

## Using hyperspectral remote sensing to map grape quality in 'Tempranillo' vineyards affected by iron deficiency chlorosis

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### Summary

The objectives of this work were (i) to investigate the relationships between chlorophyll a+b concentration in leaves ( $C_{ab}$ ) and grape composition parameters in vineyards affected by iron chlorosis, and (ii) study whether the assessment of  $C_{ab}$  from hyperspectral remote sensing imagery could be useful to map different potential quality zones in these vineyards. A field trial was conducted in a vineyard with the chlorosis susceptible cultivar, 'Tempranillo', over '110 Richter', located in Northern Spain. Three experimental treatments were applied: 0, 2, and 4 foliar sprayings with a lignin-sulphonate derived product (10 % water soluble Fe) in a randomized design with 3 replications. The yield and grape composition parameters at harvest were measured for each base-plot (10x10 m in size), and related with chlorophyll concentration in leaves. On the other hand, a total of 24 'Tempranillo' commercial vineyards were identified for field and airborne data collection with CASI hyperspectral sensor, comprising 103 study areas of 10x10 m in size. A total of 1467 leaves were collected for determining pigment concentration and optical properties. Several narrow-band vegetation indices were calculated from leaf reflectance spectra. Results of trial showed that the lack of pigmentation in leaves was a major factor limiting grape ripening. Significant linear regressions between  $C_{ab}$  and total soluble solids concentration and colour density of the must were detected. Estimation of  $C_{ab}$  using the image-calculated TCARI/OSAVI through the PROSPECT-rowMCRM model simulation for all study zones, including the specific ligninsulphonate experiment, demonstrated the potential of hyperspectral imagery for mapping  $C_{ab}$  in vineyards for chlorosis detection using remote sensing methods. Given the described relationship between  $C_{ab}$  and quality parameters in vineyards affected by iron chlorosis, high-spatial resolution imagery with narrow bands might enable the segmentation in areas of potential quality in the framework of precision viticulture.

**Key words:** chlorophyll, iron deficiency, image processing, management zones, must composition, radiative transfer, remote sensing, *Vitis vinifera* L.

### Introduction

Iron deficiency is one of the major problems affecting a variation of crop species grown in calcareous and alkaline soils, and resulting frequently in high economical losses. It is well-known that Fe deficiency is characterized by inter-venial yellowing of young leaves (chlorosis) whereas more mature leaves are frequently green. The low Fe assimilation in chlorotic plants is mainly due to the effect of the soil bicarbonate (MENGEL *et al.* 1984 a), which impairs Fe uptake mechanisms of the root system (MARSCINER *et al.* 1986).

Fe is essential for the structure and function of the photosynthetic apparatus of plants (TERRY and ABADIA 1986). The low photosynthesis rate occurring in chlorotic plants causes a slow and short shoot growth, limits potential productivity and considerably depresses yield and vigour of vineyards (ANDERSON 1982, CHIEN and BARAK 1982, TAGLIAVINI and ROMBOLÀ 2001). Fe deficiency also leads to a poor quality of must, reducing sugar and anthocyanin accumulation in berries during ripening, and increasing total acidity (CASTINO *et al.* 1987, VELIKSAR *et al.* 2005).

Fruit composition plays a critical role in the quality of wines. Phenolic compounds, anthocyanins and tannins, are responsible for the colour and astringency of red wines (RIBÉREAU-GAYON and GLORIES 1987). The synthesis and accumulation of phenolic compounds in grapes through ripening is greatly influenced by photosynthetic capacity of the vines and the bunch exposure to sunlight (PIRIE and MULLINS 1980, SMART and ROBINSON 1991); Even when cultivar and clone rootstock are identical, grape and wine quality are influenced by subtle differences in characteristics of vineyard: soil type, microclimate, slope, exposure, soil water holding capacity, drainage, etc. (SMART and ROBINSON 1991, WILSON 1998, BRAMLEY 2001).

HALL *et al.* (2002) defined photosynthetically active biomass (PAB) as a term which integrates vine-leaf biomass (canopy size, density and vigour) and leaf chlorophyll content. Spatial variations in vine PAB, or its equivalents, has been linked to spatial variations in yield (LAMB *et al.* 2001) and fruit and wine composition (JOHNSON *et al.* 2001, LAMB *et al.* 2004, CORTELL *et al.* 2005). Grapevine PAB depends on numerous ambient factors, as variation in topography, climatic conditions, physical and chemical characteristics

of the soil, or pest and disease incidence. In calcareous soils, Fe availability can be one of the major factors modifying canopy size and chlorophyll content in leaves, which have a considerable spatial variation in vineyards affected by chlorosis (ZARCO-TEJADA *et al.* 2005 a).

Several studies demonstrate the feasibility of chlorosis detection in vegetation through chlorophyll a+b concentration ( $C_{ab}$ ) estimation using spectroscopy and leaf optical properties in the 400-2500 nm spectral region (JACQUEMOUD *et al.* 1996, CARTER and SPIERING 2002, SIMS and GAMON 2002, GITELSON *et al.* 2003, Le MAIRE *et al.* 2004). In recent studies, new optical indices have been proposed to relate crop physiological status with hyperspectral data through their relationship to biochemical constituent concentrations such as chlorophyll (VOGELMANN *et al.* 1993, CARTER 1994, GITELSON and MERZLYAK 1996, ZARCO-TEJADA *et al.* 2001, 2004, 2005 a, 2005 b, HABOUDANE *et al.* 2002). Current research efforts in precision viticulture and on the temporal and spatial monitoring of *Vitis vinifera* L. show a growing interest in remote sensing methods due to its potential for estimating vine biophysical variables such as shape, size and vigour, potential indicators of vine yield and grape quality (HALL *et al.* 2002). These studies point toward the application of new techniques in viticulture based on precision agriculture, introducing methods focused on describing homogeneous management zones derived from remotely-sensed biophysical variable estimates (HALL *et al.* 2002), connecting the within-field variability and the suggested classification of the field into different vigour classes with a potential wine quality production (JOHNSON *et al.* 2001). Progress on crop condition in vineyards has been made at the leaf level studying absorbance in the visible region in the field (SCHULTZ 1996), and detecting phenology and grape colour at harvest to gather information about berry phenolics (LAMB *et al.* 2004).

Given the links between  $C_{ab}$  and the quantitative and qualitative response of vines, the potential exists for generating synoptic information concerning spatial variations in yield and quality attributes in vineyards affected by iron chlorosis. A recent study conducted on 24 vineyards flown with airborne hyperspectral sensors such as the Compact Airborne Spectrographic Imager (CASI), the Reflective Optics System Imaging Spectrometer (ROSIS) and the Digital Airborne Imaging Spectrometer (DAIS-7915) sensors demonstrated the successful estimation of vine chlorophyll concentration using coupled radiative transfer models PROSPECT (JACQUEMOUD and BARET 1990) and rowM-CRM to account for the effects of vineyard structure, vine dimensions, row orientation and soil and shadow effects on the canopy reflectance (ZARCO-TEJADA *et al.* 2005 b). Results using the Transformed Chlorophyll Absorption in Reflectance Index (TCARI), and the Optimized Soil-Adjusted Vegetation Index (OSAVI) in the form TCARI/OSAVI yielded  $R^2 = 0.67$  and  $RMSE = 11.5 \mu g cm^{-2}$  when estimating chlorophyll concentration on 103 study sites imaged on 24 fields. This methodology suggests the potential input for a successful field segmentation into homogeneous zones to facilitate segmented harvests. From a management point of view, it is interesting to characterize the spatial variability in grape quality in order to produce grapes with higher unit

values. Fruit of different quality zones can then be batched for separate winemaking.

The objectives of this work were: (i) to establish relationships between variations in foliar chlorophyll and fruit composition in a commercial vineyard affected by iron chlorosis (same clone, rootstock, age, and viticulture practices); and (ii) to study if the assessment of chlorophyll by hyperspectral remote sensing techniques could be useful in these conditions to map different potential quality zones in vineyards.

## Material and Methods

**Study site description:** The investigation, conducted in 2003, included a couple of 24 full production vineyards. Each vineyard included 5 study areas of 10 x 10 m in size. Field sampling was carried out in these areas, concurrent with the airborne overflights, for biochemical analysis of leaf  $C_{ab}$ , as well as to study the vine optical properties. Fig. 1 shows a selection of 3 vineyard fields used in this study imaged by the CASI airborne sensor at 1 m spatial resolution, and the study blocks selected for field data collection. Vineyards are located in the western area of Ribera del Duero Appellation d'Origine (Northern Spain), at an altitude of about 800 m above sea level. The



Fig. 1: Airborne hyperspectral CASI images acquired over three of the sites used in this study. The images were collected at 1 m spatial resolution, showing the 10x10 m study blocks used for field sampling collection.

study sites used for ground and airborne collection were carefully selected from a plot network currently monitored by the local government to assure a gradient in the leaf biochemistry as sought for this investigation. All vineyards correspond to 'Tempranillo' grafted on '110-Richter', and are between 7 and 16 years old. With 2200-4000 vines per hectare, plants are trained to a simple or double Cordon Royat system. The fruiting wires are at a height of about 60 cm, and all shoots are guided to a vertical position. The soils are calcareous, poor in organic matter (about 7.6 g·kg<sup>-1</sup>), with a medium-weighted texture and average pH of 8.7. Mean extractable K concentration is medium (235 mg kg<sup>-1</sup>), but extractable P (13.7 mg kg<sup>-1</sup>) and Mg (1.53 meq/100 g) concentration in the soil are low. Concentrations of active carbonate (up to 17.6 %) and DPTA extractable Fe (1.2 to 7.6 mg kg<sup>-1</sup>) are highly heterogeneous within the area.

**Ligninsulphonate experiment:** A field trial was conducted to evaluate yield and fruit quality of chlorotic vines in relation to leaf  $C_{ab}$  levels, as affected by the foliar application of different amounts of a Fe-containing product. Experiment was carried out in a commercial vineyard located in the same study site described above. The vineyard (cv. Tempranillo over 110-Richter) is strongly affected by Fe deficiency in a soil with a mean chlorotic power index of 64. Grapevines were planted in 1990, and spaced 2.5 (between rows) x 1.0 m (between plants). The pruning charge was 36000 buds per hectare. The cultural practices were the usual ones in the zone.

Brexil (10 % water soluble Fe, Valagro), a ligninsulphonate derived product, was used to correct iron chlorosis. Three experimental treatments were considered: T0 (control), T2 (two supplies on 22 May and 9 June) and T4 (4 supplies on 22 May, 9 and 25 June, and 4 July). Brexil were applied at 2.5 g l<sup>-1</sup> in three first sprayings and at 4 g l<sup>-1</sup> in the 4<sup>th</sup> application. No adjuvants were added. The treatments were given over all the foliage of plants to runoff, with a 16 L hand sprayer. The trial was conducted in a randomized design with 3 replications. Each base-plot had 40 vines distributed in 4 rows (10 x 10 m in size). At harvest (October 2003), vine yield and average berry weight were recorded. The must obtained from a sample of 100 berries in each experimental treatment was extracted to determine the concentration of total soluble solids (g l<sup>-1</sup>), total acidity (g l<sup>-1</sup> tartaric acid equiv.), total polyphenols index and colour density, according to the official methods of analysis established in the EEC 2676/90 Regulations (European Commission, 1990). The visible spectra of musts were obtained with a Jasco V-530 UV-VIS spectrophotometer. The effects of the treatments on yield and must composition were studied by analysis of variance applying models of fixed effects. The mean separations were determined by the LSD test.

**Analysis of nutrient and chlorophyll in leaves:** Concurrent with the airborne overflights, field sampling campaign were conducted in third week of July 2003 for biochemical analysis of leaf  $C_{ab}$ , as well as to measure reflectance (R) and transmittance (T) from leaf samples to study the vine optical properties. A total of 80 leaves were sampled from each 10x10 study area, including base plots of ligninsulphonate experiment.

Fifty leaves were used for measuring dry matter and elements N, P, K, Ca, Mg and Fe, 20 leaves for chlorophyll a+b concentration ( $C_{ab}$ ) determination, and 10 leaves per site for conducting reflectance and transmittance measurements.

The leaves were sampled from the top of the canopy, eliminating the small leaves indicative of low expansion. Selected leaves of ligninsulphonate experiment were grown after spray application, and thus not contaminated with Fe. Structural measurements on each study site consisted of grid size, number of vines within the 10x10 m site, trunk height, vegetation height and width, and row orientation.

Leaves were placed in paper bags to allow tissue respiration and conservation, then stored at 4 °C prior to analysis of reflectance and transmittance, and then stored in a freezer at -8°C prior to pigment determination (as described in detail in ZARCO-TEJADA *et al.* 2005 a).

**Remote sensing methods for chlorosis detection at leaf and canopy levels:** Leaves used for measuring optical properties were taken to the laboratory and reflectance and transmittance measurements made on the same day to avoid leaf structure and pigment degradation. Reflectance and transmittance measurements of vine leaves were conducted with a Li-Cor 1800-12 Integrating Sphere (Li-Cor, Inc., Lincoln, NE, USA), coupled by a 200 µm diameter single mode fiber to an Ocean Optics model USB2000 spectrometer (Ocean Optics Inc., Dunedin, FL, USA), with a 2048 element detector array, 0.5 nm sampling interval, and 7.3 nm spectral resolution in the 350-1000 nm range. Single leaf reflectance and transmittance measurements were acquired following the methodology described in the manual for the Li-Cor 1800-12 system (Li-Cor-Inc. 1984) modified by HARRON (2000) to correct for stray light in the integrating sphere. The protocol is described in detail in ZARCO-TEJADA *et al.* (2005 a) requiring a total of 5 measurements to calculate the stray-light corrected leaf hemispherical reflectance (R) and transmittance (T) using a reference target in the integrating sphere. Fig. 2 shows a selection of leaf reflectance and transmittance spectra measured from 2 leaves with extreme low and high values of chlorophyll concentration (15.5 and 53.8 µg cm<sup>-2</sup>). Leaf-level vegetation indices that demonstrated to track changes in chlorophyll concentration (see ZARCO-TEJADA *et al.* 2005a for a complete discussion of leaf-level indices for chlorophyll concentration estima-

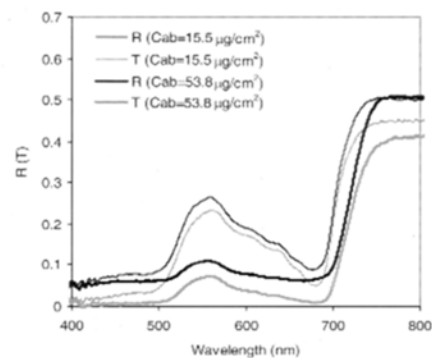


Fig. 2: Leaf reflectance and transmittance spectra measured with integrating sphere and fiber spectrometer, showing spectral differences between a chlorotic and healthy vine leaf.

tion in vineyards) were calculated from the leaf reflectance and transmittance measured on the dataset collected in the field. In such study, indices such as Vogelmann (VOGELMANN *et al.* 1993) calculated as  $(R_{734} - R_{747}) / (R_{715} + R_{726})$ , Z&M (ZARCO-TEJADA *et al.* 2001) calculated as  $(R_{750} / R_{710})$ , and TCARI/OSAVI (HABOUDANE *et al.* 2002; 2004) obtained the best results for chlorophyll a+b estimation at the leaf level, and were used in this study.

The index calculated as a combination of the Transformed Chlorophyll Absorption in Reflectance Index (TCARI) (HABOUDANE *et al.* 2002), and the Optimized Soil-Adjusted Vegetation Index (OSAVI) (RONDEAUX *et al.* 1996) in the form TCARI/OSAVI has been demonstrated as the most consistent index for estimating  $C_{ab}$  on aggregated and pure vine pixels extracted from hyperspectral imagery, demonstrating to successfully minimize soil background and LAI variation in crops, providing predictive relationships for precision agriculture applications with hyperspectral imagery in closed crops (HABOUDANE *et al.* 2002) and open tree canopy orchards (ZARCO-TEJADA *et al.* 2004). Predictive relationships for  $C_{ab}$  were developed linking PROSPECT leaf model (JACQUEMOUD and BARET 1990) with the Markov-Chain Canopy Reflectance Model (MCRM) (KUUSK 1995a, 1995b) with additions to simulate the row crop structure, called rowMCRM and developed within the frame of the Crop Reflectance Operational Models for Agriculture (CROMA) project. The PROSPECT-rowMCRM model was used to predict  $C_{ab}$  as function of input image-calculated TCARI/OSAVI index, considering vine LAI, field-measured vine dimensions, and image-extracted soil background, row-orientation and viewing geometry values (a full description of the scaling-up methodology can be found in ZARCO-TEJADA *et al.* 2005 a). The prediction model shown in Equation (1) obtained through PROSPECT-rowMCRM simulation was used to estimate  $C_{ab}$  on each 10x10 experimental block from the vineyard fields.

$$C_{ab} = 316.03 \cdot e^{-14.798 \frac{TCARI}{OSAVI}} \quad (1)$$

where TCARI/OSAVI was calculated from each spectra extracted from the CASI airborne imagery, and inputs required for PROSPECT-rowMCRM used in this study were the leaf and canopy-level parameters. At the leaf level, chlorophyll a+b ( $C_{ab}$ ), dry matter ( $C_m$ ), water concentration ( $C_w$ ), and structural parameter (N) were used as input for PROSPECT model. At canopy level, canopy layer and structure parameters such as the row leaf area index (LAI), leaf angle distribution function (LADF), relative leaf size ( $h_s$ ), Markov parameter ( $\lambda z$ ), leaf transmittance coefficient (t), leaf hair index ( $l_h$ ), canopy height ( $C_H$ ), crown width ( $C_w$ ), visible soil strip length ( $V_s$ ), the angular difference between sun azimuth and row direction ( $\psi$ ) and background and viewing geometry parameters such as soil reflectance ( $\rho_s$ ), Angstrom turbidity factor ( $\beta$ ), and the viewing geometry ( $\theta_s, \theta_v, \phi$ ) were needed for the rowMCRM model simulation to generate the predictive equation. Validation was conducted comparing the  $C_{ab}$  estimated from model simulation (Equation 1) with laboratory determinations of  $C_{ab}$  on leaf samples acquired on each study site of this experi-

ment. Spectra was extracted from the 1 m resolution CASI imagery using aggregated pixels within the 10x10 m plots, and using the brightest pixels (targeting vine and removing soil and shadow effects) within each study plot.

## Results and Discussion

**Chlorophyll concentration and mineral composition of leaves:** The average  $C_{ab}$  concentration in the sampling leaves collected in field trial was  $30.37 \mu\text{g}\cdot\text{cm}^{-2}$ , but there were wide variations, the range of which made it impossible to record differences between treatments. In addition to the spraying of different doses of Fe-ligninsulphonate-derived product, the differences in leaf pigmentation among experimental plots might be related to the spatial variation in the physical and chemical properties of the soil, and in plant vigour itself within the vineyard affected by chlorosis. Vines root systems exploit a relatively large volume of soil with heterogeneous characteristics; some roots are developed in sites favourable to Fe uptake while others are located in poor areas. On the other hand, the redistribution of Fe from green canopy parts to chlorotic ones in apical part of the shoots (sampling zone) is restricted during active vegetative growth.

The iron concentration in leaf blades, measured fifteen days after stopping the ligninsulphonate applications was higher in T4 ( $101 \text{ mg kg}^{-1}$ ) than in T0 and T2 (less than  $55 \text{ mg kg}^{-1}$ ). The variance analyses did not show statistically significant differences between the macronutrient concentrations of the different experimental treatments. The average N, P, K and Ca concentrations of the whole set of treatments were intermediate between the reference levels for blades at fruit set and at veraison proposed by GONZÁLEZ and MARTÍN (2005) for cv. Tempranillo in the Ribera del Duero Appellation d'Origine area. The Mg values ( $0.52\text{-}0.58\%$  over dry matter) were higher than the reference ones ( $0.22\text{-}0.40\%$ ) throughout the trial, while Fe values were lower than reference ( $73\text{-}141 \text{ mg kg}^{-1}$ ) in treatments T0 and T2.

The  $C_{ab}$  concentration correlated inversely with phosphorus levels ( $r = -0.69$ ;  $p < 0.05$ ) and potassium levels ( $r = -0.67$ ,  $p < 0.05$ ) in the blades, in line with the observations of other researchers (JONES and WALLACE 1992). Phosphorus behaved as an iron antagonist, creating insoluble iron phosphate both in the soil and inside the plant. In situations where there was iron chlorosis, when P leaf concentration increased, the P/Fe ratio also increased, without any correlation between soil and leaf phosphorus content (FREGONI 1998).

No significant relationships were detected between the iron concentration of sampling leaves in all study areas (sampling zones and field experiment) and chlorophyll concentration, whether expressed on dry matter or in leaf surface basis. It is well known that iron concentration in leaves is not a valid indicator for diagnosing iron chlorosis. The named iron chlorosis paradox (RÖMHELD 1997, BAVARESCO *et al.* 1999) is manifested when chlorotic plants have higher leaf Fe concentration (on the dry weight ba-



sis) than green ones. This effect is associated with a severe growth inhibition in affected leaves, which decreases its weight per area (BAVARESCO *et al.* 1999). On the other hand, large apoplasmic inactive Fe pools are accumulated both in Fe-sufficient and Fe-deficient leaves (see ABADIA *et al.* 2004).

**Relationships between foliar chlorophyll concentration and grape composition:** The decrease in photosynthesis activity caused by the lack of iron (PALLIOTI *et al.* 1994; TAGLIAVINI and ROMBOLÀ 1995; RYSER and HELLER 1997) had a negative effect on all the physiological processes of the plants, which showed overall low values of vigour and yield. The progressive recovery of net assimilation levels as a consequence of the iron input to the leaf made possible an increase in the average cluster weight and consequently in yield, which was 66 % higher in T4 than in the untreated controls (Table). However, the increase in yield was not significantly linked to the increase in chlorophyll level in sampled leaves ( $r = -0.13$ ;  $p > 0.05$ ). Due to the poor mobility of iron, it is known that plant growth is often considerably depressed independent of whether young leaves are chlorotic or green (MENGEL *et al.* 1984 b; GRUBER and KOSEGARTEN 2002).

There were no significant differences for must composition parameters between experimental treatments (Table). Like leaf chlorophyll concentration, the grape composition parameters were probably affected by parameters independent of the experimental design (such as plant vigour and canopy size), which might modify the availability of water and mineral nutrients, including iron. When there were no major constraints such as iron deficiency, a low-moderate vigour increased the polyphenol content and the colour intensity of the berries (SMART and ROBINSON 1991; LAMB *et al.* 2004; CORTELL *et al.* 2005).

Nevertheless, when we applied linear regression methods over values of grape composition parameters obtained in base plots, a clear relationship between the leaf chlorophyll concentration, soluble solids concentration and colour density of the must was detected (Fig. 3). The presence of a greater quantity of pigments in leaves increased the must absorptions at 420 nm, and more particularly at 620 nm (blue colour component), as shown in Fig. 3. Absorptions

at those two levels represented overall between 53 % and 58 % of must colour density in the different experimental treatments. We would emphasize that, despite the hetero-

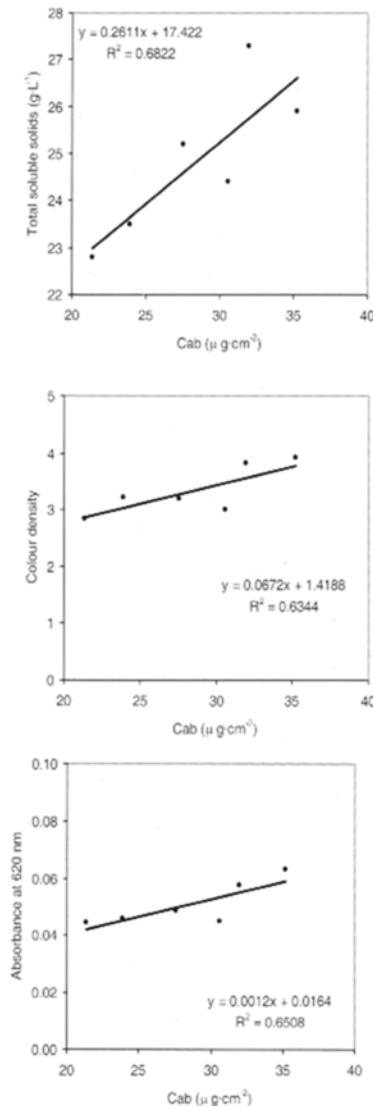


Fig. 3: Relationships obtained between chlorophyll concentration in leaf blades, total soluble solids concentration (upper), colour density (centre) and absorbance at 620 nm (lower) of the must at harvest.

Table

Mean values of yield, weight of 100 berries and must composition parameters for the different treatments with zero (T0), two (T2) and four (T4) foliar applications of Fe-ligninsulphonate

Parameters	Treatments		
	T0	T2	T4
Yield (kg m <sup>-2</sup> )	0.49 a	0.63 b	0.81 c
Weight of 100 berries (g)	161.20 a	173.80 a	179.40 a
Must composition			
Soluble solids concentration (g l <sup>-1</sup> )	255.30 a	250.50 a	239.50 a
Total acidity (g l <sup>-1</sup> tartaric acid equiv)	4.41 a	4.78 a	4.79 a
Total polyphenols index	20.60 a	18.72 a	17.61 a
Colour density	3.55 a	3.34 a	3.02 a

Values with different letters in the same line are significantly different ( $p < 0.05$ ).

geneous nature of the soil and high vine-size variations in field trial,  $C_{ab}$  in leaves was identified as a crucial factor for the processes of synthesis and accumulation of sugars and anthocyanins in the grapes during ripening.

In conclusion, the pigmentation of young leaves in summer may be a valid symptom of plant stress. In vineyards affected by iron chlorosis, it might be taken as an indicator of the extent to which the grapes will acquire a sufficient qualitative status at harvest. Thus, the potential exists for generating synoptic information concerning spatial variations in quality attributes in these vineyards.

**Chlorosis detection with remote sensing methods:** The work conducted at the leaf level for the entire experiment was based on 103 study areas, with 605 leaves for measuring the optical properties of reflectance and transmittance, and a subsample of 80 leaves from the specific ligninsulphonate trial. Results for this experiment showed that narrow-band indices VOG2 ( $(R_{734} - R_{747}) / (R_{715} + R_{726})$ ), ZM ( $R_{750} / R_{710}$ ), and TCARI/OSAVI yielded the best results for  $C_{ab}$  estimation ( $R^2 = 0.88$ ;  $R^2 = 0.89$ ;  $R^2 = 0.92$ , respectively) (Fig. 4). In addition, the Nor-

malized Difference Vegetation Index (NDVI), a traditional index related to vegetation structure and condition, and widely used for segmenting zones in precision agriculture, was shown to saturate at lower  $C_{ab}$  values than TCARI/OSAVI, obtaining poorer results for  $C_{ab}$  estimation ( $R^2 = 0.44$ ) at the leaf level in vine leaves.

The indices TCARI and TCARI/OSAVI calculated from the 1-m CASI imagery on each 10x10 m block demonstrated consistent relationships with  $C_{ab}$  at the canopy level (Fig. 5) on the ligninsulphonate experimental subset. Determination coefficients for TCARI ( $R^2 = 0.54$ ), and combined index TCARI/OSAVI ( $R^2 = 0.57$ ) demonstrated superior sensitivity for  $C_{ab}$  estimation at canopy level than the traditionally-accepted NDVI index for vegetation condition monitoring ( $R^2 = 0.03$ ) (Fig. 5). Estimation of  $C_{ab}$  using the image-calculated TCARI/OSAVI through the PROSPECT-rowMCRM simulation for this specific ligninsulphonate experiment yielded  $R^2 = 0.53$  and  $RMSE = 8.2 \mu g cm^{-2}$ , demonstrating the potential of hyperspectral imagery for mapping  $C_{ab}$  in vineyards and chlorosis detection using remote sensing methods. The successful estimation of vine

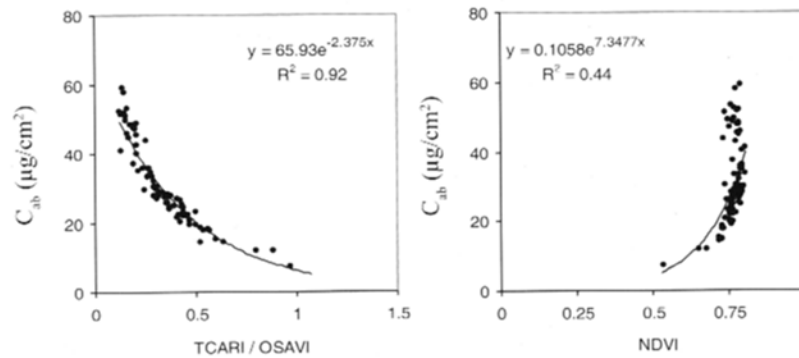


Fig. 4: Leaf-level relationships obtained between chlorophyll concentration ( $C_{ab}$ ) and indices TCARI/OSAVI (left) and NDVI (right) from leaves collected over the site used in this study.

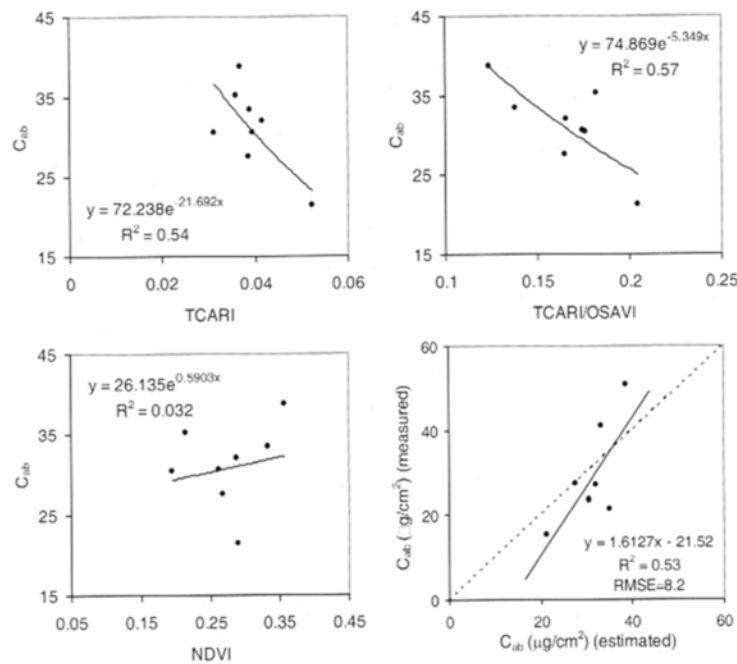


Fig. 5: Relationships obtained between chlorophyll concentration ( $C_{ab}$ ) and indices calculated from airborne CASI images: TCARI (upper left), TCARI/OSAVI (upper right), and NDVI (lower left). The relationship obtained between measured and image-estimated  $C_{ab}$  using a scaling-up algorithm through radiative transfer simulation is shown in the lower right plot.

chlorophyll concentration using high-spatial resolution imagery, and the described relationship between  $C_{ab}$  and quality parameters enable the segmentation of vineyards in areas of potentially homogeneous characteristics. Similarly to segmentation attempts using NDVI as a surrogate for LAI, this study conducts the field segmentation based on  $C_{ab}$  estimated from TCARI/OSAVI index through the coupled PROSPECT-rowMCRM radiative transfer model. The estimated  $C_{ab}$  at the vine level from 1 m CASI imagery was used as input for K-means unsupervised classification generating 3 classes of potentially high, medium and low quality as function of the chlorophyll concentration estimated (Fig. 6). The K-means unsupervised classification method calculates initial class means that are evenly distributed in the data space, iteratively clustering the pixels into the nearest class using a minimum distance technique (TOU and GONZÁLEZ 1974).

These results suggest the potential application of hyperspectral remote sensing imagery to map vine potential quality in the framework of precision viticulture. The estimation of  $C_{ab}$  concentrations in leaves before veraison could be a useful viticultural management tool for predicting grape quality at harvest in vineyards affected by iron chlorosis. The generation of quality maps from hyperspectral remote sensing would facilitate spatial segmented harvests and their separate winemaking. These techniques may require specific calibration for different cultivars. Its

application for sites with a low Mg status, which also result in chlorosis affecting must quality, should be investigated.

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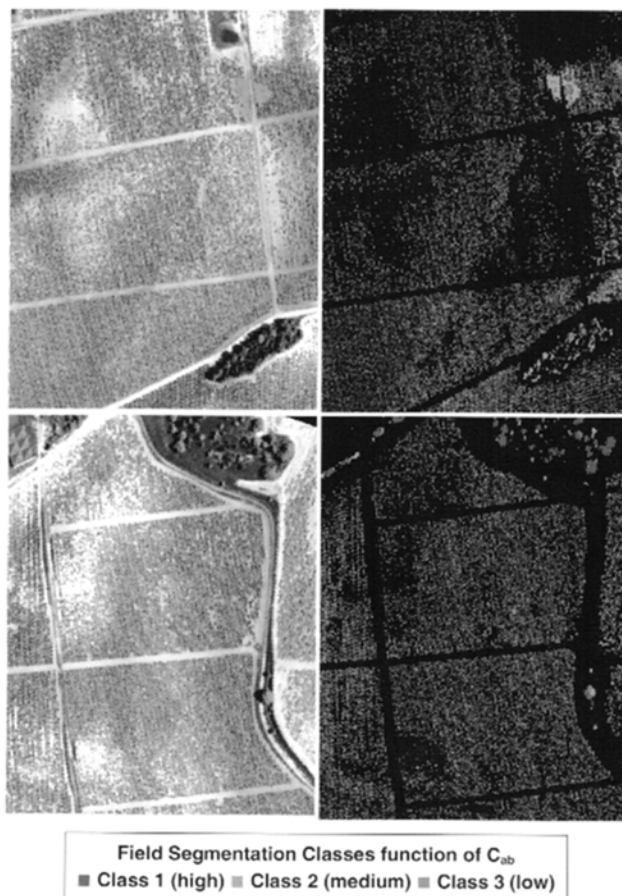


Fig. 6: Hyperspectral CASI images collected over two fields part of this study (upper and lower plot). Figs on the right show the mapping of the chlorophyll concentration using the scaling-up algorithm through radiative transfer simulation.

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