



Validation of thermal indices for water status identification in grapevine



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ABSTRACT

The use of thermal imaging represents a substantial progress in monitoring plant water status and therefore drought stress in field conditions. However, the effective use of thermal imaging requires consistent methods for data acquisition and image analysis. We determined the temperature variation of grapevine canopies by the use of thermal imaging in a proximal manner, and calculated stomatal conductance index (I_G) and crop water stress index (CWSI), aiming to assess the plant water status that was measured as variations in stomatal conductance. The study was conducted in a hillside commercial vineyard with Graciano (*Vitis vinifera* L.) vines grown under two different water statuses. Leaf stomatal conductance was measured to determine plant water status and indices derived from individual grapevine leaves, clusters and canopies were assessed by thermal imaging. Measurements were carried out under different light conditions (sunlit and shaded part of the canopy) and at different times of the day (morning, midday and afternoon) to analyze the robustness and sensitivity of thermal imaging for detecting changes in a range of plant water status and experimental conditions.

Highly significant correlations were found between the calculated indices (I_G and CWSI) and the measured stomatal conductance. The strongest relationships between I_G and CWSI and the measured stomatal conductance were obtained at midday, on the shaded side of the grapevine canopy. Therefore, those sampling conditions were the most appropriate to estimate variation in stomatal conductance in a non-contact manner through the use of thermal imaging.

Furthermore, the sensitivity of berry temperature to changes in grapevine water status was quantified. The acquired thermal images for vine clusters were corrected for berry emissivity, which was estimated to be 0.96. For water stressed grapevines, berry temperature was increased as much as 1–2 °C above clusters from non-water stressed grapevines, thus potentially affecting berry composition.

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1. Introduction

A large proportion of vineyards are located in semi-arid regions with seasonal drought (e.g. Mediterranean type climates) where soil and atmospheric water deficit, together with high temperatures, exert large constraints on yield and quality (Medrano et al., 2003; Chaves et al., 2007, 2010), thus requiring precise regulation of water supply. An improvement in the efficiency of water use is therefore necessary in vineyard management, with finely tuned deficit irrigation being able to fulfill that role. Thus, there is a real need for sensitive and robust techniques to accurately detect plant water stress. In that sense, the recent development of portable

thermal cameras has greatly extended the opportunities for the analysis of the thermal properties of plant water status in a non-invasive way (Jones, 1999a; Grant et al., 2007; Fuentes et al., 2012; Costa et al., 2012).

Infrared thermography has been proposed as an appropriate methodology for assessing grapevine water status (Jones et al., 2002; Costa et al., 2012) and as a tool to support irrigation scheduling (Idso et al., 1981; Jackson et al., 1981; Jones et al., 2002; Jones and Leinonen, 2003; Grant et al., 2006, 2007). This technique is based on the fact that when water is lost through the stomata, leaf temperature decreases, but when stomata close, transpiration no longer occurs and the temperature of leaves increases due to the fact that heat dissipation by transpiration stops (Gates, 1964; Fuchs, 1990; Jones et al., 2002).

The importance of stomatal closure in response to long term water stress was previously measured in field-grown grapevines by several authors (Winkel and Rambal, 1993; Medrano et al., 2003) and leaf temperature was tested as an indicator of such plant

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responses to various stresses: biotic (Chaerle and Van Der Straeten, 2000; Oerke et al., 2006) and abiotic, in particular limited plant water availability (Jones, 1999a; Jones et al., 2002; Grant et al., 2007). Therefore, estimation of leaf stomatal conductance to water vapor from leaf temperature measurements avoids the need for multiple and time consuming leaf gas-exchange measurements and provides important information about the degree of stomatal closure (Berni et al., 2009a,b).

The recent development of field-portable thermal imaging systems opens up the possibility to study not only the average temperatures over a defined area, but also to obtain frequency distributions of temperature over the area or region of interest (ROI). Moreover, thermal imaging can also reveal spatial heterogeneity within a leaf (Prytz et al., 2003), which has been shown to occur at the whole leaf level (Fuchs, 1990; Jones, 1999b; Oerke et al., 2006). However, factors such as solar radiation, wind speed, air temperature, and air humidity have important effects on leaf temperature (Jones, 1992). Attempts to solve these problems have been presented by several authors by the use of either “dry” (Qiu et al., 1996) or “wet” and “dry” (Jones et al., 1997; Jones, 1999a) reference surfaces. In the second case, the observed leaf temperatures are compared with the temperatures that the leaf would attain under maximum (“wet”) and zero (“dry”) transpiration levels respectively, in the same environment (Jones et al., 1997). The requirement for ancillary meteorological data is lowered by using both reference surfaces (Jones, 1999a; Leinonen et al., 2006; Alchanatis et al., 2010).

One of the best known indices for evaluating crop water stress as a function of leaf temperature is the Crop Water Stress Index (CWSI), which expresses the difference between “well-watered baseline” and “total stress” temperatures normalized against vapor pressure deficit (Idso et al., 1981; Jackson et al., 1981; Jones et al., 2002). CWSI has been used for assessing the water status of crops such as grapevine (Grant et al., 2007; Zia et al., 2009), French beans (Möller et al., 2007), wheat (Gontia and Tiwari, 2008), rice (Jones et al., 2009), maize (Romano et al., 2011) and cotton (Alchanatis et al., 2010).

More recently, other approaches have been developed in attempts to improve the sensitivity of infrared estimation for the evaluation of crop water stress indices. In that sense, the stomatal conductance index (I_G), which is proportional to the leaf conductance to water vapor transfer (Jones, 1999a), was calculated using the temperature thresholds (i.e. the use of dry and wet references) and the obtained mean T_{canopy} . However, the best choice of thermal indices to accurately identify plant water stress is still unclear.

In previous studies, leaf temperatures of non-irrigated (NI) and well-irrigated (WI) plants were used as thresholds to estimate the thermal indices (Grant et al., 2006; Jones et al., 2009; Costa et al., 2012). Here, a previously described approach for the use of reference surfaces was used to provide artificial leaves with open and fully closed stomata (Ochagavía et al., 2011). Briefly, the dry and wet references were respectively composed of a black metal (platinum) and a wick of black cotton, which continuously absorbed distilled water from a small reservoir.

Finally, different techniques have been proposed to minimize the thermal variability within grapevine canopies, such as targeting of the shaded side rather than the sunny side to obtain thermal images (Jones et al., 2002) or taking measurements at a certain time of day (Ochagavía et al., 2011). However, up to now, no consensus in the adequate use of the technique has been attained. Nowadays it is unclear whether images taken in the sunlit or shaded part of row crops, such as grapevines, can provide accurate measurements of plant water status. It is neither obvious which is the best time of the day to apply this technique. Therefore, the major aim of our study is to report an evaluation of the consistency and repeatability of methods of applying thermal imaging for the detection of stomatal

closure of grapevine leaves (i) under different times of the day (morning, midday and afternoon), (ii) on both sides of the canopy (sunlit and shaded) and (iii) under different grapevine water status.

Furthermore, the influence of plant water status on berry temperature was evaluated in this study. A number of studies have correlated grape temperatures with anthocyanin composition and concentration in the berries (Spayd et al., 2002; Tarara et al., 2008). Environmental factors may affect the production, transport, and accumulation of anthocyanins and other flavonoids in grapes in a typical bell-shaped manner, and may improve the final quantities of these compounds only when present at optimal levels (Braidot et al., 2008). Hence, a decrease of anthocyanin biosynthesis may occur when endogenous or exogenous factors, such as water (Esteban et al., 2001; Ojeda et al., 2002; Koundouras et al., 2006) and temperature (Hunter et al., 1995; Bergqvist et al., 2001; Spayd et al., 2002) are limiting or excessive.

For the berry, the emission factor (emissivity) was accurately estimated and used as the global emissivity of the whole berry image. Emissivities of common materials such as plant leaves and fruits were reported in the literature (Hellebrand et al., 2001; Jones et al., 2002; Kaplan, 2007; Bulanon et al., 2008), however the determination of thermal emissivity for grapevine berries has not been previously assessed.

2. Material and methods

2.1. Plant material and experimental conditions

The experiment was conducted in September 2011, at harvest, in a commercial hillside vineyard located in Ollauri (Northwestern La Rioja, 42°31'48"N, 2°49'25"W, 527 m), Spain.

Seventeen-year-old grapevines (*Vitis vinifera* L. cv. Graciano) grafted on Richter-110 rootstock were used in the study. The vines were spur-pruned (12 buds per vine) on a bilateral cordon, and grew in a loam to clay-loam soil with 2.7×1.15 m spacing between rows and plants, respectively, and row orientation was east-west. The training system was a vertically shoot positioned (VSP) with movable wires.

The experiment was carried out in two different slope dependent plots of the vineyard (approximately steeper than 10%). Each plot was composed of two rows running up the slope, thus taking advantage of the gradient to obtain different plant water status. In each plot, 10 vines along the two rows were randomly selected, marked and used as experimental plants. During the experiment, some indicators of vine water status, based on leaf maximum daily stomatal conductance (g_s) and stem water potential (Ψ_{stem}) were monitored, bearing in mind the positive correlation obtained by several authors (Stevens et al., 1995; Williams and Araujo, 2002) between those parameters and the soil water availability.

In each vine, and for each side of the canopy (sunlit and shaded sides), one fully expanded leaf was marked to be measured along the experiment (in days 26, 27 and 28 of September). Experimental measurements were evenly distributed between north (shaded part of the canopy) and south (sunlit or exposed to direct solar radiation) sides of the canopies, and were performed at three different times of the day: in the morning (between 10:00 and 12:00 h), at midday (between 13:00 and 15:00 h) and in the afternoon (between 16:00 and 18:00 h). The same marked vines and leaves were measured on each day. No irrigation was applied during the whole season.

Meteorological data on air temperature, relative humidity, solar radiation, wind speed and potential evapotranspiration (ET_0), were provided by an automatic meteorological station (Siar, La Rioja Government) located in Casalareina, 8.2 km distance from the vineyard. Vapor pressure deficit (VPD) was calculated from measurements of

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