



Article Water Availability Affects the Capability of Reflectance Indices to Estimate Berry Yield and Quality Attributes in Rain-Fed Vineyards

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Abstract: Remote sensing methods are known to provide estimates of berry quality. However, previous studies have shown that the Normalized Difference Vegetation Index (NDVI) failed to predict berry quality attributes in rain-fed vineyards. This study explores the association of several reflectance indices with vine biophysical characteristics and berry yield and quality attributes and their temporal stability. The study was conducted in rain-fed Chardonnay vineyards located around Masquefa (Penedès region, Catalonia, Spain) over four years. Canopy reflectance, fractional Intercepted Photosynthetic Active Radiation, predawn water potential and canopy temperature at midday were measured at veraison whereas berry yield and quality attributes were determined at harvest. Water availability and vine biophysical attributes showed large temporal stability whereas berry quality attributes was subject to the timing and extent of water deficits. The Photochemical Reflectance Index (PRI), the NDVI and the Water Index (WI) provided estimates of berry quality attributes under mild, moderate and severe water deficits, respectively. These results might have potential applications in precision viticulture activities such as selective harvesting according to grape quality attributes and the assessment of ripening.

Keywords: berry yield and quality attributes; rain-fed vineyards; reflectance indices; water availability; NDVI; WI; PRI

1. Introduction

A main current interest to viticulturists and wine industries is to identify zones of distinct grape quality for differential harvest in order to produce wines with different characteristics and properties [1–3]. In grapevines, berry quality largely depends on sugar/acid balance at harvest. Indeed, a certain amount of sugar content (i.e., Total Soluble Solids, TSS) is necessary in order to produce enough alcohol during fermentation whereas acidity (Titratable Acidity, TA) has to be reduced during ripening to a level that will be palatable. Thus, the ratio TSS/TA (maturity index, IMAD) indicates the sugar/acid balance. Both TSS and TA are commonly measured by repeated sampling during the ripening process to assess berry quality and maturity and, consequently, to determine the optimal harvest date [4,5].

Environmental conditions (i.e., climate and soil) as well as vineyard management practices influence the composition and, thus, the quality of grapes [6]. Despite the complex effects of environmental conditions on grape quality, vine water status has been long recognized as the factor most comprehensively determining berry ripening and composition [7–11]. Indeed, changes in vine water status, particularly at critical phenological stages, have a direct effect on grape composition by influencing vegetative growth, canopy microclimate and fruit growth and metabolism [7,8,10,11]. In addition, vine water status



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Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). also influences fruit composition through an indirect effect on berry size, which decreases in vines subjected to water deficits [10,12].

The extent and seasonal timing of water deficits is largely determined by site variables (i.e., soil type and topography) as well as by water supply (rainfall and irrigation) [7,13,14]. Soil type and topography are spatially variable whereas water supply varies both in space and in time, particularly in Mediterranean rain-fed vineyards where rainfall is irregular and highly unpredictable. Therefore, in rain-fed vineyards, berry quality attributes might be expected to show large variability both within a year and across years (i.e., low temporal stability). Indeed, previous studies conducted at the field level have reported low temporal stability in berry quality both under rain-fed [15] and irrigated conditions [16].

Since berry quality attributes relate to the timing and extent of water deficits—and given that water deficits vary both spatially and temporally—it is inefficient to employ conventional methods to measure berry quality. As an alternative, remote sensing offers the possibility of a rapid assessment of large vineyard areas, avoiding the need for a large number of measurements of individual samples that are cost and time consuming. Previous studies have shown the capability of remote sensing methods to estimate vine biophysical variables such as size and vigour, potential indicators of berry quality and yield while providing opportunities for cost-effective generation of spatial data amenable to precision viticulture activities [17,18]. Vegetation indices derived from the red and near infrared bands have been used to map relative differences in vine canopy vigour and to estimate differences in fruit yield and quality attributes [4,17,19]. However, studies conducted under rain-fed conditions found that vegetation indices, such as the Normalized Difference Vegetation Index (NDVI), failed to predict berry quality parameters [20–22] whereas González-Flor et al. [23] showed that the capability of NDVI to estimate TSS was subject to the timing of occurrence of water deficits (i.e., before or after veraison). Thus, characterization of canopy structure appears less useful at estimating the effects of water stress on berry composition.

Water deficits decrease the synthesis and accumulation of sugars and acids in berries not only through decreases in leaf area but also by inducing stomatal closure (which limits photosynthesis). Therefore, spectral indices capable of assessing plant water status or their effects on photosynthetic functioning might provide information potentially linked to fruit quality. Indeed, previous studies have shown the capability of the Photochemical Reflectance Index (PRI) [24,25], an indicator of the epoxidation state of xanthophyll pigments and, thus, of photosynthetic efficiency, to estimate berry quality [26] under mild to moderate water deficits. Similarly, the Water Index (WI), an indicator of plant water status [27,28], successfully estimated berry quality in grapevines experiencing water deficits [21,23]. Therefore, it appears that for in rain-fed vineyards, the timing and extent of water deficits might be critical in determining the capability of the spectral indices to assess berry yield and composition. Field spectroscopy might be useful at identifying reflectance indices to estimate berry yield and quality attributes. Indeed, in previous studies, we explored the capability of narrow-band reflectance indices at estimating berry yield and quality attributes at the canopy level while considering separately moderate to severe water deficits and mild to moderate water deficits [21,23]. This spatial scale (i.e., multifield or regional level) is of interest for wineries (industry) attempting to make decisions about harvest date at the regional scale for multiple vineyards simultaneously rather than single vineyard [22]. Herein, the capability of reflectance indices to estimate berry yield and quality attributes is further reassessed by considering single vines of different fields spreading over an area representative of the Designation of Origin (D.O.) Penedès through differing water availabilities. The main objective of this work was to examine opportunities to predict berry quality at harvest in rain-fed Chardonnay vineyards using reflectance indices of canopy structure and physiological status in order to:

- (i) determine the temporal stability of vine vigour and water status and berry yield and quality attributes
- (ii) examine the association of reflectance indices to vine vigour, water status and berry yield and quality attributes
- (iii) study the impact of the timing and severity of water deficits on the capability of reflectance indices to estimate berry yield and quality attributes at harvest.

2. Materials and Methods

2.1. Study Site

The study was conducted in ten rain-fed commercial vineyards of *Vitis vinifera* L. cv. Chardonnay plants (K5V1 clone) located in the Designation of Origins (D.O.), Penedès region (Catalonia, Spain), over four years (2007, 2008, 2009 and 2011). The location of the vineyards studied is provided in Table 1.

Table 1. Location and area of the vineyards studied. Coordinates are provided in decimal degrees for a single vine at each vineyard. Sampled vines were less than 5 m apart from each other.

Vineyard	Municipality	Area (ha)	X Coordinate (°)	Y Coordinate (°)
Batista	Masquefa	3.8	1.79620	41.49797
Hostal	Masquefa	2.3	1.78292	41.50378
Valencians	Piera	9.8	1.76192	41.52602
Les Planes	Piera	13.8	1.76615	41.47577
La Plana	Piera	3.2	1.75697	41.46499
Masover	Piera	1.4	1.74533	41.48836
Isidro	St. Llorenç d'Hortons	0.9	1.82534	41.47678
Casa	St. Llorenç d'Hortons	1.2	1.81994	41.47524
Barraca	St. Llorenç d'Hortons	0.9	1.81656	41.47701
La Creu	St. Sadurní d'Anoia	3.7	1.79590	41.42612

The region has a Mediterranean climate with an average annual temperature of 15 °C and mean annual rainfall of 550 mm. Weather data during the study period were obtained from a weather station located at Hostalets de Pierola (41°31′59″ N; 1°48′ 31″ W), which belongs to the Catalan Meteorological Network. Daily potential evapotranspiration (ET₀) was calculated by the Penman–Monteith equation using data from the same meteorological station. Weather water balance was calculated as the precipitation minus reference evapotranspiration (P – ET₀) for the entire growing cycle (from 1 November to 30 October) and at two different stages: pre-veraison (from budburst to veraison) and post-veraison (from veraison to harvest).

Vines were planted between 1990 and 2001 at variable density, ranging from 2083 to 3703 stock ha⁻¹, and the training system was Double Royat. Soil textures are loam and loamy-silt and soil depth ranges from 0.35 m to 2.0 m among vineyards. A more detailed description of the vineyards studied in terms of soil properties and plantation characteristics is provided in Serrano et al. [29] and González-Flor et al. [23]. Thirty vines (3 vines at each vineyard) with contrasting vigour were chosen to carry out measurements at the stage of veraison, when vines attained full canopy expansion. This phenological stage was chosen on the basis of previous studies aimed at estimating berry yield and composition from remote sensing data [30]. Field data collection at veraison was carried out over the third and fourth week of July, whereas harvest was carried out around the second week of August.

2.2. Vine Water Status

Predawn water potential (Ψ_p) was measured on a randomly selected mature leaf of the outer part of the canopy (1 leaf per vine) using a pressure chamber (Soilmoisture 3005, Soil Moisture Corp., Santa Barbara, CA, USA) at the stage of veraison. The canopy to air temperature difference ($T_{canopy} - T_{air}; \Delta T_m$) of field vines was determined at midday (solar noon) using a hand-held infrared thermometer (Scheduler, SN 870401 in 2007 and ST Pro Plus, Raytek Corp., Santa Cruz, CA, USA in 2008), positioned at 20 cm of the canopy and held at an angle of 60° so that the field of view was approximately 0.25 m².

2.3. Vine Development and Vigour

Fractional intercepted Photosynthetic Active Radiation (fIPAR) was determined using a ceptometer (Accupar, Decagon Devices Inc., Pullman, WA, USA). Data were gathered by placing the ceptometer at ground level. Measurements were taken at regular intervals (3 parallel and 4 perpendicular to the row) with the individual vine located in the central portion and avoiding border vines. Incident radiation readings were taken above the vines. Measurements were taken at midday (solar noon) within an interval of less than 1.5 h at the stage of veraison.

2.4. Spectral Measurements

Canopy radiance was measured over each vine using a spectroradiometer UNISPEC (PP Systems Ltd., Havervill, MA, USA) with a 2.3 mm diameter bifurcated fibre optic (model UNI410, PP Systems, Havervill, MA, USA) fitted with a 12° field of view foreoptics (UNI-710, PP Systems Ltd., Havervill, MA, USA). The detector samples 256 bands at roughly even intervals (average band-to-band spacing 3.3 nm) within a 400–1100 nm effective spectral range. The radiometer was mounted on a tripod and held in a nadir orientation at ~0.75 m above the canopy. Four scans were internally averaged for each vine. Measurements were expressed as apparent reflectance after standardizing by the irradiance determined using a cosine corrected detector lens (UNI-685, PP Systems Ltd., Havervill, MA, USA) positioned above the canopy. Data were collected on cloudless days between 11:00 h and 13:00 h (i.e., solar noon) in order to minimize disturbances from the atmosphere and changes in solar elevation.

Several hyperspectral indices related to canopy structure, water content and pigment content (i.e., chlorophyll and carotenoid) were calculated from each collected spectra. We herein report on those indices that provided significant relationships with either vine canopy structure and water status or berry quality parameters based on previous results [21,23,31]. The number of indices has been reduced in order to avoid redundancy of data. The Normalized Difference Vegetation Index (NDVI) [32] was calculated to provide canopy structure estimates (Leaf Area). In addition, The Water Index (WI) [28,33] was chosen to estimate vine water status. The Normalized Phaeophytinization Index (NPQI) [34] was determined to provide estimates of chlorophyll degradation. In addition, the carotenoid to chlorophyll ratio was assessed using the Structure Independent Pigment Index (SIPI) [35]. Finally, the Photochemical Reflectance Index (PRI) [24,25] was also determined. Indices were calculated from reflectance narrow bands as follows:

$$NDVI = (R_{900} - R_{680})/(R_{900} + R_{680})$$
$$WI = R_{900}/R_{970}$$
$$PRI = (R_{531} - R_{570})/(R_{531} + R_{570})$$
$$SIPI = (R_{800} - R_{450})/(R_{800} - R_{650})$$
$$NPQI = (R_{415} - R_{435})/(R_{415} + R_{435})$$

2.5. Berry Yield and Quality Attributes

In the years of study, harvest took place over the second and third weeks of August when berries were at optimum ripeness for cava (sparkling wine) elaboration. Experimental vines were hand harvested, and total yield per vine was weighed. Berries were then carried to the lab in coolers and pressed, and the must, after filtration, was analysed to determine quality attributes for each sample (vine). Total soluble solids (TSS, °Brix) were determined by refractometry (WM-7, ATAGO Co., Ltd., Tokyo, Japan). Total acidity (TA) was determined by titrimetry with 0.1 M NaOH to an end point of pH 8.2 using phenolphthalein indicator solution and was expressed in g tartaric acid L⁻¹. Maturity index (IMAD) was calculated as the ratio between TSS and TA. Berries for cava elaboration are considered at optimum ripeness at TSS ~ 18 °Brix and TA ~ 10 g tartaric acid L⁻¹. In addition, as a surrogate for berry size, berry weight (W_{100}) was determined on a subsample of 100 berries.

2.6. Statistical Analyses

Data were analysed using the statistical package IBM SPSS Statistics for Windows, version 25.0 (IBM Corp., Armonk, NY, USA). Differences in the variables studied were evaluated by analysis of variance (ANOVA) and means were compared using the Tukey's test. Variability (i.e., the degree of variation of a specific attribute within a year) in vine vigour, water status and berry yield and quality attributes was assessed using coefficients of variation. Temporal stability (i.e., whether a specific attribute systematically presents a similar spatial pattern over the years) was determined using the Kendall's coefficient of concordance (W) for the variables studied. Kendall's coefficient varies from 0 in case of total disagreement (i.e., no temporal stability) to 1 in case of total agreement [15]. In addition, in order to identify the association and rank the contribution of vine canopy structure and water status to berry yield and quality attributes, we carried out a series of Principal Component Analyses (PCA) where we considered several variables in conjunction: fIPAR, Ψ_{p} , berry yield and quality attributes (TSS, TA, IMAD) as well as reflectance indices (NDVI, WI, PRI SIPI, and NPQI) for the whole data set as well as for separate data sets according to the timing of occurrence (i.e., pre- or post-veraison) and severity (i.e., mild, moderate and severe) of water deficits.

3. Results

3.1. Weather Conditions

Temperatures and precipitation over the entire growing cycle—from 1 November to 30 October—(Table 2) were close to the long-term average (i.e., mean annual temperature 15 °C and annual precipitation 550 mm) in 2007 and 2008. In contrast, in 2009 and 2011, mean temperature was ca. 2 °C higher than the long term average and cumulative precipitation was above the mean annual rainfall (163 mm and 188 mm higher than the long-term average for 2009 and 2011, respectively). A detailed description of weather conditions and, particularly, of the weather water balance over the years of study can be found in Serrano et al. [21] and González-Flor et al. [23]. Briefly, in 2007 and 2009, and according to the weather water balance, the water restriction was particularly severe at pre-veraison (i.e., from bud-break to veraison) with $P - ET_0 = -275$ mm and $P - ET_0 = -216$ mm in 2007 and 2009, respectively, whereas over the ripening period (post-veraison), there was ample water availability (Table 2). In contrast, in 2008 and 2011, water constraints had a larger incidence at post-veraison with $P - ET_0 = -131$ mm and $P - ET_0 = -150$ mm in 2009 and 2011, respectively, whereas water availability at pre-veraison was abundant (Table 2). In summary, in 2007 and 2009, water availability was scarce at pre-veraison whereas water restrictions had a larger incidence at post-veraison in 2008 and 2011.

e Pierola (41°31′59″ N, 1°48′31″ W).									
T _{mean} (°C)	P (mm)	ET ₀ (mm)	P — ET ₀ (mm) Pre-Veraison	P – ET ₀ (mm) Post-Veraison					
15.0	510.8	851.4	-274.5	-41.1					
14.6	533.1	767.6	-97.0	-130.6					
17.5	687.6	928.2	-216.4	-88.7					
	[°] 31′59″ N, 1°48′ T _{mean} (°C) 15.0 14.6 17.5	[°] 31′59″ N, 1°48′31″ W). T _{mean} (°C) P (mm) 15.0 510.8 14.6 533.1 17.5 687.6	[°] 31′59″ N, 1°48′31″ W). T _{mean} (°C) P (mm) ET ₀ (mm) 15.0 510.8 851.4 14.6 533.1 767.6 17.5 687.6 928.2	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$					

1031.8

-89.1

Table 2. Average temperature (T_{mean}), precipitation (P) and reference evapotranspiration (ET₀) for the entire growing cycle, and weather water balance (P – ET₀) from bud break to veraison (pre-veraison) and from veraison to harvest (post-veraison). Data are from the meteorological station of Els Hostalets de Pierola (41°31′59″ N, 1°48′31″ W).

3.2. Vine Vigor and Water Status

17.2

662.6

2011

There were significant differences (p < 0.05) in vine vigour and vine water status at veraison among years. Vine vigour, as indicated by fIPAR values, significantly differed among years ranging from 0.46 in 2007 to 0.70 in 2009 (Table 3). In addition, Ψ_p and ΔT_m at veraison showed significant differences among years. Predawn water potential ranged from -0.88 MPa in 2007 to -0.26 MPa in 2011 with intermediate values in 2008 and 2009, whereas ΔT_m ranged from -4.62 °C to 0.92 °C (Table 3). Within a year, the coefficients of variation (CV) of fIPAR ranged from 11% in 2009 to ca. 28% in 2007. In addition, CV for Ψ_p predawn water potential ranged from \sim 24% in 2008 to \sim 41% in 2011, whereas ΔT_m showed large variation, particularly in 2007 (CV = 268%) and 2008 (CV = 110%).

Table 3. Descriptive statistics for predawn water potential (Ψ_p) , canopy to air temperature difference (ΔT_m) and fractional intercepted PAR (fIPAR): number of vines (n), minimum (Min), maximum (Max) and Mean values, standard deviation (SD) and coefficient of variation (CV).

Year	n	Min	Max	Max Mean *		CV (%)
Ψ _n (MPa)						
2007	30	-1.40	-0.30	-0.88 a	0.28	32.1
2008	27	-0.95	-0.40	$-0.67 \mathrm{b}$	0.16	24.1
2009	30	-0.73	-0.20	−0.45 c	0.12	27.6
2011	30	-0.60	-0.13	-0.26 d	0.11	40.9
ΔT_m	(°C)					
2007	30	-3.30	5.00	0.92 a	2.46	268
2008	27	-6.55	1.40	−1.96 b	2.14	110
2009	30	-7.15	-1.85	−4.62 c	1.59	34.4
2011	30	-5.50	0.30	0.30 –2.47 b		53.0
fIPA	AR					
2007	30	0.16	0.66	0.46 a	0.13	27.7
2008	27	0.30	0.80	0.61 b	0.11	18.4
2009	30	0.54	0.86	0.70 c	0.08	11.0
2011	30	0.47	0.86	0.66 bc	0.09	13.3

* Different letters indicate significant differences (p < 0.05) among years according to Tukey's test.

3.3. Berry Yield and Quality Attributes

Table 4 presents summary statistics for berry yield and quality attributes. Yield ranged from 3.4 kg vine⁻¹ in 2007 to 5.8 kg vine⁻¹ in 2011 and was significantly higher in 2011 than in the other years of study. Similarly, TSS showed significant differences among years with lower values in 2007 (17.1° Brix) than in the other years of study when TSS ranged from 18.8° Brix to 20.1° Brix. In contrast, there were no significant differences among years in TA and IMAD with an average value (calculated from annual means over the four years) of 10.2 g L⁻¹ of tartaric acid for TA and 1.92 for IMAD.

-150.0

Year	n	Min	Max	Mean *	SD	CV (%)
Yield (Kg	vine ⁻¹)					
2007	30	1.19	5.56	3.41 a	1.33	39.0
2008	27	1.50	6.93	3.77 a	1.27	33.7
2009	24	1.92	7.28	4.22 a	1.49	35.3
2011	29	1.25	11.51	5.82 b	2.25	38.6
TSS (Brix)					
2007	30	11.37	20.84	17.13 a	2.74	15.6
2008	27	16.40	22.03	19.32 b	1.58	8.2
2009	24	15.10	23.20	20.10 b	2.20	10.9
2011	29	15.50	21.80	18.78 b	1.55	8.3
TA (g tartari	c acid L^{-1})					
2007	30	6.30	14.40	9.73 a	1.85	19.0
2008	27	7.22	16.00	10.49 a	2.35	22.4
2009	24	7.39	13.13	10.02 a	1.66	16.6
2011	29	8.60	13.79	10.67 a	1.52	14.3
IMAD (1	FSS/TA)					
2007	30	1.00	3.31	1.85 a	0.59	31.9
2008	27	1.03	2.87	1.94 a	0.51	26.2
2009	24	1.37	2.80	2.06 a	0.42	20.4
2011	29	1.34	2.54	1.81 a	0.34	18.8

Table 4. Minimum (Min), maximum (Max) and mean (Mean) values, standard deviation (SD) and coefficient of variation (CV) observed on Yield, Total Soluble Solids (TSS), Total acidity (TA) and maturity index (IMAD) for the years of the study.

* Different letters indicate significant differences (p < 0.05) among years according to Tukey's test.

Yield showed a large variation with CV ranging from 33.7% to 39.0%, whereas quality attributes showed lesser variation. Indeed, coefficients of variation ranged from 8.2 in 2008 to 15.6 in 2007 for TSS, whereas TA and IMAD showed larger variation with CV ranging from 14.3% to 22.4% for TA and from 18.8% to 31.9% for IMAD.

3.4. Temporal Stability

Over the entire period of the study, the temporal stability in vine vigour and water status as well as in berry yield and quality attributes was assessed by the Kendall's coefficient of concordance (W) (Table 5). The W evaluates to which extent a variable might be considered as time stable. Variables linked to vine water status presented high W values with Ψ_p (W = 0.90), ΔT_m (W = 0.69) and W_{100} (W = 0.62), whereas variables linked to canopy vigour showed intermediate values with W = 0.53 for fIPAR and W = 0.422 for NDVI. Yield and TSS showed a lesser degree of concordance over the years of study (W = 0.22 and W = 0.19 for yield and TSS, respectively), whereas TA and IMAD did not show any temporal stability (i.e., W ~ 0).

Table 5. Kendall's coefficient of concordance (W) computed over the years of study. Variables are predawn water potential (Ψ_p), canopy to air temperature difference (ΔT_m), fractional intercepted PAR (fIPAR), Normalized Difference Vegetation Index (NDVI), Yield, weight of 100 berries (W_{100}), Total Soluble Solid (TSS), Total acidity (TA) and maturity index (IMAD).

Variable	W	x ²	Significance (p Value)
Ψ _p	0.899	72.823	0.001
ΔT_{m}	0.686	55.572	0.001
fIPAR	0.530	42.911	0.001
NDVI	0.422	29.087	0.001
Yield	0.223	13.380	0.004
W ₁₀₀ *	0.616	33.267	0.001
TSS	0.188	11.303	0.010
TA	0.055	3.300	0.348
IMAD	0.019	1.000	0.801

* W₁₀₀ was not measured in 2008.

3.5. PCA Analysis

Principal Component Analysis was carried out to examine the association among the reflectance indices and vine attributes (vine vigour and water status, and berry yield and quality parameters). For the sake of clarity, we herein report the principal component (PC) results that accounted for at least 10% of variance and for those variables with large factor loadings (i.e., absolute coefficient's magnitude > 0.4).

When the overall data set was considered (Table 6), Component 1 (PC1) showed a strong association between NDVI and PRI, Ψ_p and fIPAR and represented 31% of variation. In addition, PC1 was associated with WI and yield and was negatively correlated with ΔT_m . Component 2 (PC2) associated TSS and IMAD and was negatively correlated to TA and represented 21% of variation. Component 3 (PC3) showed a strong association between NPQI and PRI accounting for 14% of variation. Overall, these three axis explained most of the variance in the data, accounting for 66% of the total variability.

Table 6. Eigenvectors for measured vine vigour (fractional intercepted PAR, fIPAR), water status variables (predawn water potential, Ψ_p ; and canopy to air temperature difference, ΔT_m), berry yield and quality attributes (Total Soluble Solids, TSS; total titratable acidity, TA; and maturation index, IMAD) and reflectance indices (Normalized Difference Vegetation Index, NDVI; Water Index, WI; Photochemical Reflectance Index, PRI; Structural Independent Pigment Index, SIPI; and Normalized Phaeophytinization Index, NPQI). Principal Component Analysis results are shown for the whole data set (4 years, n = 101) and for separate sets considering the timing of occurrence of water deficits (pre- and post-veraison, n = 51 and n = 50, respectively). Bold characters indicate variables with large factor loadings. Vine biophysical attributes are highlighted in green, berry yield and quality attributes in red and reflectance indices in blue.

*7 • 11		4	Years]	Pre-Veraison	ı		Post-Veraison			
variables	PC1	PC2	PC3	PC4	PC1	PC2	PC3	PC1	PC2	PC3	PC4	
Ψp	0.752	0.393	-0.040	0.310	0.764	0.471	-0.036	0.486	0.648	0.294	0.291	
$\Delta \hat{T}_m$	-0.748	-0.087	0.447	0.014	-0.854	-0.109	0.247	-0.553	-0.009	0.347	-0.535	
fIPAR	0.662	0.207	-0.236	0.184	0.835	0.083	-0.212	0.030	0.324	0.473	0.008	
Yield	0.710	-0.296	0.249	0.254	0.619	-0.429	0.004	0.791	0.325	0.125	-0.098	
TSS	0.079	0.794	-0.295	0.085	0.274	0.817	-0.195	-0.726	0.275	0.018	0.265	
TA	0.485	-0.702	-0.112	-0.125	0.487	-0.697	-0.142	0.722	-0.467	-0.179	-0.236	
IMAD	-0.364	0.896	-0.070	0.144	-0.236	0.952	-0.018	-0.828	0.413	0.107	0.317	
NVDI	0.582	0.162	-0.312	-0.452	0.810	0.053	0.136	0.083	0.196	-0.654	0.363	
WI	0.645	-0.178	-0.061	0.321	0.524	0.017	0.147	0.803	0.243	0.078	0.322	
PRI	0.552	0.372	0.580	-0.304	0.466	0.145	0.751	-0.005	0.766	-0.479	-0.300	
SIPI	-0.397	-0.271	-0.409	0.670	-0.470	0.012	-0.696	0.121	-0.460	0.682	0.331	
NPQI	0.169	0.136	0.797	0.422	-0.579	0.052	0.577	0.194	0.723	0.381	-0.340	
Eigenvalue	3.687	2.506	1.662	1.266	4.467	2.511	1.589	3.608	2.513	1.764	1.159	
Variance (%)	30.7	20.9	13.9	10.5	37.2	20.9	13.2	30.1	20.9	14.7	9.7	

Vines experiencing severe water deficits mostly showed negative factor scores for PC1 whereas those experiencing moderate and mild water deficits had positive factor scores (Figure 1a). In addition, PC3 separated vines that experienced mild or moderate water deficits according to the timing of occurrence of water deficits. Thus, in vines experiencing mild water deficits at post-veraison, PC3 had positive factor scores whereas in vines experiencing moderate water deficits at pre-veraison showed negative factor scores (Figure 1b).



Figure 1. Principal component (PC) scores for the overall data set of (a) PC1 and PC2 and (b) PC1 and PC3. Each symbol corresponds to a single vine under severe pre-veraison water deficits (orange symbols, n = 29), moderate pre-veraison water deficits (red symbols, n = 26), moderate post veraison water deficits (green symbols, n = 22) and mild post veraison water deficits (blue symbols, n = 24).

When timing of occurrence of water deficits was considered, the PCA extracted three (71% of variance) and four (80% of variance) principal components for pre- and post-veraison water deficits, respectively. Under pre-veraison water deficits, PC1 accounted for 37% of variance and was associated with fIPAR, NDVI, ΔT_m and Ψ_p , whereas PC2 accounted for 21% of variance and was mainly associated with TSS and IMAD with positive factor loadings (Table 6). In addition, PC2 was associated with yield and TA with negative factor loadings. Under post-veraison water deficits, PC1 was associated with berry quality attributes (TSS, TA, and IMAD) as well as WI and yield (30% of variance) and PC2 accounted for 20% of variance and was mainly associated with PRI, NPQI and Ψ_p (Table 6).

When the data set was partitioned according to the severity of water deficits (i.e., mild, moderate and severe), the number of extracted PC was variable. Five, four and three PC were extracted for mild, moderate and severe water deficits, respectively (Table 7). Under mild water deficits, PC1 was primarily associated to berry yield and quality attributes (TSS, TA and IMAD) and accounted for 27% of variance. The second principal component (PC2) was related to vield, PRI, SIPI and NDVI (21% of variance), whereas the third component was associated to fIPAR, WI and NDVI (14% of variance). The fourth principal component (PC4) was related to Ψ_p , ΔT_m , WI and fIPAR (10% of variance). Under moderate water deficits, PC1 was related to yield, berry quality attributes (TSS, TA and IMAD) and NDVI, and accounted for 31% of variance. The second principal component (PC2) was related to WI, PRI and NPQI and to TSS (19% of variance), whereas the third component was associated to SIPI, PRI and ΔT_m (16% of variance). The fourth principal component (PC4) was related to Ψ_p and ΔT_m (11% of variance). Under severe water deficits, PC1 accounted for 32% of variance and was related to ΔT_m , WI, yield, NDVI, TA and IMAD, whereas PC2 (27% of explained variance) was mainly associated with PRI, SIPI, NDVI, Ψ_p and TSS. The third principal component (PC3) was mainly related to TSS and SIPI (11% of the accounted variance).

Table 7. Eigenvectors for vine vigour (fractional intercepted PAR, fIPAR), water status variables (predawn water potential, Ψ_p ; and canopy to air temperature difference, ΔT_m), berry yield and quality attributes (Total Soluble Solids, TSS; total titratable acidity, TA; and maturation index, IMAD) and reflectance indices (Normalized Difference Vegetation Index, NDVI; Water Index, WI; Photochemical Reflectance Index, PRI; Structural Independent Pigment Index, SIPI; and Normalized Phaeophytinization Index, NPQI). Principal Component Analysis results are shown for separate sets considering the intensity of water deficits: mild ($\Psi_p > -0.4$ MPa), moderate (-0.4 MPa < $\Psi_p < -0.8$ MPa) and severe ($\Psi_p < -0.8$ MPa). Sample size is n = 24, n = 22 and n = 55 for mild, moderate and severe water deficits, respectively. Bold characters indicate variables with large factor loadings. Vine biophysical attributes are highlighted in green, berry yield and quality attributes in red and reflectance indices in blue.

37 . 11	Mild						Moderate				Severe		
variables	PC1	PC2	PC3	PC4	PC5	PC1	PC2	PC3	PC4	PC1	PC2	PC3	
Ψ _p	0.345	0.327	-0.243	-0.476	0.428	0.445	-0.292	-0.245	0.569	0.410	0.647	0.400	
$\Delta \hat{T}_m$	0.267	-0.355	-0.070	0.625	-0.233	0.092	0.244	0.469	-0.688	-0.733	-0.222	-0.333	
fIPAR	-0.136	0.021	0.671	0.460	0.346	-0.653	0.107	0.409	0.451	0.375	0.494	0.200	
Yield	-0.530	0.709	0.077	0.057	0.101	-0.599	0.102	-0.441	-0.248	0.735	-0.116	0.091	
TSS	0.875	-0.086	-0.181	-0.151	-0.013	0.656	-0.445	-0.055	-0.283	-0.300	0.648	0.500	
TA	-0.759	0.213	-0.460	0.064	0.052	-0.757	-0.198	-0.251	-0.119	0.792	-0.325	-0.087	
IMAD	0.914	-0.187	0.260	-0.128	-0.032	0.945	-0.024	0.197	-0.038	-0.743	0.515	0.347	
NVDI	0.193	0.519	0.525	0.017	-0.341	-0.668	0.408	0.276	0.070	0.632	0.566	-0.088	
WI	-0.283	0.271	0.588	-0.484	-0.307	0.210	0.787	0.382	0.014	0.717	-0.385	0.165	
PRI	0.424	0.801	-0.171	0.171	-0.011	0.181	0.701	-0.548	-0.083	0.245	0.756	-0.435	
SIPI	-0.262	-0.602	0.386	-0.187	0.493	-0.120	-0.402	0.794	0.056	-0.318	-0.603	0.485	
NPQI	0.456	0.542	0.099	0.277	0.454	0.504	0.665	0.054	0.304	-0.295	0.513	-0.423	
Eigenvalue	3.239	2.465	1.642	1.250	1.031	3.680	2.286	1.902	1.265	3.818	3.183	1.336	
Variance (%)	27.0	20.5	13.7	10.4	8.6	30.7	19.1	15.9	10.5	31.8	26.5	11.1	

4. Discussion

4.1. Water Status, Vine Vigor and Berry Yield and Quality Attributes

Contrasted patterns of precipitation resulted in differences in the timing and extent of water deficits at veraison among years. Indeed, Ψ_p indicated severe water deficits at veraison in 2007 and 2008, whereas in 2009 and 2011, water deficits were moderate and mild, respectively [36,37]. Variation in the timing and extent of water deficits at veraison among years was translated into differences in vine vigour (i.e., fIPAR) and water status (i.e., ΔT_m) as previously reported in these vineyards [23,29]. However, over the years of study, differences in vine vigour and water status were not translated into significant differences in yield across years, except in 2011 (mild water deficits at post-veraison) when yield was significantly higher, which might be attributed to the increase in berry size resulting from ample water availability [12,23]. Similarly, variation in vine vigour and water status at veraison were not translated into significant differences in berry quality attributes across years (i.e., TSS, TA and IMAD), with the exception of TSS, which was lower in 2007 than in the other years of study.

4.2. Variability in Water Status, Vine Vigor and Berry Yield and Quality Attributes (within Year Analysis)

Vine vigour and water status showed large variability within a year with higher coefficient of variation for ΔT_m , followed by Ψ_p and fIPAR, respectively. Large variation in ΔT_m might be expected owing to the influence of environmental conditions on ΔT_m during data acquisition and to the complex interplay of water availability and canopy structure on transpiration [38]. This makes it difficult to interpret changes in ΔT_m across years and it will not be discussed from herein on. Large variation in Ψ_p (averaged CV ~ 31%) might be attributed to the spatial variability in soil water storage capacity [6]. In contrast, variation in fIPAR (averaged CV ~ 18%) was lower than for Ψ_p . This is to be expected because pruning and other crop management strategies tend to equilibrate the vegetative and reproductive growth according to the farmer's experience and criteria. In addition, berry yield showed a similar degree of variability (averaged CV 37%) than Ψ_p over the years of study which is consistent with the close association between water availability and yield [6,7,10,21]. Nonetheless, within a year, and despite the broader range of conditions encountered in this

study (multiple field survey), berry yield showed lower variation than in previous studies conducted in a single field (i.e., within field variability), both under rain-fed conditions [15] and in irrigated vineyards [39]. In the present study, variability in IMAD (CV ~ 24%) was higher than variation in TA (averaged CV ~ 18%), whereas variation in TSS was minor (CV ~ 8%), which is consistent with the fact that date of harvest was partially decided by meeting industry TSS requirements [15]. Similar results have been observed with regard to the variation in TSS and TA in previous studies conducted under rain-fed conditions [15] and irrigated vineyards [40,41]. In addition, as reported by Tisseyre et al. [15], results show that for quality attributes, CV can almost double from one year to another whereas yield presents a more stable CV over time. In summary, in spite of the seasonal/climatic variation, large intra-annual variation in berry yield and quality attributes suggests that precision viticulture techniques may be utilized in this region to identify zones that relate to differing quality levels.

4.3. Temporal Stability in Water Status, Vine Vigor and Berry Yield and Quality Attributes (Kendall's Analysis)

The existence of recurrent patterns over time (i.e., temporal stability) in the parameters studied was assessed using the Kendall's coefficient of concordance (W). In agreement with previous studies [42], Ψ_p presented the highest Kendall's value, which is consistent with the fact that soil properties, particularly those related to water storage capacity and topography (water flow), are highly time stable. Accordingly, ΔT_m and vine vigour (fIPAR), which are largely affected by water availability, were also time stable. Previous studies have reported large temporal stability in vine vigour and yield in irrigated vineyards [43] as well as in rain-fed vineyards either within a single field [15] or across multiple fields over a large area-region [44]. In contrast, in the present study, yield was moderately time stable as previously reported [45]. In addition, in the present study, TA and IMAD did not present any temporal stability whereas TSS had a low—although significant—degree of temporal stability, which, as previously mentioned, might be partly because the harvest date was based on TSS measurements. The results obtained are in agreement with previous studies conducted in irrigated [41] and rain-fed vineyards [15] that reported no temporal stability in berry quality attributes. This is consistent with the fact that berry quality results from a strong interaction between the climate of the year and water availability [6,46]. In summary, in the present study, biophysical canopy attributes (i.e., Ψ_p , ΔT_m and fIPAR) showed high temporal stability followed by yield and TSS with moderate although significant temporal stability and parameters with no temporal stability (TA and IMAD). These results highlight the need to developing indicators of berry quality in these rain-fed vineyards.

4.4. PCA Analysis

The PCA allowed for identifying the association among reflectance indices, canopy biophysical variables and berry yield and quality attributes. For the overall data set, the first component of the PCA evidenced a strong dependence of vine vigour and yield on water availability (Ψ_p), which is consistent with the fact that water is the major limiting factor of yield in Mediterranean rain-fed vineyards [7,46]. Therefore, PC1 might be related to variation in yield driven by changes in water availability either through changes in the photosynthetic capacity [21] or in berry size [12]. Moreover, NDVI, PRI and WI were also related with PC1, evidencing that these reflectance indices were able to capture variation in canopy biophysical attributes associated to yield. However, berry quality attributesgrouped in principal component 2-did not show any association with vine vigour and water status, neither with reflectance indices of canopy structure and function. Thus, it appears that, in these rain-fed vineyards, reflectance indices failed to capture the complex interaction between water availability and climatic conditions on berry quality [6,46]. The third component highlighted important adjustments in chlorophyll content with positive factor loadings related to NPQI and PRI. Although PRI is associated to dissipation of excess radiation through the xanthophyll cycle pigments [25], it has been also related to constitutive changes in the pigment pool size (i.e., chlorophyll to carotenoid ratios) [47,48]. Indeed, the grouping of PRI and NPQI—which assesses degradation of chlorophyll to phaeophytin [49]—suggests the occurrence of constitutive changes related to environmental stresses, particularly water stress [48]. Overall, the first principal component clearly distinguished vines under severe water deficits from those experiencing mild to moderate water deficits, thus, confirming the effects of water availability on vine vigour and yield. In addition, in vines experiencing mild to moderate water deficits, the third principal component allowed for distinguishing between pre and post-veraison water deficits. Thus, vines growing under pre-veraison water deficits showed negative scores indicating changes in pigment composition (i.e., lower chlorophyll content with respect to carotenoid content) as previously reported [50], whereas vines growing under post-veraison water deficits showed positive scores.

When timing of occurrence of water deficits was considered, the PCA permitted to identify the association of both vine vigour and water status with berry yield and quality attributes. Moreover, the eigenvectors—and the variables associated to them—accounted for a large proportion of variation when compared to PC extracted from the whole data set. Thus, under pre-veraison water deficits, PC1 evidenced a close association between vine water status (Ψ_p), transpiration rates (ΔT_m), vine vigour (fIPAR), yield, and the reflectance indices NDVI, WI and PRI. The association of these reflectance indices with the canopy variables is consistent with results reported in previous studies. Indeed, the association of NDVI with vine vigour has been widely reported [17,18,20,22,30]. Moreover, the WI has been associated to transpiration rates in vines as well as in other crops [23,29,51,52], whereas the PRI has been reported to provide estimates of water status in numerous studies [52–56]. In addition, PC2 evidenced a close association between berry yield and quality attributes and Ψ_p , although in a lesser extent. However, no single reflectance index was associated with berry quality attributes in PC2. Nonetheless, NDVI (which had the highest factor score) could serve as an indirect estimator of berry quality due to its association with yield (PC1) and the association of yield with berry quality attributes (PC2) under pre-veraison water deficits. This is consistent with the fact that early water deficits (i.e., before the onset of veraison) have a detrimental effect on vegetative growth and, thus, on yield [7,10,21]. Indeed, since berry quality generally decreases along with increasing yield, NDVI has been widely used to estimate berry quality attributes [17,18,30]. In contrast, when water deficits had a major incidence at post-veraison, PC1 indicated a strong association between vine water status (Ψ_p and ΔT_m), WI and both berry yield and quality attributes whereas, in agreement with previous studies conducted in rain-fed vineyards, NDVI was not related to berry yield and quality attributes [13,21]. These results suggest that, when differences in light interception (i.e., vine vigour) are minor, stomatal aperture largely determines variation in yield [57]. In addition, improved vine water status results in larger berries and, thus, higher yield [7,10,12]. The ability of WI to estimate yield has been ascribed to the scaling of this index to leaf area [27,58] and to the capability of WI to track variation in stomatal aperture [29,51,52]. Therefore, WI might provide reliable estimates of berry yield and quality attributes under post-veraison water deficits [21,23]. The association of $\Psi_{\rm p}$ and both NPQI and PRI in PC3 indicates increases in chlorophyll content (NPQI and PRI) along with increased water availability. In addition, as indicated by the association of PRI and Ψ_p , and as mentioned above, PRI was found to be a good indicator of plant water status [52–56].

Under mild water deficits, the PC1 grouped berry yield and quality attributes whereas PC2 evidenced a close association between yield and both PRI and NDVI. Therefore, PRI might provide indirect estimates of berry quality due the close association between yield and berry quality attributes as mentioned above. Indeed, PRI is often used as a water stress indicator under mild water deficits [53,56] and has provided estimates of fruit quality in irrigated orchards [54,55]. When moderate water deficits occurred, yield and quality attributes were associated with NDVI in PC1. These results are in agreement with previous studies conducted in irrigated vineyards experiencing moderate water deficits

where vegetation indices derived from reflectance imagery in the red and near infrared bands (e.g., NDVI) have provided reliable estimates of berry quality [4,17,19,43]. On the other hand, the grouping of PRI and WI with TSS in PC2 suggests that enhanced water status led to a dilution of sugars in the berry as previously reported [7,26,59]. Finally, under severe water deficits, PC1 grouped yield, TA and IMAD with both NDVI and WI, indicating that increased vigour and water status were translated in higher yield and TA and, thus, lower IMAD [21]. The relationship between TA and both vine vigour and water status might be attributed to increased shade inside the canopy, for TA levels are typically more dependent upon temperature and radiation than on soil and vine water status [45,59]. On the other hand, PC2 grouped TSS and IMAD with NDVI and PRI which suggests that enhanced photosynthetic capacity was translated into an increase in carbohydrate supply to the berry [60]. Thus, regardless of the extent of the water deficits, NDVI was associated to yield whereas the association of NDVI with quality attributes was variable. Finally, it is worth noting that, across the series of PCA analyses, PRI and WI showed higher scores than NDVI, emphasizing the role of water availability in these rain-fed vineyards.

5. Conclusions

Predawn water potential showed large temporal stability highlighting the influence of soil water availability in these rain-fed vineyards. Accordingly, vine biophysical attributes related to water status (i.e., fIPAR, ΔT_m and W_{100}) were also time stable, evidencing the effects of water restriction on vine structure and functioning regardless of the timing and severity of water deficits. In spite of the temporal stability in vine condition, yield showed moderate temporal stability whereas berry quality attributes did not show consistency across years.

Timing and extent of water deficits were critical in determining the capability of spectral indices at assessing berry yield and composition. Indeed, when data from different years were considered in conjunction, no single reflectance index neither vine biophysical attributes were related to berry quality parameters. Consideration of the timing of incidence of water deficits showed a strong association of NDVI with yield under pre veraison water deficits and—through the dependence of berry quality on yield—with berry quality attributes, whereas, under post veraison water deficits, WI was directly associated to both yield and berry quality attributes. Finally, when the extent of water deficits was considered, berry yield and quality attributes were mostly associated to PRI under mild water deficits, to NDVI under moderate water deficits and to WI under severe water deficits. Therefore, in addition to indices related to vine structure (i.e., NDVI), reflectance indices related to plant water status (i.e., WI and PRI) might be relevant at characterizing vine conditions and, hence, berry yield and quality attributes, in these Mediterranean rain-fed vineyards. More studies are needed to validate the predictive capability of spectral indices at estimating berry quality attributes in order to ensure their applicability to management practices such as selective harvesting and the assessment of ripening.

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