test with Bonferroni corrections were performed using the software package PMCMR in R studio software (R Core Team, 2013).

The yield, the soil matric potential (SMWP), the crop water stress index (CWSI), the leaf physiological, and the physical and quality grape parameters were subjected to Principal Component Analysis (PCA) to explore relationships among these variables and the treatments (i.e., slope position and year), and to analyze which variables were more effective to discriminate the treatments.

The PCA analysis was carried out using the software package FactoMineR (Husson et al., 2014) in R studio software (R Core Team, 2013). All package used in this statistical analysis are available via the Comprehensive R Archive Network (CRAN, https://cran.r-project.org).

3. Results and discussion

3.1. Potential CWSI, leaf water potential and leaf gas exchanges

At the flowering stage (Fig. 2a-d) of both years, the vines did not experience any stress in either site in terms of both indicators, potential CWSI and Ψ_l, as both soils had an optimum water content availability, because of the rainfalls in winter as reported by Bonfante et al. (2015) and in May (Fig. 1).

During the pre-veraison of 2011, the CWSI was less than 10% in both sites (Fig. 2a), while the Ψ_l in the Up slope began to be significantly lower than the Down-slope site at DOY 188 (Fig. 2c). The CWSI and the Ψ_l are crucial indicators for vine water status (Fuentes et al., 2012; Mirás-Avalos and Intrigliolo, 2017). According to the water stress thresholds based on leaf water potential (van Leeuwen et al., 2009), the Up-slope vines depicted weak water stress (Ψ_l between −0.9 and −1.1 MPa), while the Down-slope ones did not show any stress (Ψ_l >−0.9 MPa). In 2012, the CWSI at the pre-veraison was much higher than the one in 2011, reaching 56% in the Up slope (Fig. 2b), with the corresponding Ψ_l equal to − 1.35 MPa, which matches with moderate to severe stress (Ψ_l between −1.3 and −1.4 MPa) in pre-veraison (Fig. 2d). In the same way, in the Down-slope site, the CWSI achieved values of 27% and the Ψ_l was equal to − 1.17 MPa, depicting weak to moderate stress (van Leeuwen et al., 2009).

During post-veraison, in the Up-slope the maximum CWSI of 61% and the lowest Ψ_l of − 1.67 MPa occurred in 2011 (Fig. 2a, c), indicating higher stress conditions than 2012. However, the Up-slope vines reached levels of severe water stress (Ψ_l < −1.4 MPa) in the post-veraison stage in both years (Fig. 2c, d). In the same stage, the Down-slope site achieved on average the maximum values of 32% CWSI and around − 1.40 MPa Ψ_l in both years, which agreed with moderate-severe water stress levels.

The Ψ_l values registered along the season in both years fall within the common ranges for Ψ_l in pre-veraison of − 0.8 MPa till achieving − 1.7 MPa in post-veraison stage in vineyards cultivated in semi-arid environments, as reported by other authors (e.g., Schultz, 2003; Centeno et al., 2010). Although CWSI and Ψ_l are independent indicators, they followed the same dynamics along the experimental seasons and across treatments. In addition, both were able to discriminate different timing of stress occurrence and its intensity between the Up and

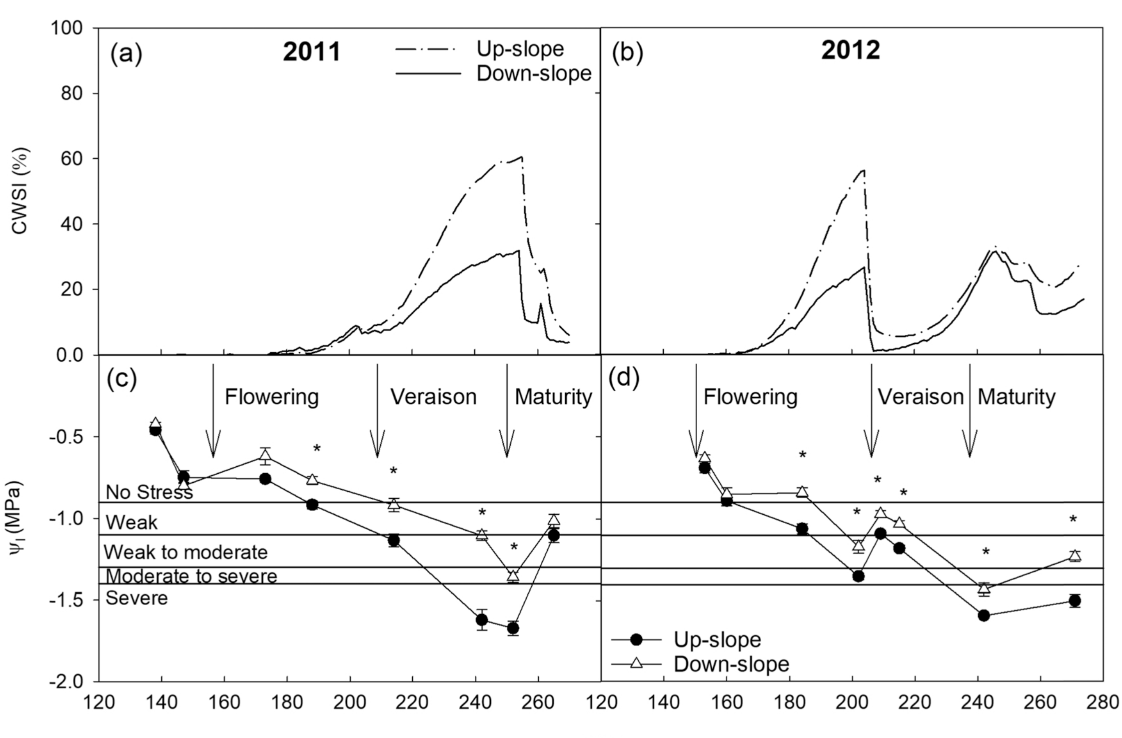

Fig. 2. Potential crop water stress index, CWSI (a, b) and midday leaf water potential, Ψ_l (c, d) on the Up- and Down-slope sites in 2011 (left panels) and 2012 (right panels). Bars indicate standard error of the mean. Asterisks indicate significant effect of slope position at P ≤ 0.05.

Down-slope sites. A positive correlation between CWSI and stem water potential with the slope position in the steep slope vineyards of Chianti terroir (Tuscany, Italy) has been reported also by Puig-Sirera et al. (2021).

It is well known that field-grown grapevines when submitted to long-term water deficit show a strong reduction of stomatal conductance and CO_2 assimilation (Escalona et al., 2000). In our experiment, stomatal conductance (g_s) and leaf CO_2 assimilation rate (A) resembled the general dynamics of Ψ_l of the two slope positions in both experimental years (Fig. 3a-d; Fig. 2c-d). Hence, both parameters were significantly lower in the Up- than in the Down-slope vines after the flowering stage (DOY 157 and 151 in 2011 and 2012, respectively), and both decreased along the season with progressive drought. Quite low values of g_s (0.038 mol m^{-2} s^{-1}) and A (3.2 mol m^{-2} s^{-1}) were registered in the Up-slope at the end of the pre-veraison 2012 (DOY 202). A rainfall event during veraison (DOY 215) allowed a significant recovery followed by a strong decrease to the lowest values of g_s and A at grape maturity (g_s =0.018 mol m^{-2} s^{-1}, A= 2.2 µmol m^{-2} s^{-1}) (Fig. 3b, d). The stress during pre-veraison could have

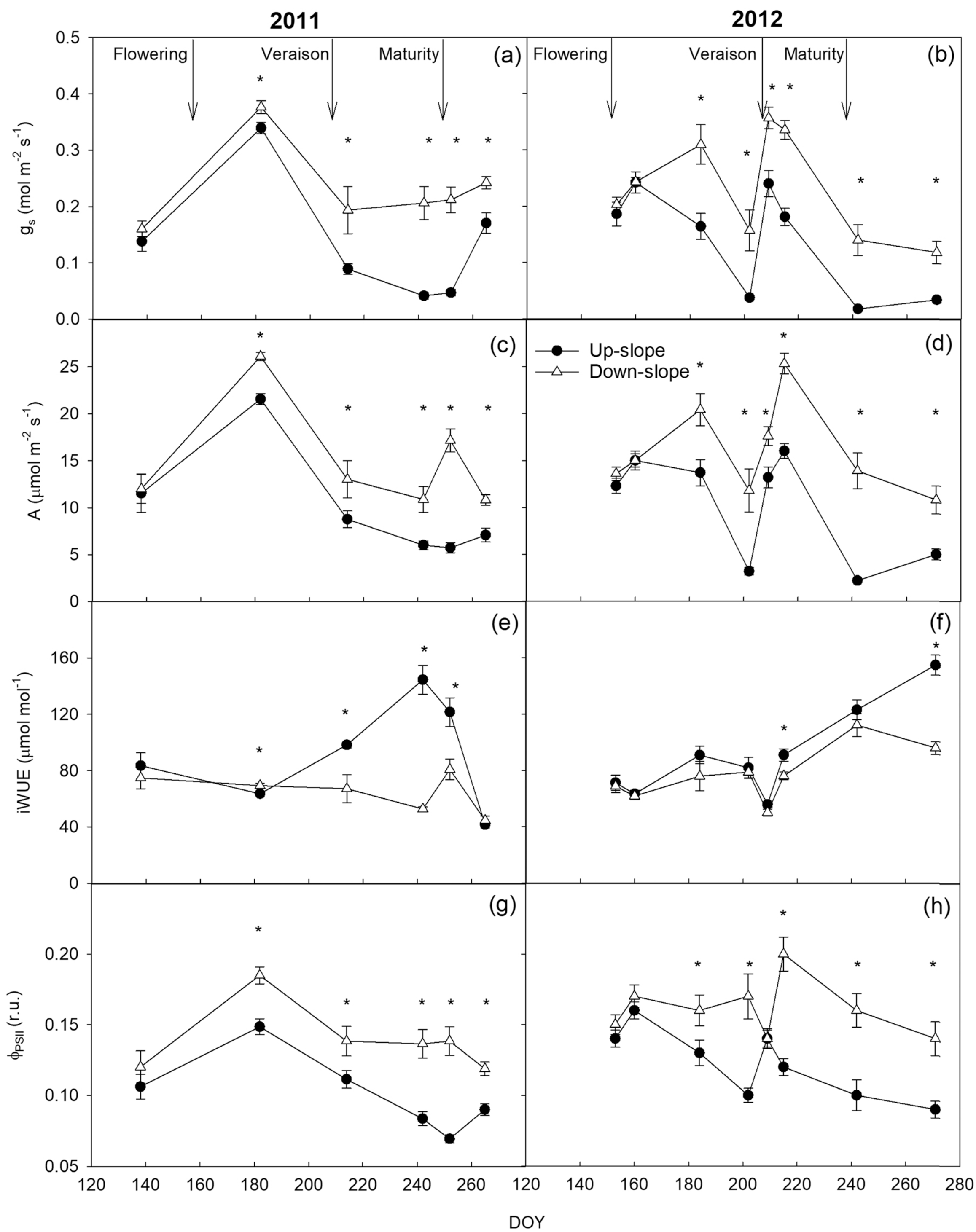

Fig. 3. Stomatal conductance, g_s (a, b), CO_2 assimilation, A (c, d), intrinsic water use efficiency, iWUE (e, f) and PSII quantum yield, (ϕ_{PSII}), (g, h) on the Up- and Down-slope sites in 2011 (left panels) and 2012 (right panels). Bars indicate standard error of the mean. Asterisks indicate significant effect of slope position at $P \leq 0.05$.

impaired both the rate and the extent of recovery after the rainy event (Flexas et al., 2009; Lovisolo et al., 2010), and thus exacerbated the subsequent decrease in the gas exchanges of the Up-slope vines.

These g_s data indicate severe vine water stress conditions in the Up-slope vines at both pre-and post-veraison, as they are lower than the threshold 0.05 mol m^{-2} s^{-1} for severe water stress, according to Cifre et al. (2005). In addition, g_s and A in the Up-slope vines were also low at grape maturity in 2011 (g_s =0.042 mol m^{-2} s^{-1}; A=5.74 μmol m^{-2} s^{-1}), indicating that severe water stress conditions also occurred in post-veraison of 2011. During this stage, we observed the maximum difference in intrinsic WUE (iWUE) (A/g_s) between Up- and Down-slope vines, mainly due to the strong g_s reduction in the Up-slope (Fig. 3e). Conversely, in 2012 differences in iWUE between the two sites were less marked during most of the sea son (Fig. 3f). The decrease of both stomatal conductance and photosynthesis and the increase of iWUE in grapevines with increased conditions of water deficit has been largely reported in many studies (e.g., Bota et al., 2016; Brillante et al., 2016; Palai et al., 2021; Poni et al., 2009).

The leaf gas exchange values of the Down-slope vines followed the same pattern as the ones in the Up-slope vines, although achieving moderate water stress levels in both experimental years (Cifre et al., 2005). The higher stress conditions in the Up-slope vines than in the Down-slope are due to the lower soil water retention capacity encountered in the Up-slope soil, as demonstrated in previous works in the same experimental sites (Basile et al., 2020; Bonfante et al., 2015, 2017). Similarly, Zsófi et al. (2009) found significant differences in soil water availability between flat and steep-slope vineyards, which were reflected in lower pre-dawn leaf water potentials, g_s and A values in the steep ones.

In addition, the severe water stress experienced in the upper soil vines, which caused a lower leaf CO_2 assimilation rate, was at the same time reflected in reduced vegetative growth. In fact, along the season, LAI in Up-slope averaged 1.18 m^2m^{-2} (± 0.21) in 2011, 12% lower than Down-slope, while in 2012 LAI averaged 1.36 m^2m^{-2} (± 0.17), 22% lower than Down-slope vines (data not shown).

The PSII quantum yield (ϕ_{PSII}) was assessed through modulated fluorescence and resembled the A rates (Fig. 3g-h). The Up-slope ϕ_{PSII} decreased to a minimum of 0.07 (r.u.) near grape maturity (DOY 252) of 2011, while in the same period the Down-slope vines showed double values (Fig. 3g). Rainfalls occurring near veraison in 2012 also induced a recovery of ϕ_{PSII} in both slope positions. Likewise, A and g_s, the Down-slope vines reached a maximum ϕ_{PSII} (0.2 r.u.) at DOY 215 (Fig. 3h), which is comparable with values measured during the flowering stage in 2011 (Fig. 3g), when no water stress was detected (Fig. 2c). Such a recovery of ϕ_{PSII} in 2012 was followed by a gradual decrease, with the lowest 0.09 (r.u.) value reached by the Up-slope vines in the late season; such value was about half of the one measured in the Down-slope (DOY 271, Fig. 3h). Ju et al. (2018), also reported a decrease in ϕ_{PSII} which can induce photoinhibition due to a light excess under water stress conditions. Maroco et al. (2002) reported for Tempranillo cv. during mid-summer a quantum yield of PSII about 0.4 in the control and 0.3 in the stressed vines at midday, because of earlier stomatal closure in the latter vines.

Therefore, both gas-exchanges and fluorescence data confirmed the intense stress conditions encountered in the Up-slope vines. The close link between impaired gas-exchanges, namely photosynthesis, caused by decreased soil water availability, and grape yield reduction is well established in the literature (Medrano et al., 2003).

3.2. Yield and its components

Grapevine yields were significantly influenced by the slope position (S) (p ≤ 0.001) and year (Y) (p ≤ 0.01) (Table 1). As expected, the less water-stressed Down-slope vine yielded significantly better (+ 40%) than the Up-slope vine. In addition, the yield of 2011 was significantly higher by 30% than the one of 2012.

Consequently, S and Y also significantly affected the related yield parameters of berry weight and volume. Both parameters mimicked the yield results and were higher in the Down-slope and 2011. Thereby, the differences in yield across treatments and years were mainly determined

Table 1
Yield and grape quality parameters as affected by slope position (S) and year (Y).

Source of variance	Yield (g $plant^{-1}$)	Berry weight (g)	Berry volume (cm^3)	TSS (°Brix)	pH	Titratable acidity (g L^{-1})	Intensity (Abs units)	Tonality (Abs 420 nm/ Abs 520 nm)	Total anthocyanins (mg L^{-1})	Total polyphenols (mg L^{-1})	Total tannins (g L^{-1})
Slope position (S)											
Up-slope	1010 ± 209 b	177.5 ± 11.6 b	164.0 ± 9.8 b	22.98 ± 0.41 a	3.32 ± 0.06 a	6.2 ± 0.4 b	5.38 ± 0.51 a	0.52 ± 0.01	610.2 ± 52.2 a	2014 ± 195 a	2.69 ± 0.14 a
Down-slope	1437 ± 378 a	186.9 ± 27.8 a	175.7 ± 21.8 a	22.07 ± 0.56 b	3.26 ± 0.14 b	6. ± 0.7 a	4.32 ± 0.54 b	0.52 ± 0.02	504.7 ± 60.9 b	1664 ± 209 b	2.43 ± 0.24 b
Scheirer Ray Hare-test	* **	* *	* *	* ** *	* *	*	* ** *	0.72	* ** *	* ** *	* **
Year (Y)											
2011	1389 ± 424 a	193.3 ± 15.4 a	179.2 ± 13.7 a	22.27 ± 0.76 b	3.23 ± 0.09 b	6.5 ± 0.4 a	4.66 ± 0.8	0.518 ± 0.01 b	525.5 ± 79.4 b	1779 ± 352	2.46 ± 0.28 b
2012	1058 ± 213 b	171.1 ± 21.4 b	160.6 ± 16.6 b	22.78 ± 0.45 a	3.35 ± 0.1 a	5. ± 0.7 b	5.04 ± 0.66	0.528 ± 0.02 a	589.4 ± 61.7 a	1899 ± 122	2.66 ± 0.13 a
Scheirer Ray Hare-test	* *	* **	* **	*	* *	* **	0.09	*	*	0.09	*
S x Y											
Up-slope x 2011	1028 ± 204 b	182.4 ± 10.6	168.2 ± 7.3	22.94 ± 0.39	3.3 ± 0.05 a	6.4 ± 0.5	5.34 ± 0.5	0.52 ± 0.01	596.0 ± 35.6	2069 ± 249 a	2.7 ± 0.14 a
Down-slope x 2011	1750 ± 220 a	204.2 ± 11.1	190.2 ± 8.8	21.6 ± 0.3	3.16 ± 0.06 b	6.5 ± 0.2	3.99 ± 0.32	0.52 ± 0.01	455.1 ± 32.6	1489 ± 113c	2.23 ± 0.14 b
Up-slope x 2012	992 ± 221 b	172.6 ± 10.9	159.9 ± 10.5	23.02 ± 0.45	3.34 ± 0.06 a	6.0 ± 0.3	5.42 ± 0.54	0.52 ± 0.01	624.5 ± 63.2	1959 ± 106 ab	2.68 ± 0.15 a
Down-slope x 2012	1124 ± 191 b	169.6 ± 28.9	161.3 ± 21.5	22.54 ± 0.3	3.36 ± 0.13 a	6.0 ± 0.9	4.65 ± 0.53	0.53 ± 0.02	554.35 ± 36.1	1838 ± 110.12 b	2.63 ± 0.12 a
	*	0.11	0.08	0.09	*	0.77	0.22	0.32	0.12	*	* *

ns,* ,**, *** ,**** Non significant or significant at P ≤ 0.05, 0.01,0.001 and 0.0001, respectively. Different letters within each column indicate significant differences according to Dunn-Bonferroni pairwise comparisons test (P = 0.05). All data are expressed as mean ± standard deviation by the berry weight and volume.

The yield was also significantly affected by the Slope × Year (S × Y) interaction. Specifically, the Down-slope vines in 2011 showed the highest yield, while the Up-slope vines in 2012 had the lowest grape yield (−43%).

These results on yield differences between the Up- and Down-slope vines could be explained, by the different hydraulic properties and, specifically by the lower water retention of the soil horizons of the upper site (Basile et al., 2020; Bonfante et al., 2017). The Up-slope soil hydrological properties led to a lower soil water content and also lower soil matric potential that imposed more intense stress in those grapevines. Moreover, the significant rainfall event at the beginning of veraison 2012 probably avoided more intense stress in post-veraison. On the other hand, the Down-slope vines in 2011 were able to produce the highest yield when there was no stress in pre-veraison and moderate post-veraison stress.

The pre-veraison stress that took place in 2012 was the main driver of yield reduction in both sites. The results on the influence that pre-veraison stress has on yield and berry growth parameters, agree with previous data reported in the literature. For example, Casassa et al. (2015) found a significant reduction of berry weight that turned into limited yields in Cabernet Sauvignon grapes subjected to pre-veraison stress, while post-veraison stress caused no yield reduction in comparison to full irrigated grapes. Wenter et al. (2018) on an experiment in hilly viticulture reported significantly lower yield in rainfed vines subjected to severe stress in pre-veraison compared to those with full or deficit irrigation. Similarly, Palai et al. (2021) reported the lowest value of yield, fresh and dry berry weight for vines subjected to pre-veraison stress.

In addition, Intrigliolo et al. (2016) found that severe post-veraison stress in rainfed Cabernet Sauvignon vines decreased berry weight and yield by 30% when compared to irrigated vines at 50% of ET_c. These results are in line with our yield results of 2011, whose differences were driven by severe post-veraison stress in Up-slope vines. Intermediate yields were generated in the Up-slope vines in 2011 and the Down-slope vines in 2012 (Table 1). Both of them were significantly lower than the Down-slope 2011, but not significantly different from the Up-slope 2012. In this way, these yield differences were significant in the Down-slope vines between the two years, while no significance was found between the Up-slope vines. In particular, the Up-slope yield of 2011 was not significantly higher than the Up-slope yield of 2012, even with different severity and timing of stress.

Furthermore, the yield results of 2012 resulted in a non-significant 12% difference between the two sites. This small yield difference arose even if the Up-slope site encountered moderate to severe levels of pre-veraison stress and the Down-slope site weak to moderate levels. At the same time, both sites experienced severe post-veraison water stress levels in 2012.

These results could be explained by the improved efficiency of water management by the upper grapevines for production purposes. The grapevines from the Up-slope under more stressed conditions could have developed internal adaptation mechanisms that enabled them to maintain a steady production. Indeed, Pagay et al. (2022) observed in an Australian Mediterranean-type climate area that rain-fed Cabernet Sauvignon showed better resilience to drought when grown in shallow soils as compared to vines in deep soils, due to drought adaptation mechanisms induced by the cyclical droughts over many years.

3.3. Berry composition

A great significant effect ($p \leq 0.0001$) of slope position (S) was detected for total soluble solids, total polyphenols, total anthocyanins, and color intensity of berries (Table 1). Specifically, the Up-slope vines were 4%, 21%, 21%, and 25% higher than the Down-slope vines, respectively for each of the above parameters.

Several studies reported that must quality improves significantly with mild to moderate water stress (Castellarin et al., 2007; Munitz et al., 2017; Ojeda et al., 2002). However, to our knowledge, there are not many studies that investigate the ranges of berry quality parameters between two different slope positions regarding the vine water stress development. In the same experimental site, Bonfante et al. (2017) found significant correlations between the CWSI and tannins, total anthocyanins, color intensity and sugar content in both sites.

Moreover, there was also a significant influence ($p \leq 0.001$) of the S in the relative composition of phenolic compounds. Grapes from the Up-slope position were 10% higher in tannins and 8.3% lower in the tannins/anthocyanin's ratio. These results are important for the "Aglianico" cultivar, as an excess of tannins and tannins/anthocyanins ratio could give astringents and tawny wines (Muccillo et al., 2014; Picariello et al., 2020).

The S x Y interaction was statistically significant for total polyphenols and tannins. In 2011, both parameters were statistically higher in the Up- than in the Down slope site, while even if these must parameters showed no significant difference in 2012, there was a clear trend indicating better quality in the Up- than in the Down-slope.

The effect of the S on the phenolic compounds, anthocyanins and tannins of the Up-slope berries could be related to an excessive exposure to light irradiation of the berries. In this way, specific key enzymes such as phenylalanine ammonia lyase, chalcone synthase, and stilbene synthase of flavonoid pathway would be more activated (Ferrer et al., 2008). Moreover, the slight shift towards higher production of anthocyanins with respect to tannins could be due to more specific activation of genes involved in this specific branch of pathway for "Aglianico" grapes.

Although grapes from the Up-slope position were in higher water stress conditions with respect to the Down-slope position, differences in titratable acidity and pH between the two sites were almost negligible (Table 1). This result is important considering future drier climate, as the upper vines seem to be able to maintain an acidic equilibrium similar to the one of the Down-slope vines which were under less water stress conditions. Regarding the year factor (Table 1), the main effect was detected for TSS and titratable acidity, probably due to the higher temperature and ET_o of 2012 than in 2011 (Fig. 1). Specifically, the titratable acidity was 7.4% higher in 2011 compared to 2012, while the TSS were 2.3% lower in 2011 than in 2012. Similar findings were reported in previous studies (Mira de Orduña, 2010; Wenter et al., 2018), that reported significantly higher TSS and lower titratable acidity in vineyards during the experimental year with the highest ET_o, temperatures, and limited soil water availability.

Despite the limitation of the only two-year dataset, our results show that Up-slope grapes could produce red wines with high levels of total phenolic compounds and tannins, good visual and gustatory scores, and thus with greater commercial value (Fanzone et al., 2012).

3.4. Correlation matrix and Principal Components Analysis

The Pearson correlation matrix (Fig. 4) allows the analysis of the relationships among the investigated variables. The yield variation was more positively correlated with berry weight and volume, g_s (Pre and Post) and A_Pre, whereas it was moderately positively correlated with Ψ_1 (Pre and Post), SMWP (Pre and Post), and low positively correlated with titratable acidity and A_Post. On the other hand, the grape yield was highly negatively correlated with quality parameters such as TSS, pH, Total anthocyanins, Total polyphenols, and Total tannins, whereas it was moderately negatively correlated with iWUE_Post and CWSI (Pre and Post), and low negatively correlated with iWUE_Pre. Total anthocyanins, Total polyphenols, Total tannins, and TSS quality parameters were more strongly correlated with water stress indicators (CWSI, Ψ_1 and SMWP) in post-veraison, as compared to pre-veraison, indicating that water deficiency in post-veraison ameliorated the must quality (Fig. 4).

Our results indicate that yield is strongly related to the

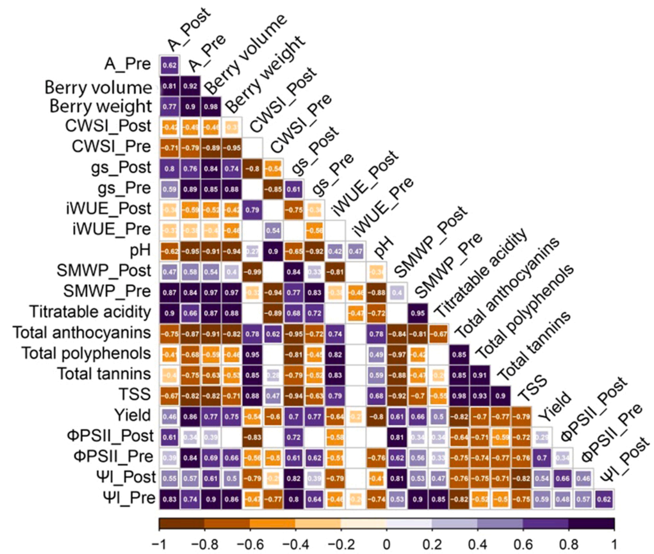

Fig. 4. **:** Correlation matrix for soil and vine physiological parameters, physical and quality grape characteristics, and yield. The white cases indicate not significant correlation at p = 0.05.

photosynthetic rate (A) in the pre-veraison stage because carbohydrate assimilation takes place for cell proliferation and expansion in this stage (Chaves et al., 2010). Whereas the plant water status and soil matric potential in both pre- and post-veraison stages strongly influence yield (Van Leeuwen et al., 2009). These outcomes have been reported in previous works, but with some differences in the relationship between these variables and the final yield (Girona et al., 2009; Intrigliolo et al., 2016). This fact may be due to the diverse variety sensitivity and/or to the application of different levels of stress (Vaz et al., 2016).

To obtain a comprehensive overview of the soil and vine physiological parameters, physical and quality grape characteristics, and yield in response to slope position (S) and years (Y), the whole data set, including the climatic parameters from May to early October of the two years, was subjected to principal component analysis (PCA). For this trial, the first two principal components (PCs) were associated with eigenvalues > 1 and explained 86.3% of the cumulative variance. PC1 accounted for 68.5% and PC2 for 17.8% (Table 2).

Fig. 5 shows the PCA results for the first two components (PC1 and PC2). In the first PC the highly positively and significantly weighted variables were berry volume, g_s_Post, A_Pre, berry weight, SMWP_Pre, Ψ_1_Pre, yield, g_s_Pre, A_Post, ϕ_{PSII}_Pre, SMWP_Post, titratable acidity, Ψ_1_Post, while the moderately weighted were ϕ_{PSII}_Post and the climatic parameters (Solar radiation, Rain and Tavg). In the PC1 Total anthocyanins, TSS, pH, Total tannins, Total polyphenols, potential CWSI_Pre were instead highly negatively weighted and ET_o only lowly. Moreover, in the second PC the potential CWSI_Post was positively and strongly weighted (Table 2). In contrast with what was expected, a positive weak correlation between titratable acidity and the average temperature was shown in the PCA (Fig. 5). However, previous studies showed that the relationship between temperature and acidic profile of grapes is cultivar-dependent; for instance, in Shiraz cv, in contrasts with Cabernet franc cv, a lack of plasticity of pH with regards to temperature was observed (Sadras et al., 2013). This result is of great interest for "Aglianico" grape, especially considering the dramatic effect that climate change can have on the base parameters of grapes, and the fundamental role of low pH and high titratable acidity for gustative equilibrium and microbial and pigment stability of "Aglianico" wine (Forino et al., 2020). Data could be, instead, considered consistent with previous studies on "Aglianico" grapes where shoot-trimming that usually determines an increase of bunch temperature was applied, but no significant effect on pH and titratable acidity of berry juice was

Table 2
: Eigen values, relative and cumulative percentage of total variance, and factor loadings for yield, physiological, and physical and grape quality traits, soil matric potential and climatic parameters with respect to the two principal components (PC1 and PC2).

Principal Components	PC1	PC2
Eigen value	14.39	3.74
Relative variance (%)	68.50	17.80
Cumulative variance (%)	68.50	86.30
Eigen vectors		
Yield	**0.827**	0.013
TSS	**-0.948**	0.314
pH	**-0.850**	-0.415
Titratable acidity	**0.753**	0.504
Berry Weight	**0.894**	0.434
Berry Volume	**0.953**	0.281
Total anthocyanins	**-0.982**	0.135
Total polyphenols	**-0.787**	0.568
Total tannins	**-0.802**	0.450
Ψ_1_Pre	**0.864**	0.212
Ψ_1_Post	**0.748**	-0.386
A_Pre	**0.924**	0.170
gs_Pre	**0.796**	0.412
ϕPSII_Pre	**0.768**	-0.094
A_Post	**0.790**	0.210
g_s_Post	**0.933**	-0.203
ϕPSII_Post	**0.584**	-0.575
SMWP_Pre	**0.883**	0.430
CWSI_Pre	**-0.727**	-0.684
SMWP_Post	**0.756**	-0.649
CWSI_Post	-0.692	**0.714**
Supplementary variables		
Rad	**0.440**	0.401
Rain	**0.440**	0.401
ET_o	**-0.440**	-0.401
Tavg	**0.440**	0.401

Boldface factor loadings indicate the most relevant characters for each principal component.

observed (Caccavello et al., 2019). It is also possible that, the lack of a great effect could be due to the higher content of exchangeable potassium in the Down-slope site compared to the upper site (Marcuzzo et al., 2021). Further and more specific experiments to study the separate effects of light and temperature, the potassium soil content and the K^+ accumulation capacity could help to better understand the behavior of this grape cultivar regarding these environmental factors.

The PC1 and PC2 score plot (Fig. 5) discriminated the variables that more influenced each slope x year treatment (S x Y). The positive side of PC1 in the lower right quadrant included most of the observations of the Down-slope site of 2011. This site was characterized by high (relative) values of soil and vine water potential, thus good water status, which was associated to the highest yield (Table 1).

The lowest yield was recorded in the Up-slope 2012 treatment (Table 1), which observations fall within the left lower quadrant, characterized by the highest CWSI_Pre and ET_o. Moreover, as the highest relative values of both SMWP_Pre and Ψ_1_Pre were observed for the opposite quadrant (right-upper), it means that the lowest values of these water status parameters (i.e., stressful conditions) occurred in Up-slope 2012. Therefore, we can infer that water stress in pre-veraison caused the lowest yield observed in the Up-slope 2012. The observations from the upper and lower left quadrant characterized the Up-slope 2011 and 2012, respectively. Both sites were identified by lower yield and high-quality parameters (Total anthocyanins, Total polyphenols, TSS and Total tannins), mainly for the Up-slope 2011, and pH for the Up-slope 2012. Interestingly, the CWSI in post-veraison was clustered within the Up-slope 2011, and tightly correlated with polyphenols, TSS, and tannins (Figs. 4 and 5). Thus, a certain level of post veraison stress improved these quality parameters (Intrigliolo et al., 2016). Conversely, the CWSI in pre-veraison, which was clustered in the Up-slope 2012, had no such effect on most quality parameters.

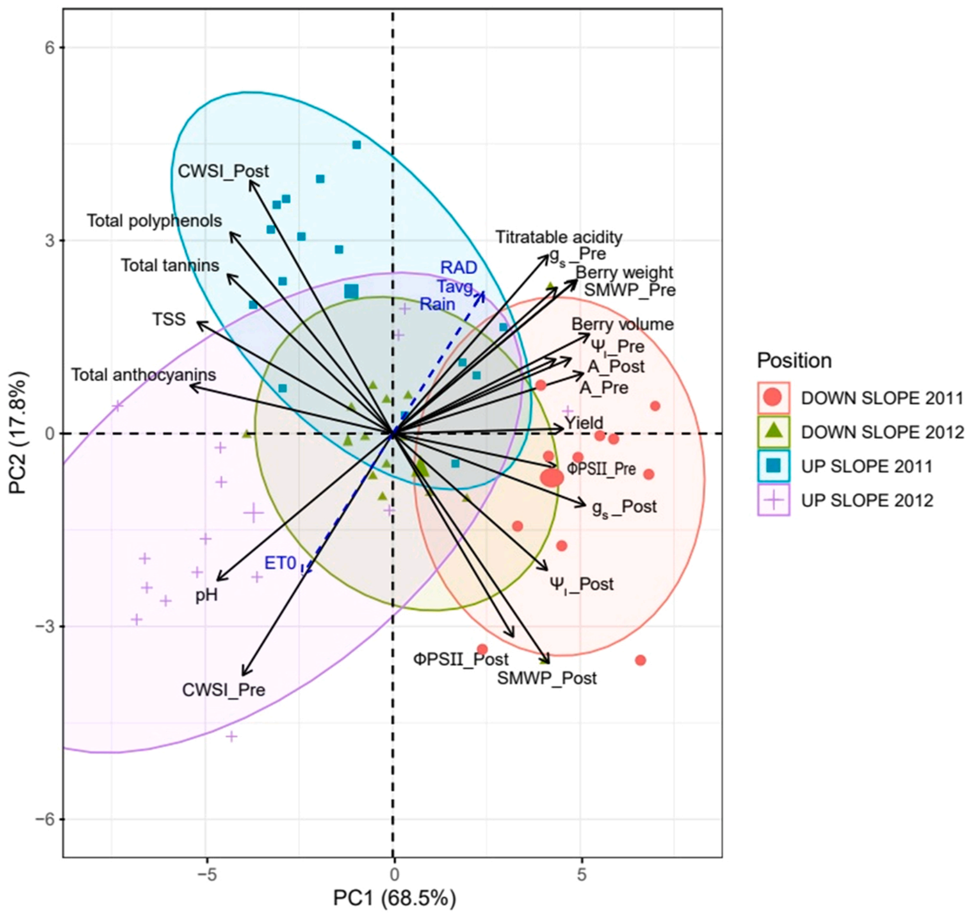

Fig. 5. : Principal component loading plots and scores of principal component analysis for physiological, yield and quality parameters, soil matric water potential and climatic variables as function of slope position and year.

Unfortunately, the Down-slope 2012 observations are somewhat in the middle and less tightly clustered than the other groups. When the observations are close to the center, some information is carried on other principal components, which means that the data set does not provide any relevant information regarding the Down-slope 2012 group.

4. Conclusions

The results achieved in this study reinforce the importance that grapevines grown in hilly soils with varying soil hydrological properties experience different water stress development. More specifically - compared to Down-slope site, vine water stress was more intense in the Up-slope site, that had lower soil water retention capacity, worse vine responses in terms of both ecophysiology and yield, but better must quality.

The pre-veraison water stress, rather than post-veraison stress, prevailed the reduction of yield and its related parameters. The must quality parameters such as total soluble solids, total polyphenols, total anthocyanins, and color intensity of berries performed better in the Up-slope vines than in the Down-slope: this effect can be attributed to water stress during post-veraison. Consequently, two different kinds of “Aglianico” wines could be produced in the two sites: high quality wine from the Up-slope grapes, and lower commercial quality wine from the Down-slope grapes.

In addition, grapevines in the upper site with more stressful conditions were able to maintain similar ranges of must acidity parameters to the ones in the down site. This fact is important for growers, as the “Aglianico” cultivar can keep its high wine quality due to its resilient behaviour considering a changing climate with more frequent drought events.

This study highlights the importance of identifying viticultural zones with different soil physical properties, vine eco-physiological, yield and quality grape characteristics. In this way, the growers could perform specific management for each zone and produce the best wine according to the specific characteristics of each viticulture zone.

CRediT authorship contribution statement

Conceptualization: RA, PG, Methodology: RA, MHS, ABas, ABon, PG, Field and lab investigation: GG, AG, Data curation: RA, MHS, GG, AG, PG, Writing—original draft preparation: RA, APS, MHS, AG, PG, Writing—review: RA, APS, MHS, ABas, ABon, AG, PG.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

Data will be made available on request.

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