Is PCD a Reliable Indicator of Berry Quality Attributes in Water Stressed Vineyards?

J.M. Pons, I. Serrano, C. Gonzales-Flor and G. Gorches
Departament d’Enginyeria Agroalimentària i Biotecnologia
Universitat Politècnica de Catalunya
Barcelona
Spain

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Abstract
Grape berry composition in terms of sugars and titratable acidity is strongly dependent on vine water status. The objective of this study was to evaluate the feasibility of using airborne derived Plant Cell Density (PCD) and canopy reflectance derived Simple Ratio (SR) measurements - both remote indicators of leaf area and intercepted light - at estimating berry yield and composition in vineyards experiencing water deficits. In addition, the capability of the Water Index (WI) - a hyperspectral reflectance based indicator of water status - derived from canopy reflectance at estimating berry yield and composition was tested. The study was conducted in vineyards of Vitis vinifera L. “Chardonnay”. Remote optical imagery was acquired at veraison and PCD derived. Field measurements of fractional intercepted Photosynthetic Active Radiation (fPAR), predawn water potential (Ψp), canopy to air temperature difference at midday (ΔTmidday) and canopy hyperspectral reflectance were also conducted at the stage of veraison. Yield, TotalSoluble Solids (TSS), TitratableAcidity (TA) and maturation index (IMAD) were determined at harvest. Predawn water potential ranged from -0.85 ± 0.06 MPa to -0.50 ± 0.07 MPa among the vineyards studied indicating moderate to severe water stress at the stage of veraison. Berry quality attributes did not show significant relationships against fPAR but were related to ΔTmidday in a variable extend. Consistently, PCD derived from airborne imagery, as well as the SR, did not show significant correlation against berry quality attributes. On the other hand, the WI derived from canopy reflectance showed significant correlation against ΔTmidday (r = -0.82, P<0.05) and successfully estimated TA and IMAD (r = 0.70, P<0.05, for both indices). The results obtained suggest that, in vineyards experiencing water deficits, remote estimates of vine water status through the WI might be more appropriate to characterize berry composition than remote estimates of vine vigor such as SR or PCD.

INTRODUCTION
Current remote sensing methods in precision viticulture use vegetation indices derived from reflectance in the red and near-infrared regions of the spectrum to estimate vine biophysical variables such as size and vigour (Hall et al., 2002; Johnson et al., 2001). Since vine vigor has a considerable effect on yield which, in turn, relates to berry composition (Chaves et al., 2010; Medrano et al., 2003), remote measurements of canopy vigor (e.g., PCD or NDVI) have been used to estimate berry yield and quality (Hall et al., 2002, 2011; Johnson et al., 2001). However, recent studies conducted in vineyards experiencing water deficits showed that while NDVI was related to vine vigor and yield, it failed to predict berry quality attributes (Acevedo-Opazo et al., 2008; Serrano et al., 2012). Reflectance indices such as the NDVI are related to canopy structure and leaf pigment concentration, and, thus, potential photosynthetic activity, but have proven less useful for monitoring photosynthetic functioning under stress conditions, particularly, water stress (Garnot et al., 1995). In vineyards, water status has been recognized to largely influence berry ripening and composition (Chaves et al., 2010; Medrano et al., 2003; Koundouras et al., 2006; Van Leeuwen and Seguin, 1994). Recent studies have
shown the capability of the WI (Peñuelas et al., 1993) derived from hyperspectral reflectance for monitoring variation in stomatal conductance in vineyards (Serrano et al., 2010). Therefore, the capability of the WI to monitor vine water status might be of great value in the context of precision viticulture as a potential tool for the estimation of grape yield and quality.

The objective of this study was to evaluate the feasibility of using remote measurements of PCD—a remote indicator of leaf area and intercepted light—derived from airborne imagery to estimate berry yield and quality attributes. In addition, the potentiality of the Water Index—a remote indicator of vine water status—at estimating berry yield and quality in vineyards experiencing water deficits is assessed.

**MATERIALS AND METHODS**

The study was carried out in nine vineyards of *Vitis vinifera* L. ‘Chardonnay’ plants (KSVI clone) located in the Cava Designation of Origin region (Catalonia, Spain, 41° 48’22”W; 1° 28’54”N) in 2008. The region has a Mediterranean climate with an average annual temperature of 15°C and mean annual rainfall of 550 mm. A more detailed description of the vineyards studied in terms of soil properties and plantation characteristics is provided in Serrano et al. (2010, 2012). In each vineyard, 3 vines with contrasting vigor were chosen to carry out measurements at the stage of veraison. Subsequently, at harvest, yield and quality attributes were determined on the same vines.

Predawn water potential (Ψ_P) was measured on a randomly selected mature leaf using a pressure chamber (Soilmoisture 3005, Soil Moisture Corp., Santa Barbara CA, USA). In addition, the canopy minus air temperature (ΔT_ambient) was measured at midday (solar noon) using a hand-held infrared thermometer (ST Pro Plus, Raytek Corp., Santa Cruz CA, USA in 2008). Transmitted PAR (PAR_1) was measured at regular intervals by placing a radiometer (Accupar, Decagon Devices Inc., Pullman, WA, USA) at ground level while incident radiation (PAR_0) was measured above the vines. Subsequently, Fractional intercepted Photosynthetic Active Radiation (fPAR) was calculated as (fPAR = PAR_1 / PAR_0).

Canopy radiance was measured over each vine using a spectroradiometer UNISPEC (PP Systems Ltd., Havervill, MA, USA) with a 12° field of view foreoptics (UNI-710, PP Systems Ltd., Havervill MA, USA) mounted on a tripod and held in a nadir orientation at -0.75 m above the canopy and expressed as apparent reflectance after standardizing by the irradiance determined using a cosine corrected detector lens (UNI-885, PP Systems Ltd., Havervill MA, USA) positioned above the canopy. The Simple Ratio (SR) and the Normalized Difference Vegetation Index (NDVI) were derived to provide vine vigor estimates while the Water Index (WI) was chosen to estimate vine water status. Hyperspectral indices were derived from each collected spectra using narrow band apparent reflectance values as follows:

\[
\text{SR} = \frac{R_{590}}{R_{660}}
\]

\[
\text{NDVI} = \frac{(R_{560} - R_{660})}{(R_{560} + R_{660})}
\]

\[
\text{WI} = \frac{R_{590}}{R_{670}}
\]

where R indicates apparent reflectance and the sub indices the respective wavelengths in nanometers. Digital multispectral imagery was acquired in July 2008 at 0.50 m spatial resolution using narrow band-pass interference filters (20 nm) and PCD derived as the ratio between near infrared (780 nm) and red reflectance (675 nm). Georeferencing of the PCD individual images to historical ortho-rectified aerial photography (www.ice.cat) was performed by means of a second-order polynomial warping and nearest neighbor resampling using GVSIG 1.11 (GVSIG Association, Valencia, Spain) and ARCGIS 9 (LSR Inc., California, USA). Within each vineyard, the vines under study were spatially located using a differential GPS (Leica Viva GS15, Leica Geosystems AG, Heerbrugg, Switzerland). At each vine, PCD was determined by choosing the highest value within a 3 x 3 neighborhood pixel window.
Harvest took place when berries were at optimum ripeness (i.e., at TSS-18 °Brix and TA-10 g tartaric acid L⁻¹) for cava (sparkling wine) elaboration. Yield was measured and, afterwards, berries were carried to the lab in coolers and pressed. The must, after filtration, was analyzed to determine total soluble solids (TSS, °Brix) by refractometry (WM-7, ATAGO Co. Ltd., Tokyo, Japan) and titratable acidity (TA, g tartaric acid L⁻¹) by titration with 0.1 M NaOH. Maturity index (IMAD) was calculated as the ratio between TSS and TA.

Data were analyzed using the statistical package SPSS 20.0 (SPSS Inc., Chicago, ILL, USA). Correlation and regression analyses were used to study the relationships among grape vigor and water status parameters, yield and quality attributes and spectral indices. Differences among vineyards in the variables studied were evaluated by analysis of variance (ANOVA) and means were compared using the Tukey's test.

RESULTS AND DISCUSSION

Temperatures from budbreak to harvest were close to the long-term average while cumulative precipitation was 347 mm. The weather water balance indicated moderate water deficits over the growing season (P - ET₀ = 235 mm) with larger incidence from veraison to harvest (P - ET₀ = 131 mm) (see Serrano et al., 2012 for more details).

Water status parameters (i.e., Ψₑd and ΔT_midday) showed significant (P<0.05) variation among vineyards. Thus, Ψₑ ranged from -0.85 ± 0.06 MPa to -0.50 ± 0.07 MPa, indicating moderate to severe water deficits at the stage of veraison (Carbonneau, 1998), while ΔT_midday ranged from -5.0 + 0.75°C to 0.42 - 0.34°C. Contrastingly, no significant differences in iPAR emerged among vineyards, which might be attributed to the minor incidence of water deficits over the vegetative growth period (i.e., from budbreak to veraison).

The effects of water deficits on vine vigor and yield have been long documented (Chaves et al., 2010; Schultz and Matthews, 1988; Van Leeuwen and Seguin, 1994). In contrast with previous studies, yield was found to decrease along with increasing iPAR (Table 1), which might be attributed to a competition effect between vegetative and reproductive growth. In agreement with previous studies (Chaves et al., 2010; Medrano et al., 2003), higher yield was accompanied by a decrease in quality attributes as indicated by the correlation between yield and both TA and IMAD (r = 0.75 and r = -0.78, P<0.05, for TA and IMAD, respectively). In addition, a marginal relationship between yield and TSS was observed (r = -0.57, P<0.10). These results are consistent with previous reports under rain fed conditions with decreased TSS and titratable acidity at harvest when water deficit progresses from moderate to severe (Koundouras et al., 2006; Van Leeuwen et al., 2009). In addition, berry quality attributes were not significantly related to iPAR but to vine water status as indicated by the significant correlation against AT_midday (Table 1), suggesting that vine water status was an important determinant of variation in grape composition.

Spectral indices were variably related to vine biophysical properties (Table 2). Thus, higher PCD was associated to lower ΔT_midday, although marginally, suggesting that more vigorous plants had higher transpiration rates. Contrastingly, and despite the close correlation between PCD and both SR and NDVI (Fig 1), SR and NDVI were not related to ΔT_midday but were significantly related to Ψₑd (R² = 0.48, P<0.05; Table 2), suggesting that water availability largely determined vine vigor, in agreement with previous studies conducted in rain fed vineyards (Acevedo-Opazo et al., 2008). In addition, none of the spectral indices showed significant correlation against iPAR, which might be attributed to the narrow range of variation in iPAR encountered in our study (see above).

In contrast with previous studies (Acevedo-Opazo et al., 2008; Hall et al., 2002, 2011), yield was poorly related to NDVI but related to W₁ (r = 0.54, P<0.01 and r = 0.75, P<0.05, for NDVI and W₁, respectively) (Fig. 2). Water deficit decreases yield due to reduced leaf area and intercepted light (Pellegrino et al., 2005) but also due to a decrease in the production of assimilates as a result of stomatal closure (Flexas et al., 2002). Thus, because the W₁ has been shown to respond to the water content of vegetation (i.e., leaf
area and relative water content) as well as to changes in vine stomatal aperture (Serrano et al., 2000, 2010), WI might better characterize the effects of water deficits on berry yield than remote estimates of intercepted light (i.e. SR or NDVI). Moreover, in our study, berry quality attributes were found to be dependent on vine water status rather than on vine vigor (Table 1). Consistently, WI was significantly related to TA and IMAD (r = -0.70, P<0.05, for both TA and IMAD) and marginally related to TSS (r = -0.59, P<0.10) (Fig. 3) while no significant relationships emerged between NDVI and berry quality attributes (results not shown). In contrast, PCD did not show significant relationships against berry quality attributes (Fig. 3), probably due to the low dependence of berry composition on vine vigor (see Table 1). The results obtained suggest that remote estimates of vine water status through the WI might be more appropriate to characterize berry quality attributes than remote estimates of canopy structure (e.g., PCD) in vineyards experiencing water deficits.

Our results add on a growing body of studies in precision viticulture that attempt to assess vine yield and quality attributes using hyperspectral indices of physiological condition (Monzón et al., 2007; Mezzio et al., 2010; Suarez et al., 2010; Serrano et al., 2012; Zarco-Tejada et al., 2005). More studies are needed, particularly at the field scale (i.e., from airborne platforms), and under a wide range of conditions and cultivars, in order to ensure the applicability of the Water Index to management practices.

CONCLUSIONS
In our study, berry yield and quality attributes were found to be significantly dependent on vine water status rather than on canopy vigor. Accordingly, and despite the dependence of vine canopy vigor on water availability, PCD and NDVI failed to predict berry yield and quality attributes while the Water Index, an indicator of stomatal aperture, provided reliable estimates of titratable acidity and maturity index in the vineyards under study. Our results suggest that remote estimates of vine water status through the WI might provide a valuable tool to predict berry composition in vineyards experiencing water deficits.

ACKNOWLEDGEMENTS
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Literature Cited


Table 1. Pearson correlation coefficients between yield and quality attributes (TSS: Total Soluble Solids, TA: Titratable Acidity, and IMAD: TSS/TA) and vine vigour and water status (fIPAR: fractional Intercepted PAR; ΔT<sub>midday</sub>: canopy minus air temperature at midday) (n=9).

<table>
<thead>
<tr>
<th></th>
<th>Yield</th>
<th>TSS</th>
<th>TA</th>
<th>IMAD</th>
</tr>
</thead>
<tbody>
<tr>
<td>fIPAR (%)</td>
<td>-0.62*</td>
<td>0.39</td>
<td>-0.47</td>
<td>0.51</td>
</tr>
<tr>
<td>ΔT&lt;sub&gt;midday&lt;/sub&gt; (°C)</td>
<td>-0.49</td>
<td>0.66**</td>
<td>-0.61*</td>
<td>0.66**</td>
</tr>
</tbody>
</table>

* and ** indicate significant correlations at P<0.10 and P<0.05, respectively.

Table 2. Pearson correlation coefficients between spectral indices and vine vigour (fIPAR) and water status (Ψ<sub>pd</sub> and ΔT<sub>midday</sub>) at veraison (n=9).

<table>
<thead>
<tr>
<th></th>
<th>fIPAR (%)</th>
<th>Ψ&lt;sub&gt;pd&lt;/sub&gt; (MPa)</th>
<th>ΔT&lt;sub&gt;midday&lt;/sub&gt; (°C)</th>
</tr>
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<tbody>
<tr>
<td>PCD</td>
<td>0.14</td>
<td>0.49</td>
<td>-0.72*</td>
</tr>
<tr>
<td>NDVI</td>
<td>-0.28</td>
<td>0.72**</td>
<td>-0.40</td>
</tr>
<tr>
<td>SR</td>
<td>-0.25</td>
<td>0.74**</td>
<td>-0.50</td>
</tr>
<tr>
<td>WI</td>
<td>-0.21</td>
<td>0.30</td>
<td>-0.88**</td>
</tr>
</tbody>
</table>

* and ** indicate significant correlations at P<0.10 and P<0.05, respectively.
Fig. 1. Relationships between Plant Cell Density (PCD) derived from aerial imagery and the Normalized Difference Vegetation Index (NDVI) and the Simple Ratio (SR) measured at the canopy level. Values are mean ± standard error of the mean of 3 vines measured at each vineyard (n=7).

Fig. 2. Relationships between berry yield and the spectral indices Normalized Difference Vegetation Index (NDVI) and Water Index (WI). Values are mean ± standard error of 3 vines measured at each vineyard (n=9).
Fig. 3. Relationships between spectral indices (PCD: Plant Cell Density, and WI: Water Index) and berry quality attributes (TSS: Total Soluble Solids, TA: Titratable Acidity, and IMAD: Maturity Index). Values are mean ± standard error of the mean determined on 3 vines at each vineyard (n=7 and n=9, for PCD and WI, respectively).