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Methods paper

Using foliar spectral properties to assess the effects of drought on plant water potential

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Drought frequency is predicted to increase in future environments. Leaf water potential (Ψ_{LW}) is commonly used to evaluate plant water status, but traditional measurements can be logistically difficult and require destructive sampling. We used reflectance spectroscopy to characterize variation in Ψ_{LW} of *Quercus oleoides* Schltdl. & Cham. under differential water availability and tested the ability to predict pre-dawn Ψ_{LW} (PDΨ_{LW}) using spectral data collected hours after pressure chamber measurements on dark-acclimated leaves. Ψ_{LW} was measured with a Scholander pressure chamber. Leaf reflectance was collected at one or both of two time points: immediately (Ψ_{LW}) and ~5 h after pressure chamber measurements (PDΨ_{LW}). Predictive models were constructed using partial least-squares regression. Model performance was evaluated using coefficient of determination (R^2), root-mean-square error (RMSE), bias, and the percent RMSE of the data range (%RMSE). Ψ_{LW} and PDΨ_{LW} were well predicted using spectroscopic models and successfully estimated a wide variation in Ψ_{LW} (light- or dark-acclimated leaves) as well as PDΨ_{LW} (dark-acclimated leaves only). Mean Ψ_{LW} R^2, RMSE and bias values were 0.65, 0.51 MPa and 0.09, respectively, with a %RMSE between 8% and 20%, while mean PDΨ_{LW} R^2, RMSE and bias values were 0.60, 0.44 MPa and 0.01, respectively, with a %RMSE between 9% and 20%. Estimates of PDΨ_{LW} produced similar statistical outcomes when analyzing treatment effects on PDΨ_{LW} as those found using reference pressure chamber measurements. These findings highlight a promising approach to evaluate plant responses to environmental change by providing rapid measurements that can be used to estimate plant water status as well as demonstrating that spectroscopic measurements can be used as a surrogate for standard, reference measurements in a statistical framework.

Keywords: drought, leaf water potential, partial least-squares regression, *Quercus oleoides*, reflectance spectroscopy, water content.

Introduction

Climate change is predicted to increase the frequency of extreme weather conditions (IPCC 2013). One of the most critical environmental stressors currently affecting plant metabolism is drought, which adversely affects growth, development and longevity (Hasanuzzaman et al. 2014). Changes in the severity, frequency and duration of drought events will likely have significant impacts on hydrological cycling, ultimately affecting plant productivity and mortality (Allen et al. 2010, Nam et al. 2015, Cotrozzi et al. 2016). While the effects of drought on plants are

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now gaining considerable attention in natural, agricultural and urban systems, advancements in techniques to monitor the effects of drought on plants are required to improve management practices under predicted changes in climate (Hasanuzzaman et al. 2014, Sjöman et al. 2015).

One of the most frequently used indicators of the effects of drought on plants is the measurement of plant water status. A preferred methodology for determining plant water status is the Scholander pressure chamber (Scholander et al. 1965), which measures leaf water potential (Ψ_{LW}, Santesteban et al. 2011, Pellegrini et al. 2015). While Ψ_{LW} has been used successfully to understand plant water relations, there is a clear trend in research studies towards the collection of pre-dawn leaf water (PDΨ_{LW}), when the plant is in relative equilibrium with the soil in terms of water status (Santesteban et al. 2011). If plants are in equilibrium with the soil pre-dawn, then PDΨ_{LW} can be a reasonable estimate of soil water availability. Pre-dawn leaf water also is a measure of maximum water status of the plant and thus influences plant–water relation parameters, leaf transpiration rates, daily minimum stomatal resistance, leaf expansion and photosynthesis (Sato et al. 2006).

Traditional measurements of Ψ_{LW} and PDΨ_{LW} are destructive, negating the possibility of making time-integrated measurements, and point-based, involving single leaves or shoots, thus limiting the ability to up-scale to generalize regional leaf water status (Peñuelas et al. 1993). In addition, most techniques for measuring leaf or plant hydraulic conductance require measurements of Ψ_{LW} (Bartlett et al. 2012), but often this is done on experimental seedlings that have a limited number of leaves (Brodribb and Holbrook 2006, Franks 2006, Cavender-Bares et al. 2007, Blackman et al. 2009). This approach has several limitations, including potentially altering plant function through the removal of the necessary number of leaves required for the measurements. Similarly, when measuring leaf hydraulic conductance, there is often the need to measure the flow of water through one leaf while using a paired leaf, assumed to be at the same water potential, to measure water potential destructively in a pressure chamber (e.g., Sack and Scoffoni 2012). Thus, the possibility of measuring Ψ_{LW} non-destructively would eliminate the need to remove leaf material, allowing temporal analyses, limiting the impact of the measurements on plant function and potentially improving measurement accuracy for many applications, including leaf hydraulic measurements.

In this paper, we address the possibility of using foliar spectral measurements to estimate water potential. Recent advances in methods utilizing hyperspectral data have enabled estimation of a variety of plant traits and physiological processes based on foliar optical properties, including concentrations of nutrients, structure and secondary metabolites (Petisco et al. 2006, Asner and Martin 2008, Kleinebecker et al. 2009, Asner et al. 2011, Couture et al. 2013, 2016, Serbin et al. 2015) and plant physiological status (Gamon et al. 1992, 1995, Serbin et al. 2012). Plant traits are modeled as a function of reflectance features of leaves in the visible, near-infrared (NIR) and short-wave infrared (SWIR) spectral regions. The model calibration is accomplished by pairing spectra with reliable biochemical, structural and physiological measurements and then the model is validated by comparing relationships between withheld and predicted values. This calibration model can then be used to predict the variable of interest in unknown samples on the basis of their spectral signature alone (Couture and Lindroth 2012). Importantly, measurements of leaf optical properties are rapid, taking only seconds, non-destructive, and relatively inexpensive and therefore can be collected in the field on a large number of individual plants over multiple time periods (Couture et al. 2013, Ainsworth et al. 2014).

In the last few decades, plant water status has been the focus of a number of studies utilizing foliar optical properties (e.g., Hunt et al. 1987, Gamon et al. 1992, Peñuelas et al. 1993, Gao 1996, Gamon et al. 1997, Ceccato et al. 2001, 2002*a*, 2002*b*, Sims and Gamon 2003, Zarco-Tejada et al. 2003, Stimson et al. 2005, Cheng et al. 2008, González-Fernández et al. 2015). These studies have primarily focused on foliar water content or changes in water content. To date, few studies (Rodríguez-Pérez et al. 2007, Santos and Kaye 2009, De Bei et al. 2011, Gallo et al. 2014, Rapaport et al. 2015) have examined the ability of reflectance spectroscopy to estimate Ψ_{LW}. These previous efforts estimating Ψ_{LW}, however, have been limited to grapevine with minimal variation in plant water stress (Ψ_{LW}:−0.9 to −2.3 MPa, De Bei et al. 2011), and none of these studies examined the ability of leaf optical properties to model PDΨ_{LW} as a function of spectral measurements.

Here, we present the results of a study exploring the relationships among foliar optical properties and Ψ_{LW} in live oak (*Quercus oleoides* Schltdl. & Cham.) trees from four countries in Central America grown under varying levels of drought. Specifically, we assessed if plant water status (i.e., Ψ_{LW}) could be effectively estimated using full-range (350–2500 nm) reflectance spectroscopy. Our objectives in this study were (i) to develop spectroscopic models for instantaneous estimation of Ψ_{LW} and (ii) to assess the possibility of estimating PDΨ_{LW} on dark-acclimated leaves, collected pre-dawn and stored in a manner that preserves PDΨ_{LW}, using subsequent spectral measurements.

Materials and methods

Experimental design

Experimental activities were conducted in the field-station of Zamorano University, Honduras (14°00′N, 87°00′W, 797 m). This region is characterized by a seasonally dry tropical climate with a dry season that extends from approximately early January to early May. Live oak acorns from individual mothers from four regions of Central America (Costa Rica, Honduras, Belize and Mexico) were collected, germinated in a nursery under

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protected conditions for 1 year, then subsequently transferred into the ground in one of three years (2010, 2011 or 2012) in two gardens, containing 12 blocks each. In a fully factorial and randomized block design, each block was assigned one of four watering treatments: (i) no watering; (ii) water added during the dry season to match that of the climate in upland Costa Rica (18 mm water per week) (dry season irrigation); (iii) up to 25 mm was added in the wet season during weeks where precipitation dropped below 25 mm (wet season irrigation); and (iv) water added during both wet and dry seasons as reported above (year-round irrigation). These different treatments created a range of water potential values (pre-dawn and midday) used in this study (Ramírez-Valiente et al. 2017). All measurements used in the current study were collected in March 2014.

Estimation of Ψ_{LW}

Leaf water potential was measured at pre-dawn (leaves collected between 05:20 and 09:00 h) or midday (13:30–15:20 h) by a Scholander-type pressure chamber (Soil Moisture Equipment Co., Santa Barbara, CA, USA). Measurements were conducted on one fully expanded leaf per tree (423 trees). Fully expanded leaves were collected across various developmental stages and distributed among the experimental drought treatments and countries. Fifty percent of the sampled leaves used in the current study were from seeds originating in Costa Rica, 23% were from seeds originating in Honduras, 17% from Belize and 10% were from Mexico.

Collection of leaf spectra

Full-range (350–2500 nm) reflectance profiles of live oak leaves were collected using an ASD FieldSpec 3 spectroradiometer (Analytical Spectral Devices, Boulder, CO, USA), starting immediately after the pressure chamber measurement. Measurements were made on the adaxial surface of the same leaves assessed for Ψ_{LW} using a leaf-clip assembly attached to a plant probe with an internal halogen light source. Reflectance was measured on three areas per leaf, with five measurements per area, and all measurements were combined to produce an average leaf spectrum. The relative reflectance of each leaf was determined from the measurement of leaf radiance divided by the radiance of a white reference (Spectralon, Labsphere Inc., North Dutton, NH, USA), measured every fourth spectral collection.

Spectral measurements were collected throughout the day immediately (<5 min) after pressure-chamber measurements for 206 samples (49 midday Ψ_{LW} and 157 from PDΨ_{LW}). Immediately following foliar collections, intact leaves were wrapped with aluminum foil, sealed in plastic bags and placed in a cooler in the dark. A small piece of moist paper towel was kept in the bag without touching the leaf to increase humidity and reduce transpiration to near zero. This procedure is a commonly used practice to prevent changes in leaf water potential after collection. An additional 217 leaf samples were bagged following pressure-chamber measurements, sealed and stored in a dark cooler, as before to preserve Ψ_{LW}. Spectral measurements were conducted on these ~5 h (range, 4–6.5 h) after pre-dawn pressure chamber measurements. In a subset of the leaves, we confirmed that Ψ_{LW} values were stable and did not change under the storage conditions (J.C.-B., unpublished data). We collected foliar spectral measurements both immediately and ~5 h following pressure chamber measurements on a total of 89 plants. These two sets of measurements allowed us to determine if (i) we could model Ψ_{LW} instantaneously, regardless of whether the reference and spectral measurements were made pre-dawn or midday and (ii) we could model PDΨ_{LW} as a function of spectral measurements made a posteriori, i.e., several hours after the pressure chamber measurements, on leaves that were stored to maintain dark-acclimation and constant water potential. The dark-acclimated test also simulated potential measurement conditions using spectroscopy, in which samples are often collected at one time but may not be measured for several hours due to logistics.

Collection of osmolyte standard spectra

We collected reflectance profiles of osmolyte standards following Couture et al. (2016). Briefly, spectral measurements were collected from 300 mg of laboratory grade, powdered glucose, sucrose, fructose and malic, citric and oxalic acid (Fisher or Sigma Aldrich, St. Louis, MO, USA) by using a plant probe attached to a Dremel® drill press, to maintain constant measurement pressure and geometry. Material from each standard was packed loosely machined aluminum sample cup painted flat matte black and placed underneath the vertically mounted plant probe. Each measurement was compressed uniformly to 2 N m^{-1} during spectral collection through the use of a precision torque wrench (Effetto Mariposa, Giustaforza™ Professional) that acted as the handle for the drill press. Nine spectral measurements were collected for each sample in three sets of three measurements; the material was mixed and repacked in the sampling cup and the sample cup rotated by 90° between each set of collections. These steps were conducted to reduce any potential bias from sample cup orientation or sensor characteristics. All nine spectra were then averaged to determine the mean dry material reflectance and subsequently converted to first-derivative reflectance to highlight spectral absorption features.

Statistical modeling of Ψ_{LW} and PDΨ_{LW} using reflectance spectroscopy

We modeled the variation in Ψ_{LW} and PDΨ_{LW}, based on coincident variation in untransformed foliar spectral properties, using jackknifed, partial least-squares regression (PLSR; Serbin et al. 2012, Couture et al. 2013, 2016). When predictor variables are highly correlated and cases where predictor variables outnumber observations, as with hyperspectral data, standard regression approaches can produce unreliable coefficients as correlated predictor variables lead to bias in beta coefficients

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and error estimates (Grossman et al. 1996). In contrast to classical regression techniques, PLSR reduces a large number of collinear predictor variables into relatively few, uncorrelated latent variables and has become the standard in chemometric approaches (Wold et al. 1984, 2001). While it is possible to use as many latent variables as predictors, doing so may lead to overfitting the model. To avoid potentially overfitting the model, we optimized the number of components included in the model by minimizing the prediction residual error sum of squares (PRESS) statistic (Chen et al. 2004). We calculated the PRESS statistic iteratively, through a sequential leave-one-out cross-validation procedure. Once minimized, the final set of extracted components was combined into a linear model predicting Ψ_{LW} or PDΨ_{LW} based on leaf spectral profiles.

We developed a preliminary model to identify poorly predicted outliers where the ratio of the standard deviation to the mean of the prediction were on average greater than 30% (Couture et al. 2016). Outliers were a result of spectral and/or reference measurement errors, detectable from elevated reflectance in the visible wavelengths that occurs when the leaf clip is not fully closed on either the reference or target measurement. Outliers removed accounted for 10% and 11% of the initial data for the Ψ_{LW} and PDΨ_{LW} measurements. Performance of the final models predicting Ψ_{LW} and PDΨ_{L} was evaluated using 80/20 split of the data for calibration/validation, respectively, over 500 randomized permutations of the dataset. For each such permutation, we tracked the model fit (R^2), overall error rate (RMSE, root-mean-square error), the percentage of error over the data range (%RMSE) and bias in the external validation dataset. These analyses generated a distribution of model coefficients and fit statistics and allowed for the assessment of model stability as well as uncertainty in predictions.

We further determined the contribution of individual wavelengths to the model performance using the variable important to the projection (VIP) selection statistic (Chong and Jun 2005). The VIP metric evaluates the importance of individual wavelengths in explaining the variation in both the response and predictor variables; larger weightings confer greater value of contribution by specific wavelengths to the predictive model (Wold et al. 2001, Chong and Jun 2005). Models were built using the packages *caret* and *vegan* in R (www.r-project.org).

We examined relationships among leaf water potential values and other foliar chemical concentrations (nitrogen, carbon, fiber and lignin), morphology (leaf mass per area, LMA) and vegetation indexes (normalized differential water index, NDWI; chlorophyll estimation; photochemical reflectance index, PRI) using Pearson correlations to determine the influence of other spectrally derived leaf characteristics with predictions of leaf water potential predictions. We also examined relationships between immediate and a posterori Ψ_{LW} using Pearson correlations. Nitrogen, carbon, fiber, lignin and LMA were generated from spectra using calibrations from Serbin (2012); NDWI (a measure of foliar water content) was determined as the relative difference in reflectance at wavelengths 857 and 1241 (Gao 1996); chlorophyll was determined as the relative difference between reflectance at wavelengths 750 and 705 (Gitelson and Merzlyak 1994) and PRI (a measure of photosynthetic light-use efficiency and plant stress) was calculated as the relative difference between reflectance at wavelengths 570 and 531 (Gamon et al. 1997).

We were interested in whether the data collected using spectroscopy could be used as a surrogate for the reference data to determine the effect of the different conditions of water availability on PDΨ_{LW}. To test this question, we used one-way analyses of variance (ANOVA) separately for both reference and predicted data using water treatment as the main effect. We excluded country from this analysis as there were not enough samples from Mexican plants irrigated only during the dry season to complete the full, two-way ANOVA (water availability treatment × country). Correlation and ANOVA analyses were performed in JMP 10.0 (SAS Institute Inc., Cary, NC, USA).

Results

Estimating Ψ_{LW} from leaf reflectance spectra

We initially examined numerous models containing multiple different wavelength ranges and components to optimize model performance (see Table S1 available as Supplementary Data at *Tree Physiology* Online). Final models for Ψ_{LW} utilized the wavelength range 950–2400 nm and included 16 components. Elimination of the visible bands (and some of the near-infrared) reduces the likelihood of relationships between water potential and spectra due to correlations between pigments/greenness and water status. Predictive models accurately characterized Ψ_{LW} in *Q. oleoides*, with average R^2, RMSE and bias values of 0.65, 0.51 MPa and 0.09, respectively (Figure 1a–c). The distribution ranges for R^2, RMSE and bias also suggest that the model predicting Ψ_{LW} was relatively stable, as 97% of the models produced a high R^2 (0.50–0.87), RMSE values (range, 0.32–0.70 MPa) that had a %RMSE between 8% and 20% of the data range, and 85% of the models showed minimal bias (range, −2.49 and 1.04). The range of Ψ_{LW} reported in the plants used in this study determined using the Scholander pressure chamber ranged from −3.7 to −0.1 MPa, and was closely matched by the range of Ψ_{LW} predicted via spectroscopy: −3.5 to 0.0 MPa (Figure 1d). Standardized coefficients of specific wavelength regions used in the prediction of Ψ_{LW} varied around the 950–2400 nm spectrum, although consistently positive or negative coefficients we found in water absorption regions (Figure 2a). The VIP statistics confirmed that a number of wavelength regions with the highest contributions to the model were between 950 and 1010 nm, 1370 and 1580 nm, 1650 and 1685 nm, 1860 and 2040 nm, 2070 and 2140 nm, and 2240 and 2400 nm regions (Figure 2c). Wavelengths strongly related

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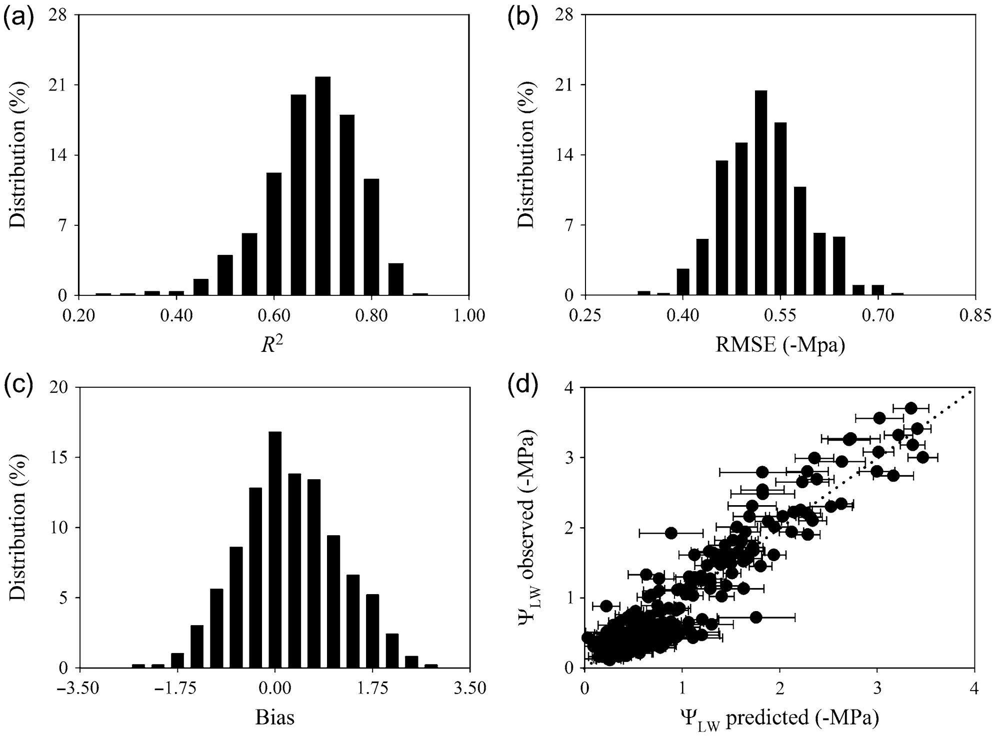

Figure 1. Error distributions of (a) R^2, (b) root-mean-square error (RMSE) and (c) bias for validation data generated using 500 random permutations of the data with 80% used for calibration and 20% used for validation to predict Ψ_{LW} values from foliar spectra from live oak. (d) Predicted vs observed values of Ψ_{LW} of live oak. Dotted line is 1:1 relationship.

with standardized coefficients and VIP values overlapped with prominent spectral features collected from the reflectance profiles of standards of fructose, glucose, fructose, citric acid, malic acid and oxalic acid (Figure 2; see Figure S1 available as Supplementary Data at *Tree Physiology* Online). The PRI and NDWI had the strongest positive and negative relationships, respectively, with instantaneous retrievals of Ψ_{LW} (Table 1).

Estimating PDΨ_{LW} from dark-acclimated leaves using leaf reflectance spectra

Numerous PLSR models containing different wavelength ranges and components were also tested to estimate PDΨ_{LW} on dark-acclimated leaves (see Table S1 available as Supplementary Data at *Tree Physiology* Online). Final models of predictions of PDΨ_{LW} used the wavelength range 1400–2400 nm and included 14 components. Models estimated PDΨ_{LW} with relatively high accuracy and precision in *Q. oleoides*: average R^2, RMSE and bias values were 0.60, 0.44 MPa and 0.01, respectively (Figure 3a–c). Similar to models of Ψ_{LW}, models estimating PDΨ_{LW} models were stable, with 95% of the models having a R^2 ranging between 0.50 and 0.79, RMSE values (range, 0.27–0.59 MPa) with a %RMSE between 9% and 20% of the data range, and 83% of the models showed minimal bias (range, −2.45 to 1.05). Again, similar to models predicting Ψ_{LW}, the observed values (−3.0 to −0.1 MPa, determined using Scholander pressure chamber) and predicted PDΨ_{LW} values were comparable (Figure 3d). Standardized coefficients showed strong relationships with two water absorption features centered at 1940 and 2400 nm, as well as the 1640–1660 nm region (Figure 2b). The VIP statistics confirmed the importance of the 1400–1470 nm, 1655–1675 nm and 1840–1950 nm regions, as well as features in the 2125–2400 nm range, with specific peaks occurring at 2181, 2248 and 2375 nm (Figure 2c). Similar to instantaneous Ψ_{LW} predictions, wavelengths strongly related with standardized coefficients and VIP values again overlapped with prominent spectral features collected from the reflectance profiles of standards of fructose, glucose, fructose, and citric, malic and oxalic acids (Figure 2; see Figure S1 available as Supplementary Data at *Tree Physiology* Online). Similar to instantaneous Ψ_{LW} predictions, NDWI and PRI had the strongest positive and negative relationships, respectively, with field measurements of PDΨ_{LW} (Table 1). While statistically significant, the strength of the relationships of NDWI and PRI with PDΨ_{LW} was much less than the relationships of those of indexes with Ψ_{LW}. The relationship between PDΨ_{LW} values predicted instantaneously and those predicted a posteriori demonstrated a statistically significantly, positive linear correlation (Figure 4). In addition, ANOVA of treatment effects on PDΨ_{LW} yielded similar statistical results when using either the pressure chamber collected data or the spectroscopy predicted data (Table 2).

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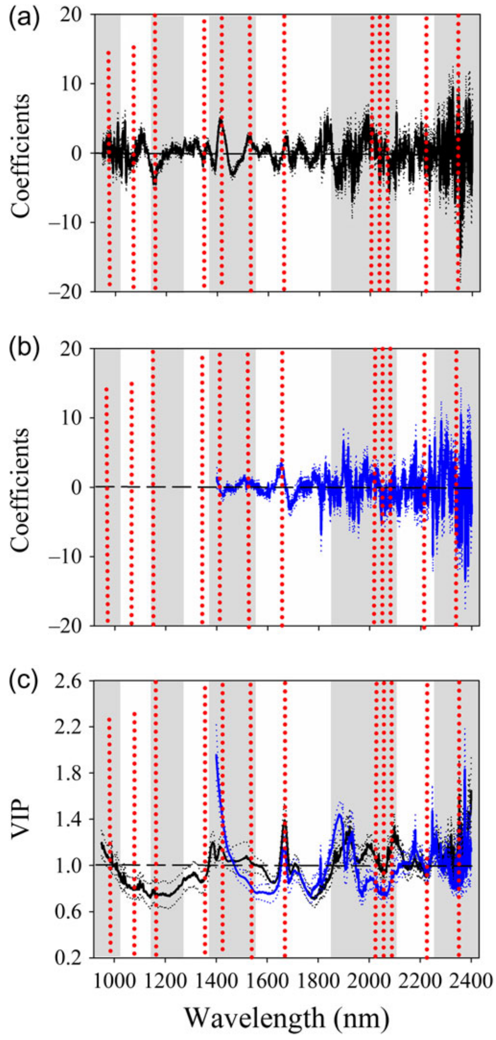

Figure 2. Mean (solid), 5th and 95th percentile (dotted) of Ψ_{LW} (a, black) and PDΨ_{LW} (b, blue) standardized model coefficients and (c) variable important to the projection (VIP) selection values by wavelengths. Gray vertical bars represent the main absorption water features of the spectrum. Red vertical dotted lines represent prominent optical features of common foliar osmolytes (see Figure S1 available as Supplementary Data at *Tree Physiology* Online).

Discussion

Measuring Ψ_{LW} using a pressure chamber is currently the most widely used method to estimate the water status of plants exposed to drought (González-Fernández et al. 2015). However, this method has several limitations: it is destructive, user-dependent and point based (Peñuelas et al. 1993, Santos and Kaye 2009). In this study, we showed that live oak Ψ_{LW} and PDΨ_{LW} can be reliably estimated using NIR-SWIR spectroscopy with appropriate PLSR calibrations. To our knowledge, our study is the first to have explored the potential to estimate Ψ_{LW} changes using spectroscopy in a tree species, while capturing a wide range (0 to −4 MPa) of ecologically relevant drought stress conditions experienced by plants in a variety of climatic regions, in contrast to the narrower range of soil conditions in managed agricultural settings. In addition, this study is the first to predict PDΨ_{LW}, instantaneously or a posteriori from dark-acclimated leaves, in any species.

Table 1. Correlation matrix of foliar traits predicted from spectra with immediate predictions of leaf water potential (Ψ_{LW}, $n = 206$) and predictions of pre-dawn leaf water potential (PDΨ_{LW}, $n = 217$) on dark-acclimated leaves. % dm, % dry mass; LMA, leaf mass per area; NDWI, normalized differential water index; PRI, photochemical reflectance index. Significant relationships ($P \leq 0.05$) are in bold.

Trait	Ψ_{LW}	PDΨ_{LW}
Nitrogen (% dm)	−0.12	−0.06
Carbon (% dm)	**−0.16**	−0.04
LMA g m^{-2}	−0.07	−0.03
Fiber (% dm)	**−0.22**	−0.08
Lignin (% dm)	**−0.28**	−0.06
Cellulose (% dm)	**−0.17**	**−0.17**
NDWI	**−0.41**	**−0.19**
Chlorophyll (g m^{-2})	**−0.32**	−0.07
PRI	**0.59**	**0.15**

Our study utilized narrow-band spectroscopy to exploit fundamental relationships among leaf optical properties and plant water status. These approaches can be broadly divided into those based either (i) on the amount of leaf water (e.g., relative water content) or (ii) on leaf energy status, usually measured by the total water potential (Ψ_W, the sum of a number of component potentials: $\Psi_W = \Psi_\pi + \Psi_P + \Psi_g$, where Ψ_π is the osmotic potential due to dissolved solutes, Ψ_P is pressure potential, equal to the hydrostatic pressure and Ψ_g is the gravitational potential reflecting elevation differences between the site of interest and the reference level and, thus, ignorable except in tall trees; Jones 2007). Leaf spectroscopy likely incorporated information from both approaches because reflectance is influenced by (i) the amount of water, as well as by (ii) the composition and concentration of osmolytes that ultimately affect variation in Ψ_{LW}. Changes in osmotic adjustments have been considered an important physiological adaptation associated with plant drought response, including *Q. oleoides* (Ramírez-Valiente and Cavender-Bares 2017), involving the net accumulation of plant cell solutes in response to drops in water potential in the root medium. Solutes such as glycerol, proline, glycebetaine and especially sugars contribute to this process (Silva et al. 2010).

We found that the spectral region best predicting Ψ_{LW} was 950–2400 nm. Similarly, other studies (Santos and Kaye 2009, De Bei et al. 2011) have obtained good calibrations for the prediction of Ψ_{LW} in multiple grapevine varieties using a similar spectral range (1100–2300 nm). Indeed, the NIR-SWIR region of the electromagnetic spectrum contains several wavelengths that are strongly influenced by the presence of water, and water status of leaves (Shenk et al. 2001). As such, it is not surprising that we observed that the most important wavelengths to predicting Ψ_{LW} were in the water absorption ranges, as well as the 1650–1685 nm regions. In addition, two spectrally derived

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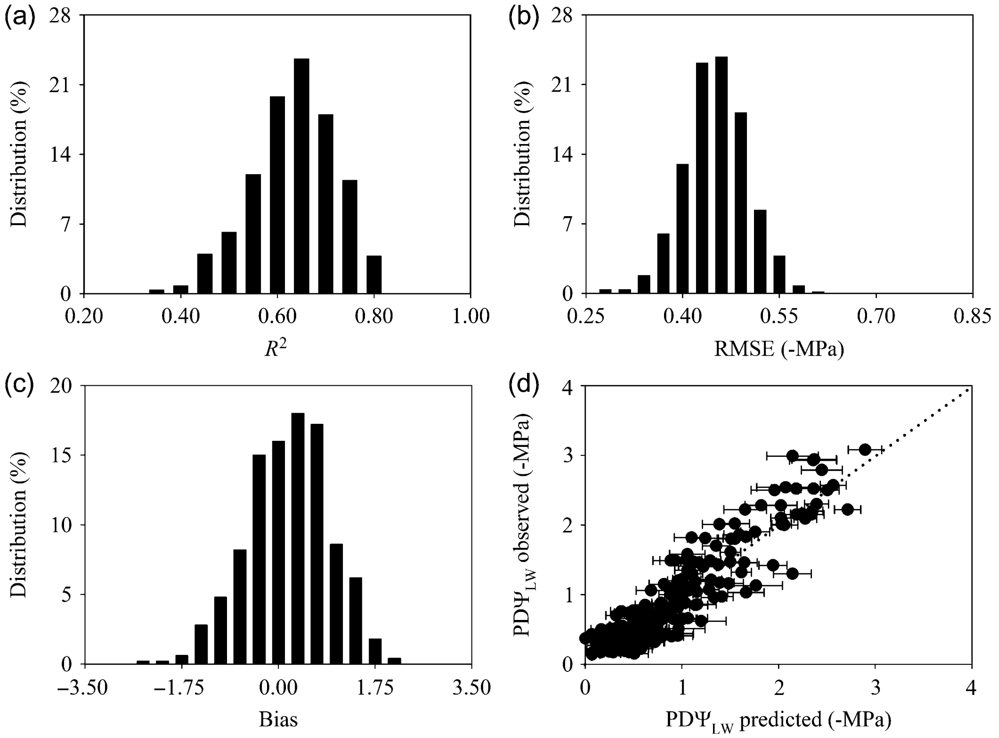

Figure 3. Error distributions of (a) R^2, (b) root-mean-square error (RMSE) and (c) bias for validation data generated via cross-validation using 500 random permutations of the data with 80% used for calibration and 20% used for validation for models predicting PDΨ$_{LW}$ values from foliar spectra collected ~5 h after the reference measurement from live oak. (d) Instantaneous predicted vs a posteriori predicted values of PDΨ$_{LW}$ of live oak. Dotted line is 1:1 relationship.

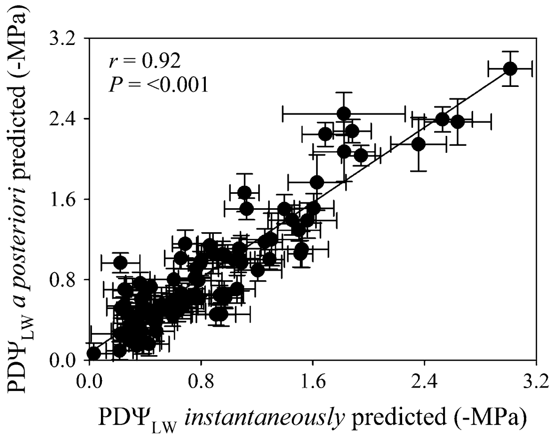

Figure 4. Correlation between PDΨ$_{LW}$ predicted values, generated from models built with the data collected instantaneously after the Scholander pressure chamber measurements, and PDΨ$_{LW}$ predicted values, generated from models built from a posteriori measurements on dark-acclimated leaves in live oak. $N = 89$.

Table 2. *F* and *P* values of the one-way ANOVA examining the effects of the different water availability conditions on PDΨ$_{LW}$ in live oak in both reference (i.e., pressure chamber) measurements and spectroscopy predicted values using dark-acclimated leaves measured stored to preserve PDΨ$_{LW}$ and measured later in the day. df, degrees of freedom.

Treatment	Reference PDΨ$_{LW}$	Spectroscopy predicted PDΨ$_{LW}$
Water availability	df: 3 *F*: 2.98 *P*: <0.05	df: 3 *F*: 3.91 *P*: <0.05

indexes, PRI (commonly used as a measure of plant stress) and NDWI (a measure of leaf water content), had strong positive ($r = 0.59$) and negative ($r = -0.41$) relationships, respectively, with Ψ$_{LW}$. These relationships indicate that as Ψ$_{LW}$ increased (became increasingly negative), plant stress increased and leaf water content decreased. Our findings agree with those of other studies on grapevine reporting that relevant water absorption peaks were influential in prediction of Ψ$_{LW}$ (Santos and Kaye 2009, De Bei et al. 2011, Rapaport et al. 2015) but also highlight that spectroscopy can be used as an effective indicator of plant responses to water stress as indicated explicitly by water potential (Peñuelas et al. 1993).

A novel outcome of this study is our ability to estimate PDΨ$_{LW}$ collected using spectral data collected at a time several hours following the pre-dawn pressure chamber measurements. In contrast to the Ψ$_{LW}$ model we describe above, we found that the most relevant spectral region for predicting PDΨ$_{LW}$ was 1400–2400 nm, suggesting that the spectral features associated with the two models are potentially differentiating physiological conditions from spectral collections associated with measurements only made at pre-dawn and those made over the span of pre-dawn to midday. The spectral region used in the PDΨ$_{LW}$ model is dominated by water content and outside of

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wavelengths commonly associated with pigments and leaf structure (which could potentially confound models). This wavelength region excluded the secondary water absorption features centered at 970 nm and 1200 nm (Carter 1991) and restricted spectrum utilized to the SWIR region. We found that wavelengths in the regions between 1650 and 1680 nm, 1860 and 1950 nm, and over 2200 nm were important for model prediction, potentially due to a higher importance of osmolyte concentration as a driver of the relationships between spectra and pre-dawn water potential values. By eliminating the NIR portion of the spectrum we potentially amplified the contribution of osmolytes in the prediction of water potential in dark-adapted leaves. We also found limited relationships of PDΨ_{LW} with other leaf traits and indexes derived from spectroscopy, compared with relationships in predictions of Ψ_{LW}. Our interpretation that osmolytes influence PDΨ_{LW} predictions is supported in part by findings from multiple studies (Shetty and Gislum 2011, Rubert-Nason et al. 2013, Asner and Martin 2015, Ramirez et al. 2015) reporting that wavelength regions important for predicting non-structural carbohydrate concentrations using spectroscopy align or closely overlap with the wavelength regions of importance we document. Moreover, our theory is bolstered by an overlapping of wavelengths strongly related with standard coefficients and VIP values and prominent spectral features of common foliar osmolytes (see Figure S1 available as Supplementary Data at *Tree Physiology* Online). Osmolytes are important for regulating plant water potential (Dichio et al. 2009), and thus spectral features associated with osmolyte concentrations may potentially assist in the spectroscopic retrievals of instantaneous Ψ_{LW} and PDΨ_{LW}. Future research of reflectance profiles of specific osmolytes will help disentangle the relative contributions of water content and osmotic potential to predictions of leaf water potential.

Importantly, we found a close linear relationship between PDΨ_{LW} values predicted immediately and a posteriori. In addition, PDΨ_{LW} estimates predicted using spectroscopy resulted in a similar statistical outcome as reference data when examining the effects of water availability on plant water status. While other studies (De Bei et al. 2011, Rapaport et al. 2015) have showed qualitative differences in spectra among various drought treatments, our study is the first where drought treatments were analyzed quantitatively using both observed and predicted values, highlighting the potential for implementing this method in ecological studies. The %RMSE for the PDΨ_{LW} model was between 9% and 20% of the data range; the absolute prediction error, however, was greater at low (well-hydrated) rather than high values of PDΨ_{LW} (see Figure S2 available as Supplementary Data at *Tree Physiology* Online), suggesting this approach may have limitations discriminating fine scale differences in leaf water potential, especially in conditions of adequate water availability and low to moderate drought stress. The current model captures a wide range in water potential and targeted models may need to be developed to adequately capture differences in a narrower range of water potential values or species for which a narrower range is meaningful, thus, we encourage caution when interpreting results from a narrow range of low values of PDΨ_{LW}. Regardless of this limitation, here we demonstrate that measurements made several hours after pre-dawn on properly stored leaves can be used as a surrogate for either spectral measurements made immediately or standard pressure chamber measurements determining PDΨ_{LW}, and provide a similar return in statistical analysis as the former methods of data collection.

In summary, we show that a non-invasive method using spectral reflectance can be an alternative method for monitoring Ψ_{LW} and also making a posteriori measurements of PDΨ_{LW} on tropical live oak. Spectroscopic approaches are quick, non-destructive and relatively low cost, providing the possibility to screen more samples in the field and over multiple time periods (Serbin et al. 2012, Couture et al. 2013, Ainsworth et al. 2014). In addition, we have demonstrated that spectroscopic retrievals of PDΨ_{LW} in response to environmental variation (e.g., water availability) can be used as a surrogate for standard approaches and analyzed with the same statistical rigor. The expanding research of relating hyperspectral data to a broad variety of plant biochemical and physiological traits can provide novel insights into the ecology and evolution of plant responses to environmental change, and can do so by rapidly and simultaneously measuring multiple plant characteristics.

Supplementary Data

Supplementary Data for this article are available at *Tree Physiology* Online.

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Conflict of interest

None declared.

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Authors' contribution

J.C.-B., P.A.T. and J.J.C. planned and designed the research objectives of this study. J.C.-B. secured funding and designed the layout for the common gardens at Zamorano and G.P. contributed to the design, maintenance and management of the common gardens at Zamorano. C.C.K. designed the spectral field measurement effort, and C.C.K., J.C.-B. and B.F. collected field data. L.C. and J.J.C. analyzed the data and wrote the manuscript, with input from J.C.-B. and P.A.T. E.P. and C.N. supported L.C. during the writing of this manuscript while L.C. conducted this work as a visiting scholar with P.A.T.

References

Ainsworth EA, Serbin SP, Skoneczka JA, Townsend PA (2014) Using leaf optical properties to detect ozone effects on foliar biochemistry. Photosynth Res 119:65–76.

Allen CD, Macalady AK, Chenchouni H et al. (2010) A global overview of drought and heat-induced tree mortality reveals emerging climate change risks for forests. For Ecol Manage 259:660–684.

Asner GP, Martin RE (2008) Spectral and chemical analysis of tropical forests: scaling from leaf to canopy levels. Remote Sens Environ 112: 3958–3970.

Asner GP, Martin RE (2015) Spectroscopic remote sensing of non-structural carbohydrates in forest canopies. Remote Sens 7: 3526–3547.

Asner GP, Martin RE, Tupayachi R, Emerson R, Marinez P, Sinca F, Powell GVN, Wright SJ, Lugo AE (2011) Taxonomy and remote sensing of leaf mass per area (LMA) in humid tropical forests. Ecol Appl 21: 85–98.

Bartlett MK, Scoffoni C, Sack L (2012) The determinants of leaf turgor loss point and prediction of drought tolerance of species and biomes: a global meta-analysis. Ecol Lett 15:393–405.

Blackman CJ, Borbribb TJ, Jordan GJ (2009) Leaf hydraulics and drought stress: response, recovery and survivorship in four woody temperate plant species. Plant Cell Environ 32:1584–1595.

Brodribb TJ, Holbrook NM (2006) Declining hydraulic efficiency as transpiring leaves dessicate: two types of response. Plant Cell Environ 29: 2205–2215.

Carter GA (1991) Primary and secondary effects of water content on the spectral reflectance of leaves. Am J Bot 78:916–924.

Cavender-Bares J, Sack L, Savage J (2007) Atmospheric and soil drought reduce nocturnal conductance in live oaks. Tree Physiol 27:611–620.

Ceccato P, Flasse S, Tarantola S, Jacquemond S, Grégoire JM (2001) Detecting vegetation leaf water content using reflectance in the optical domain. Remote Sens Environ 77:22–33.

Ceccato P, Gobron N, Flasse S, Pinty B, Tarantola S (2002*a*) Designing a spectral index to estimate vegetation water content from remote sensing data: Part 1. Theoretical approach. Remote Sens Environ 82: 188–197.

Ceccato P, Flasse S, Gregoire JM (2002*b*) Designing a spectral index to estimate vegetation water content from remote sensing data: Part 2. Validation and applications. Remote Sens Environ 82:198–207.

Chen S, Hong X, Harris CJ, Sharkey PM (2004) Spare modeling using orthogonal forest regression with PRESS statistic and regularization. IEEE Trans Syst Man Cybern 34:898–911.

Cheng YB, Ustin SL, Riano D, Vanderbilt VC (2008) Water content estimation from hyperspectral images and MODIS indexes in Southeastern Arizona. Remote Sens Environ 112:363–374.

Chong I-G, Jun C-H (2005) Performance of some variable selection methods when multicollinearity is present. Chemom Intell Lab Syst 78: 103–112.

Cotrozzi L, Remorini D, Pellegrini E, Landi M, Massai R, Nali C, Guidi L, Lorenzini G (2016) Variations in physiological and biochemical traits of oak seedlings grown under drought and ozone stress. Physiol Plant 157:69–84.

Couture JJ, Lindroth RL (2012) Atmospheric change alters performance of an invasive forest insect. Glob Chang Biol 18:3543–3557.

Couture JJ, Serbin SP, Townsend PA (2013) Spectroscopic sensitivity of real-time, rapidly induced phytochemical change in response to damage. New Phytol 198:311–319.

Couture JJ, Singh A, Rubert-Nason KF, Serbin SP, Lindroth RL, Townsend PA (2016) Spectroscopic determination of ecologically relevant secondary metabolites. Methods Ecol Evol 7:1402–1412.

De Bei R, Cozzolino D, Sullivan W, Cynkar W, Fuentes S, Dambergs R, Pech J, Tyerman S (2011) Non-destructive measurement of grapevine water potential using near infrared spectroscopy. Aust J Grape Wine Res 17:62–71.

Dichio B, Margiotta G, Xiloyannis C, Bufo SA, Sofo A, Cataldi TRI (2009) Changes in water status and osmolyte contents in leaves and roots of olive plants (*Olea europea* L.) subjected to water deficit. Trees 23: 247–256.

Franks PJ (2006) Higher rates of leaf gas exchange are associated with higher leaf hydrodynamic pressure gradients. Plant Cell Environ 29: 584–592.

Gallo G, Mincapilli M, Ciraolo G, Provenzano G (2014) Detecting crop water status in mature olive groves using vegetation spectral measurements. Biosyst Eng 128:52–68.

Gamon JA, Peñuelas J, Field CB (1992) A narrow-waveband spectral index that tracks diurnal changes in photosynthetic efficiency. Remote Sens Environ 41:35–44.

Gamon JA, Field CB, Goulden ML, Griffin KL, Hartley AE, Joel G, Peñuelas J, Valentini R (1995) Relationship between NDVI, canopy structure, and photosynthesis in 3 Californian vegetation types. Ecol Appl 5: 28–41.

Gamon JA, Serrano L, Surfus JS (1997) The photochemical reflectance index: an optical indicator of photosynthetic radiation use efficiency across species, functional types, and nutrient levels. Oecologia 112: 492–501.

Gao B-C (1996) NDWI – a normalized difference water index for remote sensing of vegetation liquid water from space. Remote Sens Environ 58:257–266.

Gitelson AA, Merzlyak MN (1994) Spectral reflectance changes associate with autumn senescence of *Aesculus hippocastanum* L. and *Acer platanoides* L. leaves. Spectral features and relation to chlorophyll estimation. J Plant Physiol 143:286–292.

González-Fernández AB, Rodríguez-Pérez JR, Marabel M, Álvarez-Taboada F (2015) Spectroscopic estimation of leaf water content in commercial vineyards using continuum removal and partial least squares regression. Sci Hortic 188:15–22.

Grossman YL, Ustin SL, Jacquemound S, Sanderson EW, Schmuck G, Verdebout J (1996) Critique of stepwise multiple linear regression from the extraction of leaf biochemistry information from leaf reflectance data. Remote Sens Environ 56:182–193.

Hasanuzzaman M, Nahar K, Gill SS, Fujita M (2014) Drought stress responses in plants, oxidative stress, and antioxidant defense. In: Tuteja N, Gill SS (eds) Climate change and plant abiotic stress

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tolerance Vol. 1, 1st edn. Wiley-VCH Verlag GmbH & Co, Weinheim, Germany, pp 209–250.

Hunt ER Jr, Rock BN, Nobel PS (1987) Measurement of leaf relative water content by infrared reflectance. Remote Sens Environ 22: 429–435.

IPCC (2013) Climate change 2013: the physical science basis. Contribution of working group I to the fifth assessment report of the Intergovernmental Panel on Climate Change. Stocker TF, Qin D, Plattner M, Tignor SK, Allen J, Boschung A, Nauels Y, Bex V, Midgley PM (eds) Cambridge University Press, Cambridge, UK; New York, NY.

Jones HG (2007) Monitoring plant and soil water status: established and novel methods revisited and their relevance to studies of drought tolerance. J Exp Bot 58:119–130.

Kleinebecker T, Schmidt SR, Fritz C, Smolders AJP, Holzel N (2009) Prediction of delta ^{13}C and delta ^{15}N in plant tissues with near-infrared reflectance spectroscopy. New Phytol 184:732–739.

Nam W-H, Hayes MJ, Svoboda MD, Tadesse T, Wilhite DA (2015) Drought hazard assessment in the context of climate change for South Korea. Agric Water Manage 160:106–117.

Pellegrini E, Campanella A, Paolocci M, Trivellini A, Gennai C, Muganu M, Nali C, Lorenzini G (2015) Functional leaf traits and diurnal dynamics of photosynthetic parameters predict the behavior of grapevine varieties towards ozone. PLoS One 10:e0135056.

Peñuelas J, Filella I, Biel C, Serrano L, Save R (1993) The reflectance as the 950–970 nm region as an indicator of plant water status. Int J Remote Sens 14:1887–1905.

Petisco C, Garcia-Criado B, Mediavilla S, de Aldana BRV, Zabalgogeazcoa I, Garcia-Ciudad A (2006) Near-infrared reflectance spectroscopy as a fast and non-destructive tool to predict foliar organic constituents of several woody species. Anal Bioanal Chem 386:1823–1833.

Ramirez JA, Posada JM, Handa IT, Hoch G, Vohland M, Messier C, Reu B (2015) Near-infrared spectroscopy (NIRS) predicts non-structural carbohydrate concentrations in different tissue types of a broad range of tree species. Methods Ecol Evol 6:1018–1025.

Ramírez-Valiente JA, Cavender-Bares J (2017) Evolutionary trade-offs between drought resistance mechanisms across a precipitation gradient in a seasonally dry tropical oak (*Quercus oleoides*). Tree Physiol 37:902.

Ramírez-Valiente JA, Center A, Sparks JP, Sparks KL, Etterson JR, Longwell T, Pilz G, Cavender-Bares J (2017) Population-level differentiation in growth rates and leaf traits in seedlings of the neotropical live oak *Quercus oleoides* grown under natural and manipulated precipitation regimes. Front Plant Sci 8:585.

Rapaport T, Hochberg U, Shoshany M, Karnieli A, Rachmilevitch S (2015) Combining leaf physiology, hyperspectral imaging and partial least squares-regression (PLS-R) for grapevine water status assessment. ISPRS J Photogramm Remote Sens 109:88–97.

Rodríguez-Pérez JR, Riaño D, Carlisle E, Ustin S, Smart DR (2007) Evaluation of hyperspectral reflectance indices to detect grapevine water status in vineyards. Am J Enol Viticult 58:302–317.

Rubert-Nason KF, Holeski LM, Couture JJ, Gusse A, Undersander DJ, Lindroth RL (2013) Rapid phytochemical analysis of birch (*Betula*) and poplar (*Populus*) foliage by near-infrared reflectance spectroscopy. Anal Bioanal Chem 405:1333–1344.

Sack L, Scoffoni C (2012) Measurements of leaf hydraulic conductance and stomatal conductance and their responses to irradiance and dehydration using the evaporative flux method (EFM). J Vis Exp 70: e14179.

Santesteban LG, Miranda C, Rojo JB (2011) Suitability of pre-dawn and stem water potential as indicator of vineyard water status in cv. Tempanillo. Aust J Grape Wine Res 17:43–51.

Santos AO, Kaye O (2009) Grapevine leaf water potential based upon near infrared spectroscopy. Sci Agric 66:287–292.

Sato T, Abdalla OS, Oweis TY, Sakuratani T (2006) The validity of predawn leaf water potential as an irrigation-timing indicator for field-grown wheat in northern Syria. Agric Water Manage 82:223–236.

Scholander PF, Bradstreet ED, Hemmingsen EA, Hammel HT (1965) Sap pressure in vascular plants. Negative hydrostatic pressure can be measured in plants. Science 148:339–346.

Serbin SP (2012) Spectroscopic determination of leaf nutritional, morphological, and metabolic traits. PhD dissertation, UW Madison, Madison, WI, USA.

Serbin SP, Dillaway DN, Kruger EL, Townsend PA (2012) Leaf optical properties reflect variation in photosynthetic metabolism and its sensitivity to temperature. J Exp Bot 63:489–502.

Serbin SP, Singh A, Desai AR, Dubois SG, Jablonski AD, Kingdon CC, Kruger EL, Townsend PA (2015) Remotely estimating photosynthetic capacity, and its response to temperature, in vegetation canopies using imaging spectroscopy. Remote Sens Environ 167:78–87.

Shenk J, Workman JJ, Westerhaus MO (2001) Application of NIR spectroscopy to agricultural products. In: Burns D, Ciurczak EW (eds) Handbook of near-infrared analysis. Marcel Dekker, New York, NY, pp 419–474.

Shetty N, Gislum R (2011) Quantification of fructan concentration in grasses using NIR spectroscopy and PLSR. Field Crop Res 120: 31–37.

Silva EN, Ferreira-Silva SL, Viégas RA, Silveira JAG (2010) The role of organic and inorganic solutes in the osmotic adjustment of drought-stressed *Jatropha curcas* plants. Environ Exp Bot 69:279–285.

Sims DA, Gamon JA (2003) Estimation of vegetation water content and photosynthetic tissue area from spectral reflectance: a comparison of indices based on liquid water and chlorophyll absorption features. Remote Sens Environ 84:526–537.

Sjöman H, Hirons AD, Bassuk NL (2015) Urban forest resilience through tree selection – variation in drought tolerance in *Acer*. Urban For Urban Greening 14:858–865.

Stimson HC, Breshears DD, Ustin SL, Kefauver SC (2005) Spectral sensing of foliar water conditions in two co-occurring conifer species: *Pinus edulis* and *Juniperus monosperma*. Remote Sens Environ 96:180–118.

Wold S, Ruhe A, Wold H, Dunn WJ (1984) The collinearity problem in linear-regression: the partial least-squares (PLS) approach to generalized inverses. SIAM J Sci Stat Comp 5:735–743.

Wold S, Sjostrom M, Eriksson L (2001) PLS-regression: a basic tool of chemometrics. Chemometr Intell Lab 58:109–130.

Zarco-Tejada PJ, Rueda C, Ustin SL (2003) Water content estimation in vegetation with MODIS reflectance data and model inversion methods. Remote Sens Environ 85:109–124.

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