Thermal infrared imaging of the temporal variability in stomatal conductance for fruit trees

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ABSTRACT

Repeated measurements using thermal infrared remote sensing were used to characterize the change in canopy temperature over time and factors that influenced this change on 'Conference' pear trees (*Pyrus communis* L.). Three different types of sensors were used, a leaf porometer to measure leaf stomatal conductance, a thermal infrared camera to measure the canopy temperature and a meteorological sensor to measure weather variables. Stomatal conductance of water stressed pear was significantly lower than in the control group 9 days after stress began. This decrease in stomatal conductance reduced transpiration, reducing evaporative cooling that increased canopy temperature. Using thermal infrared imaging with wavelengths between 7.5 and 13 μm, the first significant difference was measured 18 days after stress began. A second order derivative described the average rate of change of the difference between the stress treatment and control group. The average rate of change for stomatal conductance was 0.06 (mmol m^{-2} s^{-1}) and for canopy temperature was -0.04 ($^\circ$C) with respect to days. Thermal infrared remote sensing and data analysis presented in this study demonstrated that the differences in canopy temperatures between the water stress and control treatment due to stomata regulation can be validated.

1. Introduction

Water stress is the most damaging of all environmental stresses to agricultural productivity (Lambers et al., 2008). For this study, we defined water stress as the lack of adequate precipitation combined with high moisture demand needed for normal plant growth and development to complete a plant's life cycle (Mishra and Cherkauer, 2010). In fruit trees, the effect of water stress on fruit production is highly dependent upon its intensity, duration and phenological stage during the occurrence of cell division, vegetative growth fruit or fruit filling. Severe water stress may for example induce biennial bearing, a phenomenon whereby large fruit production occurs one year, alternated by a lesser amount of fruit production the following year (Tromp et al., 2005). Plants acquire water from the soil and CO_2 from the atmosphere to support photosynthesis. This is achieved by CO_2 absorption through the stomatal pores where water is simultaneously transpired. Stomata generally respond to three key mechanisms: (1) the direct response of stomata to meteorological factors including vapor pressure deficit (VPD), ambient air temperature, solar radiation, and wind speed (Hetherington and Woodward, 2003), (2) a response to soil water deficit primarily through the hormone abscisic acid (Ludlow, 1980; Farquhar and Sharkey, 1982; Schulze, 1982, 1986), and (3) changes in mesophyll water status caused by an electrical signal (Grams et al., 2007). Stomatal closure leads to a reduction in CO_2 uptake and transpiration causing an increase in plant temperature (Monteith, 1995). As a result, rising canopy temperatures are an indicator of lower water status in the plant.

The sensitivity of stomatal conductance to VPD is species dependent. VPD is the measure of difference in the amount of water

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vapor in the air at a constant temperature and the amount of water vapor saturated air can hold. In woody species such as fruit trees, an increase in VPD reduced stomatal conductance (Mediavilla and Escudero, 2003). Stomatal response to VPD is not consistent across all woody species (Bunce, 2006). Water stress in Asian pears 'Hosui' (*Pyrus serotina* Rehder var. culta Rehder) without soil moisture stress, stomatal response closely followed the pattern of incoming solar radiation (Renquist et al., 1994). Similar results were discovered by in cassava (*Mannihot esculenta* Crantz) grown with soil moisture approaching field capacity where it was determined that stomatal conductance was reduced as VPD increased (Sharkawy, 1990). The Mediterranean species *Eucalyptus marginata*, showed stomatal conductance was more responsive to soil moisture than leaf water potential (Crombie et al., 1987). In contrast, Scots Pine (*Pinus sylvestris* L.) stomata were most influenced by VPD (Pereira et al., 1987). In apple trees (*Malus domestica* Borkh.), stomatal conductance was lower for shoots that yield fruit than those without fruit (Warrit et al., 1980). In addition to VPD that affects stomatal conductance, a vertical gradient exists within the canopy (Wilson et al., 2000). Also, stomatal response to VPD may be different for leaves in the upper versus lower part of the canopy (Streck et al., 2003). Those differences are dependent upon leaf location and their distinct microclimate that are causing the variability in leaf anatomy and stomatal conductance (Terashima et al., 2006). Even though stomatal conductance is a very good proxy for the water status of a tree, when stomatal conductances are measured with a leaf porometer, the number of leaves per tree needed to be approximately 150 to estimate the mean to be within 10% of the mean value of the canopy (Leverenz et al., 1982). A more efficient technique is thus needed to estimate plant water status.

Thermal imaging has been successful as an integrative technique to measure water status due to the sensitivity of the canopy temperature to changes in stomatal conductance (Jones et al., 2009). However, there are challenges when using thermal imaging such as the measured temperature of the plant is affected not only by stomatal conductance and environmental variables, but also by reflected heat and objects other than leaves (e.g., background, trunk and branches of the canopy). Different methods including thresholding have been used to calibrate thermal imagery and derive leaf temperatures. Thresholding is pixel classification that considers the intensity of the pixels (Otsu, 1979). Many factors complicate thresholding including ambient light, variation of pixel values within the canopy and the background, insufficient contrast, shape and size of the target that is disproportionate with the surroundings (Parker, 1991). Wet and dry reference leaves may be used in place of empirical leaf temperatures where only partial meteorological variables are available (Jones, 1999). The wet/dry leaves allow estimation of canopy temperatures for the current environmental conditions, if one can maintain consistent leaf wetness. When reference leaves are not used, precise environmental measurements including temperature, incoming solar radiation, wind speed and VPD allow for canopy temperature correction (Jones, 2004).

Thermal imaging has been shown to measure canopy temperature variability between canopies and can be repeatedly collected over the same canopy to monitor responses over time (Grant et al., 2007). The variability between treatments can be used as an indicator of water stress (Gardner et al., 1981). However, determining the presence and intensity of water stress through the variability of canopy temperature has been difficult due in part to physiological and imaging constraints (Moller et al., 2007). Because environmental conditions are dynamic, using the average canopy temperature to determine the water status is not recommended as many abiotic factors influence canopy temperature (Grant et al., 2007). The diurnal variability, in canopy temperatures of almond trees (*Prunus dulci* (Mill.) D.A.Webb, cv. Non Pareil and cv. Monterey) under

varying levels of water stress was imposed by decreasing stomatal conductance (Gonzalez-Dugo et al., 2012).

The time required to measure 150 leaves with one leaf porometer whereby each leaf measurement takes 30 s, would require one-hour-fifteen minutes per canopy. Thermal imaging can provide accurate, precise and rapid canopy measurements enabling one to monitor entire fruit orchards in the same time instead of measuring just a few canopies with a leaf porometer. Many studies have examined the change in stomatal conductance over a period of one or a few days. However, few studies have examined the progress and change in status of stomatal conductance due to physiological and meteorological affects over a period of 60 days.

This study elected to use a controlled environment over a field study in order to regulate water input, to eliminate competition for water and nutrients, to reduce within-field variability, and to allow for precise repeated measurements. A soil water deficit was imposed to affect stomatal conductance and to separate the stress treatments from the control group. Meteorological variables and their affect in regulating water status in Conference pear trees is not well understood (Bunce, 2006). Therefore, they were included in the models in order to measure their influence on canopy water status. Because stomatal conductance is well researched and known as an early indicator of water status, leaf porometer data were used as a reference for comparison purposes. Models with residual covariance structures that correct for the repeated nature of the data were applied to analyze differences in water status using stomatal conductance and thermal infrared temperature measurements. There are three specific objectives of this study: (1) analyze stomatal conductance and canopy temperature for the stress treatment and control group at each time point, (2) construct multivariate analysis of time contrast variables for stomatal conductance and canopy temperature corrected for meteorological factors, and (3) estimate the average rate of change in stomatal conductance and canopy temperature between the stress treatments and control group.

2. Experimental

2.1. Experimental design

Stomatal conductance and thermal infrared measurements were taken at the KU Leuven, Leuven, Belgium (50°51′N, 4°40′E; 60 m above sea level). The experimental design was a completely randomized design consisting of 903-year old Conference pear trees planted in individual containers (24 cm × 29 cm). Three containers were placed in a triangular pattern forming in total 30 canopies. The stress treatment consisted of 18 canopies and the control group of 12 canopies. The experiment was laid out with 5 rows containing 6 canopies with spacing of 1.3 m × 3.0 m. Fig. 1a provides a view of the experimental design. A border row surrounded the experiment to minimize border effects. The growing medium was a greenhouse mixture containing of 40% peat moss, 30% perlite and 30% vermiculite. Water stress was initiated during the period of tree growth and development where minimal fruit growth occurs (Anderson, 1936). Beginning March 20, 2012 when approximately 50% of the buds emerged until June 10, 2012, the time water stress was imposed; all control trees were provided with water and nutrients to the water holding capacity of the media in the containers. The period of water stress ended July 27, 2012, 47 days after stress began. In Belgium, the three basic growth and development periods for pear trees are approximately: March through May where fruit cell division takes place (full irrigation for all trees), June through July when the most vigorous vegetative growth occurs (water stress), and August through the first half of September when fruit filling is most dominant (full irrigation for all trees). The containers were covered with a solid waterproof