The effects of variable nitrogen and water supply on grapevine canopy reflectance

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Abstract

Optical remote sensing has been used in precision viticulture to study the spatial variability of grapevine performance because vine vegetative growth, yield, and berry composition relate strongly to properties of grapevine canopy. Most of the research on optical sensing in vineyards has been conducted with remote sensors over large areas, but limited data exist with proximal sensors under controlled N and water conditions. The aim of the present study was to evaluate the potential of vegetation indices, acquired by proximal sensing, in predicting vine productivity and berry composition responses in a factorial experiment of N and water supply over 2 years. Three N [0 (N_0), 60 (N_{60}) and 120 (N_{120}) kg/ha] and two irrigation [irrigated (I): 70% of crop evapotranspiration and non-irrigated (NI)] treatments were triplicated in randomized blocks in two vineyards located in Northern Greece, planted with cvs. Cabernet-Sauvignon and Xinomavro (Vitis vinifera L.). Vegetation indices were obtained with two active canopy reflectance sensors with different operational properties: Crop Circle Acs-210 (reflectance recording at 590 nm and 880 nm) and Crop Circle Acs-430 (630, 730 and 780 nm). The Acs-430 sensor performed better compared to the Acs-210 sensor. NDVI measurements were efficient in predicting yield in both varieties. Regarding berry composition, all sensors presented limited efficacy in predicting sugar and acid content of the juice while NDVI of the Acs-430 sensor was inversely correlated with total phenols suggesting a negative effect of dense canopies on berry color. These findings suggest that the use of active canopy sensing in

vineyards has potential limitations.

Key words: Precision viticulture, canopy reflectance, active sensors, anthocyanins, phenols

Introduction

Precision viticulture is a decision making and management support system that aims at a more efficient control of the production process in vineyards, in order to optimize the use of vineyard resources while high grape quality and low costs of production being achieved (Bramley 2010). To meet these objectives, monitoring of the spatial variability of physical, chemical and biological variables related to grapevine performance is essential in the context of precision viticulture (Hall et al. 2002). This information on spatial variability of grapevine performance can be acquired through the adoption of optical remote sensing technologies. A relatively new technology in that field is the technology of active canopy sensors that operate at a close distance from the plant's canopy (Barker et al. 2010). Active sensors use their own source of light and advanced electronics and optics to differentiate between natural light and modulated light, which reduces the effect of shadows and minimizes background soil interferences (Stamatiadis et al. 2010).

Multiple-wave band data representing canopy reflectance are easily transformed into a single numerical value (vegetative index) that relates canopy characteristics to reflectance (Hatfield et al. 2008). The normalized difference vegetation index (NDVI) is the most commonly used vegetation index that quantifies the difference between the canopy reflectance in the near infrared and visible bands (Rouse, Jr. et al. 1973). In grapevines, NDVI can detect changes in canopy size and structure and, thus, changes in vine microclimate – the climate within and immediately around the vine canopy – that affect berry composition (Smart 1991). Positive relationships between NDVI values and vine pruning weight has been reported in proximal sensing studies (Stamatiadis et al. 2006; Stamatiadis et al. 2010), but late in the season, when the grower's ability to undertake any corrective actions is severely restricted. Significant relationships between berry composition variables and canopy reflectance have also been reported in remote sensing (Lamb et al. 2004; Meggio et al. 2010; Hall et al. 2011) and proximal vineyard sensing (Stamatiadis et al.

2006; Stamatiadis et al. 2010) literature.

The objective of our study was to investigate the potential of two proximal active canopy sensors to detect changes in grapevine productivity and berry composition under conditions of variable N and water supply. Developing relationships that characterize vine productivity and berry composition were the goals. Active sensor vegetation indices provided the data for this study. A traditional field response trial in two commercial winegrape vineyards over two consecutive years facilitated these objectives.

Materials and Methods

Experimental vineyards

This study was conducted during the growing seasons of 2009 and 2010 in two vineyards located in Goumenissa, northern Greece (40°52' N, 22°29' S) planted with Vitis vinifera L. cvs. Cabernet Sauvignon (CS) and Xinomavro (XM), respectively, and grafted onto 1103 Paulsen rootstock. Row and vine spacing in both experimental blocks were 2.2 m and 1.3 m, respectively, in a north to south orientation. The vines were spur-pruned and trained to a bilateral Royat system with three fixed foliage wires.

In each vineyard, three blocks were delineated, each containing a 2×3 grid of six plots with six vines per plot. The combination of two irrigation levels [Irrigation (I): 70 % of crop evapotranspiration (ETc) and no irrigation (NI)] and three rates of ammonium nitrate [0 (N0), 60 (N60) and 120 (N120) kg N ha-1] were randomly assigned to each block. A randomized complete block design was applied with six treatments (2 irrigation levels × 3 N fertilizer rates × 3 blocks) resulting in 18 plots per vineyard. Only the mean of each plot was used in the statistical analysis of the results.

Drip irrigation started at berry set in both seasons and was continued at weekly intervals, according to the estimates obtained from the potential evapotranspiration measured by an automated weather station located in the XM vineyard. Ammonium nitrate (34-0-0) was added to the soil surface at budburst in both years of experimentation.

Grapevine performance

Yield and berry composition were determined at technological maturity (late September of each year for both varieties). All clusters from each plot were collected, counted and weighted, then transferred to the laboratory where two samples of 200 berries from all parts of the cluster were randomly selected. One sample was weighed and hand-pressed for juice extraction and submitted to analysis for reducing sugars, pH and malic and tartaric acid content of the juice on a FTIR (Fourier Transform Infrared Spectroscopy) interferometer (Wine Scan FT120, FOSS Analytical A/S, Denmark, Hillerod). The second sample of berries was processed for the estimation of berry anthocyanin content and total phenolics according to the extractability assay described by Saint-Criq et al. (1998). The total anthocyanin concentration and berry total phenolics index were estimated by measuring the absorbance of the samples at 520 nm (Ribéreau-Gayon et al. 1965) and 280 nm (Ribéreau-Gayon et al. 1966) respectively. Total weight and number of dormant canes per vine were measured during the following winter pruning in both years.

Canopy reflectancere

Canopy reflectance was measured at the berry set (BS) and bunch closure (BC) growth stages with a handheld active sensor (Crop Circle ACS-210, Holland Scientific, Lincoln, NE) in 2009, whereas a newer model of active sensor (Crop Circle ACS-430, Holland Scientific, Lincoln, NE) sensor was used in 2010. The two sensors had different operational properties. The ACS-210 recorded reflectance at two wavelengths (amber at 590 nm and NIR at 880 nm) and the ACS-430 at three wavelengths (red at 630 nm, red-edge at 730 nm and NIR at 780 nm). Active sensors measure relative reflectance by calibrating reflectance of modulated light from a grey standard. Radiance from soil or adjoining rows can affect the readings of these active sensors, but the portion of the signal attributed to objects further away from the target is small because reflectance in individual wavelengths follows the inverse square of the distance rule (Stamatiadis et al. 2010). However, NDVI values remain constant when the distance of the sensor increases beyond 40 cm from the canopy (Stamatiadis et al. 2010). Multi-spectral readings were taken from each plot with the sensor placed at ~60 cm directly above the vine canopy and traveling at walking speed along the row. The data were recorded in a data logger. From the reflectance recorded in each individual waveband, the Normalized Difference Vegetation Index (NDVI) was computed as follows:

NDVIa = (NIR - amber) / (NIR + amber)

NDVIr = (NIR - red) / (NIR + red)

NDVIre = (NIR - red edge) / (NIR + red edge)

NIR is the reflectance of the near infrared waveband and amber, red, red edge the reflectance in the corresponding channels (amber for ACS-210 in 2009; red and red-edge for ACS-430 in 2010).

Statistical analysis

In each of the two experimental vineyards, a randomized complete block design, replicated in three blocks and repeatedly measured in two years, was used to evaluate the effects of two irrigation levels and three fertilization rates on the vine properties. To estimate the fixed and random effects, a linear mixed model with the restricted maximum likelihood method for the estimation of covariance parameters was used. For leaf data that were collected repeatedly within each growing season, the growth stage at each sampling was incorporated as an additional factor in the mixed model. Vineyard, year, irrigation, fertilization and growth stage were considered fixed whereas blocking as random effects. The least significant difference test was used to detect differences between the means of the fixed effects at $p<0.05$. The relations between the measured variables were evaluated by linear correlation and regression analysis. Data analysis was conducted using Statistical Analysis System software, version 9.3 (SAS Institute, Cary, NC).

Results and discussion

Vine performance

Fertilization increased the pruning weight, shoot vigor, and yield in the N_{120} treatment (data not shown). Similar effects of nitrogen fertilization have been documented in other studies where higher N availability has been associated with increased shoot vigor and modification of vine microclimate (Bell et al. 1999). Irrigated vines had more vigorous shoots, but pruning weight and yield was higher only in the XM vineyard (data not shown). In both vineyards, fertilization decreased the total phenol content, whereas irrigation increased the concentration of malic acid. pH levels did not respond to treatments in either vineyard and neither did must reducing sugars (data not shown).

Main effects on canopy reflectance

In both vineyards, the vines in the N_{120} treatment had higher values of NDVIa at 2009 and of NDVIre at 2010 (Table 1). The NDVIr index, however, had higher values in the CS vineyard only. Irrigation had no significant effects on canopy reflectance (Table 1). These effects were consistent with the increased biomass production of the N_{120} vines and the generally limited effects of irrigation on vine growth. A significant growth stage effect was observed on NDVIre in both vineyards and on NDVIr in XM (Table 1); the values of these vegetation indices were higher at the BS stage compared to their corresponding values at the BC. That growth stage effect was probably the result of the reduction in the near infrared reflectance (data not shown), after the application of a shoot trimming before the reflectance measurements at the BC stage.

Table 1 Treatment and growth stage effects on vine canopy reflectance in the vineyards of Cabernet Sauvignon and Xinomavro

	Cabernet Sauvignon			Xinomavro		
Effect	$^{1}NDVI_{a}$	$^{2}NDVI_{r}$	$^{2}NDVI_{re}$	$^{1}NDVI_{a}$	$^{2}NDVI_{r}$	$^{2}NDVI_{re}$
Stage						
BS	0.66	0.82	0.29a	0.56a	0.74a	0.25a
BC	0.66	0.82	0.27b	0.51b	0.71b	0.22b
Irrigation						
NI	0.65	0.82	0.29	0.53	0.71	0.24
I	0.66	0.82	0.27	0.54	0.74	0.23
Nitrogen						
N_0	0.64b	0.81b	0.27b	0.51b	0.71	0.22b
N_{60}	0.66ab	0.81b	0.27b	0.54a	0.73	0.23ab
N_{120}	0.67a	0.84a	0.31a	0.56a	0.74	0.25a

N_0: unfertilized; *N_{60}*: 60 kg N/ha; *N_{120}*: 120 kg N/ha; *NI*: non irrigated; *I*: irrigated at 70% of ETc; *BS*: berry set; *BC*: bunch closure

Amber based NDVI (*$NDVI_a$*); red-based NDVI (*NDVIr*); rededge-based NDVI (*NDVIre)*

Means within factors and vineyards followed by different letters are significantly different at the 0.05 probability level according to LSD

[1], [2]: data of 2009 and of 2010, respectively

Canopy reflectance and vine productivity

At the first year of the experiment, the values of NDVIa were positively and linearly correlated with the dormant wood weight in both vineyards at the BS stage (r^2=0.39, $p<0.05$, n=15 for CS and r^2=0.29, $p<0.05$, n=17 for XM), but not at the BC stage. Strong positive, but curvilinear, relationships of NDVIa values with dormant wood weight at the color change stage have been reported in Merlot vineyards with spatially variable water and nitrogen availability (Stamatiadis et al. 2006). Contrary to these earlier reports, our data showed a weak or not significant relationship of NDVIa with dormant wood production. In addition, the NDVIa values were more strongly correlated with the mean cane weight compared with the total wood weight (r^2=0.62, $p<0.001$, n=15 for CS and r^2=0.47, $p<0.01$, n=17 for XM). These findings indicate that the variation in the NDVIa values was probably more the result of the variation in the individual shoot vigor rather the result of the total canopy size variation.

At the second year of the experiment, the NDVIr and NDVIre indices, obtained by the ACS-430 canopy sensor, better predicted the vegetative growth of the vines compared to the first year. However, these relationships were stronger in the CS vineyard and at the BC stage (r=0.84, $p<0.0001$, n=16 for NDVIr and r=0.73, $p<0.001$, n=16 for NDVIre). Although stronger at the BC stage, the relationship of NDVIr with the pruning weight in the CS vineyard it was best described by a quadratic regression model. This saturation effect of higher biomass density on the values of red-based NDVI is well known: with increasing biomass density, red-based NDVI indices tend to saturate because reflectance in the red band (~675nm) decreases quickly to levels as low as 2-3%, even when the quantity of chlorophyll in the canopy increases (Gitelson et al. 2003; Samborski et al. 2009; Stamatiadis et al. 2010). According to these findings, and from a practical point of view, the NDVIr was less efficient compared to NDVIre in predicting the canopy size of CS vines. The variation of vine yield was best described by NDVIre and NDVIr indices in the 2010 year (Fig. 1); in 2009 the relationships of NDVIa with grape yield were inconsistent or not significant (data not shown).

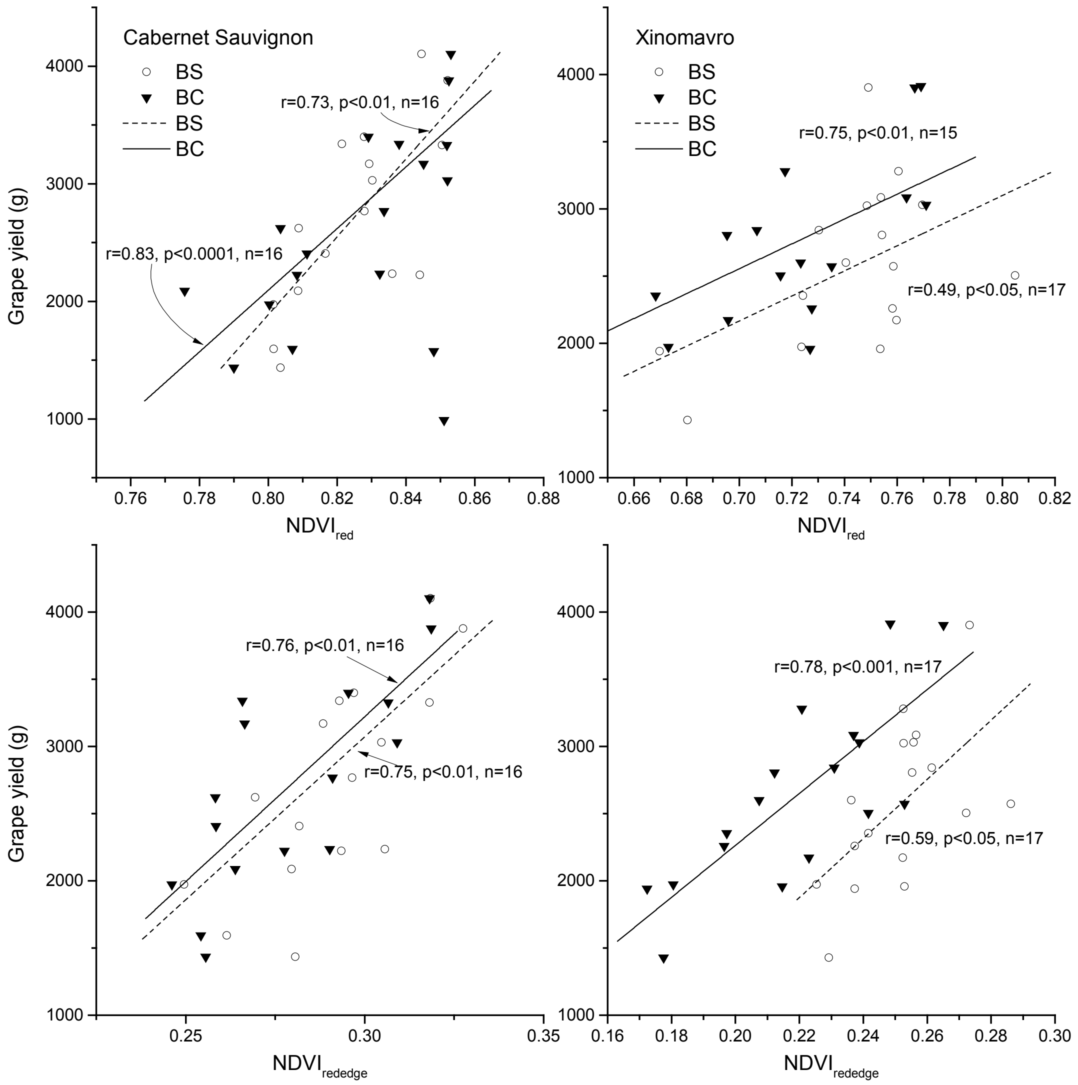

Figure 1 The relationship of NDVIred and NDVIrededge with grape yield in the Cabernet Sauvignon and Xinomavro vineyards, at berry set (BS) and bunch closure (BC).

Canopy reflectance and berry composition

The NDVIr and NDVIre values were negatively correlated with the concentration of total phenols and of total anthocyanins at the BS and BC stages in CS vineyard; but in XM only the relationship of NDVIre with total phenols was consistent at both growth stages (Fig. 2). These results indicate that the denser foliage of the more vigorous vines affected negatively berry composition because of the increased cluster shading that normally exists in vines with higher vigor (Koundouras et al. 2009). Many studies have shown that prediction of berry phenolic content by canopy reflectance data it is possible, but with varying accuracy (Lamb et al. 2004; Stamatiadis et al. 2006; Meggio et al. 2010; Stamatiadis et al. 2010; Hall et al. 2011). Our data from the ACS-430 sensor agree with the findings of these previous reports. However, all the measured indices in both year of the experiment had limited efficacy in predicting berry sugar concentration, a variable used commonly in grape pricing in the wine industry, and pH (data not shown). Berry sugar concentration depends not only on the photosynthetic activity of grapevine foliage (Guidoni et al. 2008), but also on the ability of the grapevines to remobilize carbohydrate reserves from their permanent parts (Candolfi-Vasconcelos et al. 1994). These vine properties probably explain the absence of any relationship between berry sugar concentration and canopy reflectance.

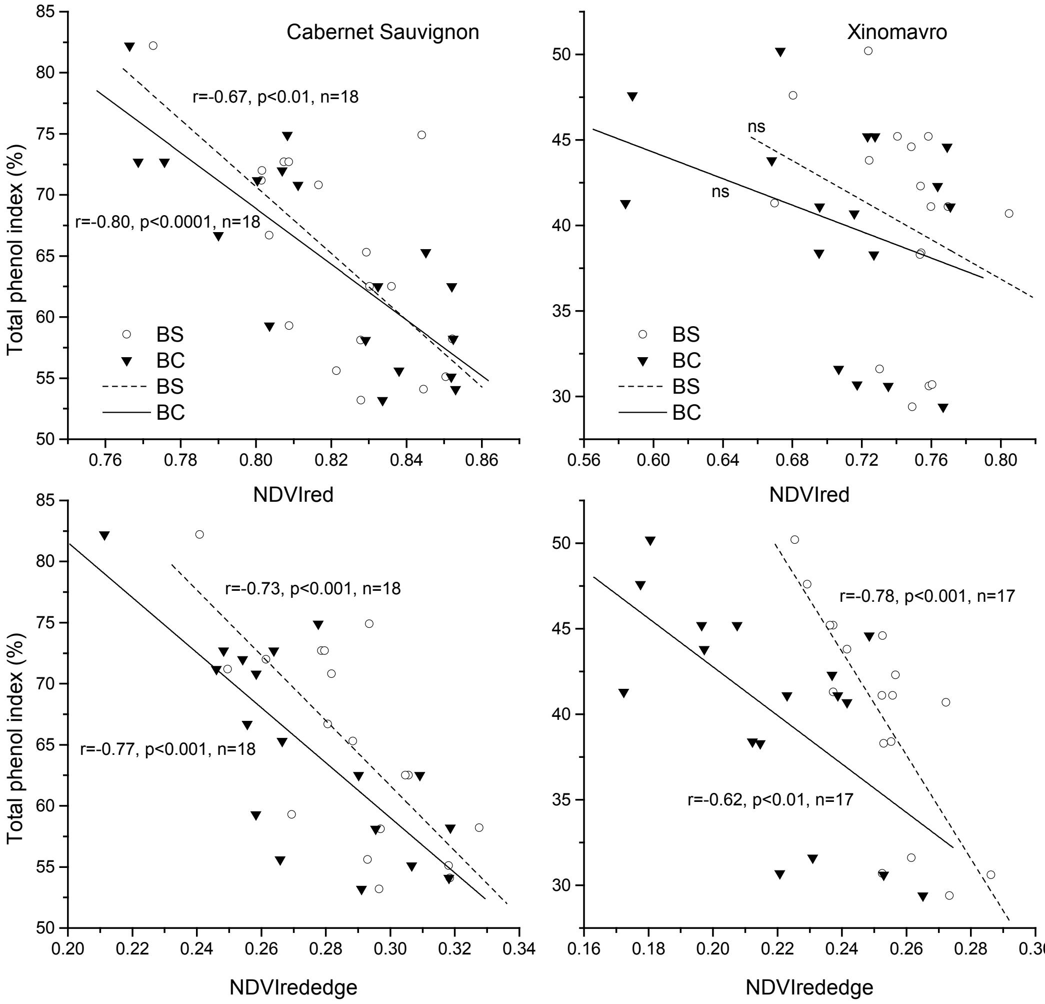

Figure 2 The relationship of NDVIred and NDVIrededge with the total phenol concentration in the berries of Cabernet Sauvignon and Xinomavro, at berry set (BS) and bunch closure (BC) stages.

These findings show that limitations exist in the use of proximal and active canopy sensing of berry composition. Moreover, further investigations are necessary to verify that the statistical significance of any correlation between berry composition and canopy reflectance has also practical value for vineyard management.

Conclusions

Treatment effects on vine productivity, berry composition, and on the readings of active sensors were observed, bur mainly as a response to nitrogen application. Consistent relationships of all ACS-430 indices with yield and of the red edge-based with total phenols were observed early in the season in both vineyards. However, the information on vine performance derived from canopy reflectance data was either vineyard or instrument specific. The Acs-430 sensor was more efficient than the Acs-210 sensor in the estimation of vine performance variables. According to these results, active canopy sensing can provide information on grapevine performance early in the season but several limitations exist.

References

Bramley, R. G. V. (2010). Precision viticulture: managing vineyard variability for improved quality outcomes. *In* Managing wine quality: Viticulture & wine quality. Vol. 1. pp. 445Pages-480. Woodhead Publishing Limited.

Rouse, J. W., Jr., Haas, R. H., Schell, J. A. & Deering, D. W. (1973). Monitoring vegetation systems in the Great Plains with ERTS. In *Third Earth Resources Technology Satellite-1 Symposium- Volume I: Technical Presentations. NASA SP-351, compiled and edited by Stanley C. Freden, Enrico P. Mercanti, and Margaret A. Becker, 1994 pages, published by NASA*. pp. 309-317.

Smart, R. (1991). Canopy microclimate implications for nitrogen effects on yield and quality. . In:M. J. Rantz (Eds.), In *Proceedings of the International Symposium on Nitrogen in Grapes and Wine* . Seattle, Washington, Usa. 18-19 June 1991. pp. 90-101. American Society for Enology and Viticulture, ASEV.

Barker D. & Sawyer J. (2010). Using active canopy sensors to quantify corn nitrogen stress and nitrogen application rate. *Agronomy Journal*, 102, 964-971.

Bell S. J. & Robson A. (1999). Effect of nitrogen fertilization on growth, canopy density, and yield of Vitis vinifera L. cv. Cabernet Sauvignon. *American Journal of Enology and Viticulture*, 50, 351-358.

Candolfi-Vasconcelos M. C., Candolfi M. P. & Kohlet W. (1994). Retranslocation of carbon reserves from the woody storage tissues into the fruit as a response to defoliation stress during the ripening period in Vitis vinifera L. *Planta*, 192, 567-573.

Gitelson A. A., Gritz Y. & Merzlyak M. N. (2003). Relationships between leaf chlorophyll content and spectral reflectance and algorithms for non-destructive chlorophyll assessment in higher plant leaves. *Journal of Plant Physiology*, 160, 271-282.

Guidoni S., Ferrandino A. & Novello V. (2008). Effects of seasonal and agronomical practices on skin anthocyanin profile of Nebbiolo grapes. *American Journal of Enology and Viticulture*, 59, 22-29.

Hall A., Lamb D., Holzapfel B. & Louis, J. (2002). Optical remote sensing applications in viticulture - A review. *Australian Journal of Grape and Wine Research*, 8, 36-47.

Hall A., Lamb D. W., Holzapfel B. P. & Louis J. P. (2011). Within-season temporal variation in correlations between vineyard canopy and winegrape composition and yield. *Precision Agriculture*, 12, 103-117.

Hatfield J., Gitelson A., Schepers J. & Walthall, C.L. (2008). Application of spectral remote sensing for agronomic decisions. *Agronomy Journal*, 100, S117-S131.

Koundouras S., Hatzidimitriou E., Karamolegkou M., Dimopoulou E., Kallithraka S., Tsialtas J. T., et al. (2009). Irrigation and rootstock effects on the phenolic concentration and aroma potential of vitis vinifera L. cv. Cabernet Sauvignon grapes. *Journal of Agricultural and Food Chemistry*, 57, 7805-7813.

Lamb D. W., Weedon M. M. & Bramley R. G. (2004). Using remote sensing to predict grape phenolics and colour at harvest in a Cabernet Sauvignon vineyard: Timing observations against vine phenology and optimising image resolution. *Australian Journal of Grape and Wine Research*, 10, 46-54.

Meggio F., Zarco-Tejada P. J., Nunez L. C., Sepulcre-Canto G., Gonzalez M. R. & Martin P. (2010). Grape quality assessment in vineyards affected by iron deficiency chlorosis using narrow-band physiological remote sensing indices. *Remote Sensing of Environment*, 114, 1968-1986.

Ribéreau-Gayon P. & Stonestreet E. (1965). Le dosage des anthocyanes dans les vins rouges. *Bulletin de la Societé Chimique de France*, 9, 2649-2652.

Ribéreau-Gayon P. & Stonestreet E. (1966). Dosage des tanins du vin rouge et détermination de leur structure. *Chim. Anal.*, 48, 188-196.

Saint-Criq N., Vivas N. & Glories Y. (1998). Maturité phénolique: définition et contrôle. *Rev. Fr. Oenol.*, 173, 22-25.

Samborski S., Tremblay N. & Fallon, E. (2009). Strategies to make use of plant sensors-based diagnostic information for nitrogen recommendations. *Agronomy Journal*, 101, 800-816.

Stamatiadis S., Taskos D., Tsadila E., Christofides C., Tsadilas C. & Schepers J. S. (2010). Comparison of passive and active canopy sensors for the estimation of vine biomass production. *Precision Agriculture*, 11, 306-315.

Stamatiadis S., Taskos D., Tsadilas C., Christofides C., Tsadila E. & Schepers J. S. (2006). Relation of ground-sensor canopy reflectance to biomass production and grape color in two merlot vineyards. *American Journal of Enology and Viticulture*, 57, 415-422.