

How do Plant Traits Affect Thermoregulation?

Student: Sabastian Escobar

Mentor: Benjamin Blonder

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Abstract

Plants are known to regulate their leaf temperature, undergoing limited homeothermy. This may be advantageous as it allows plants to utilize important photosynthetic and respiratory enzymes. We studied which traits affect a plant's ability to regulate their temperature across highly variable air temperatures, helping build better models to predict which species will have a higher performance across different environmental gradients with a changing climate. Using a thermal camera, we took diurnal videos for seven separate sites over a 1,401 meter elevation gradient. After the diurnal measurements, we took trait measurements that include: leaf area, leaf height, stomatal conductance, and wet and dry mass. Using MATLAB, we cross-referenced thermal images with visible images to find individual leaves in a given plot. We determined which traits were responsible for the greatest deviation in the leaf's temperature from ambient air temperature. The traits responsible for how quickly a plant can regulate its temperature can be found by finding the derivatives of air temperature versus time and the leaf temperature versus time functions and comparing them.

Introduction

For many years, biologists have considered a plant's leaf temperature to match ambient air temperature, classifying them as poikilotherms (Levitt 1980). However, there is evidence that plants experience limited homeothermy, (Mahan and Upchurch 1988), allowing them to regulate their temperature in a narrower range. A narrow range is beneficial because plants can passively regulate their temperatures to avoid unfavorable extremes (Leigh 2012). This allows for the employment of photosynthetic enzymes that would otherwise be denatured in high temperatures or non-reactive at lower temperatures. This may be advantageous as it would allow plants to maximize their instantaneous CO_2 assimilation rate as well as their CO_2 assimilation rate over

their lifetime (Michaletz 2015), potentially leading to an overall increase in both growth and fitness.

As the climate is warming, plant species are required to either acclimate, move, or adapt to novel conditions. While some species are currently moving up in elevation (Corlett 2013), plant functional trait adaptations will become necessary for many other species to persist in lower elevation sites. Certain leaf functional traits such as leaf area, height from ground, angle, fresh to dry mass ratio, and stomatal conductance may affect how plant species react with a variable environment and therefore can be used as predictors of which species will be more successful in a given micro-climate.

I hypothesize that all species of plants within a community will have leaves whose temperatures are warmer than ambient air temperature in both the mornings and evenings, but will have cooler leaves relative to the ambient air temperature in the afternoons. Individual species, however, will have temperature deviations that differ from one another. I predict that certain plant traits, as mentioned above, will increase the deviation of a species' leaf temperature relative to air temperature as well as increase the rate at which they react to variable atmospheric temperatures.

Methods

Diurnal Measurement

I observed six separate sites ranging from 2195m to 3596m in elevation with one plot at each site that is measured approximately one meter by one-meter square. Using two ladders for stabilization, we set up a FLIR A615 thermal imaging camera to maintain an overhead view. From this point of view, the camera took thermal (640 x 480 px) and visible pictures(1280 x 720 px) every five seconds. We simultaneously recorded atmospheric temperature, and for

calibration, blackbody reference temperature measurements at five minute intervals. Beginning at least fifteen minutes before local sunrise and ending at least fifteen minutes after local sunset, we collected one diurnal measurement per site. The images were imported into MATLAB version R2016a (MathWorks, Inc.) for further analysis.

Leaf Trait Measurements

On the day following each diurnal measurement, leaf trait measurements were collected for each species present. Each species had one or two individuals were measured with one to six leaves measured per individual. Leaves were chosen at random and had a visible image taken to identify using a Cannon 6D with 17-40mm f/4L lens set to 30 mm focal length. The leaf chosen had its height above measured in centimeters from its highest point as well as its angle rest. Stomatal conductance measurements were taken using a steady state porometer (LiCOR Li- 1600). Leaves chosen were taken back to the lab to analyze area using ImageJ and have wet and dry measurements recorded. Each leaf will have a stomatal peel applied to both top and bottom.

Image Analysis

Utilizing MATLAB, the diurnal thermal images were stitched together into a single thermal video and calibrated from raw radiance pixel counts to temperature in degrees Kelvin using a FLIR-supplied algorithm and duct tape reference temperature. Each visible picture that was taken to identify each leaf was then co-registered with this thermal video using a set of control points and projective image transformation. Using the visible image as a reference, a region of interest was delineated corresponding to a single identified leaf in the thermal video. A custom MATLAB program then extracted data that included median temperature of the leaf, atmospheric temperature, and time of day for each individual from the video.

Data Analysis

The first analysis conducted was a time versus temperature graph that will show how each different species reacted to the daily temperature variation. Each species' reaction to air temperature can be observed by taking the deviation from ambient air temperature. Five of the seven plant traits were then compared to each species' deviation from air temperature to determine which traits are more responsible for this deviation. In order to determine the rate at which leaf temperature reacts to air temperature, we compared the derivatives of both the function that represents the change in air temperature over time as well as the function that represents the change in leaf temperature over time. Then, by comparing these values using a linear regression, we determined which species of leaves are responding more quickly to the change in air temperature. By taking the value of $\frac{dT_{leaf}}{dt} - \frac{dT_{air}}{dt}$, and comparing it to five of the seven traits, the traits responsible for the variation in rates of change were calculated. A principal components analysis was conducted following the scaling of the data (centering by means and scaling by standard deviation) and both a scree and bi plot were plotted to determine the correlation between the independent variables. All unrelated variables were used as predictors in a multiple regression analysis between the traits and with either the leaf-air temperature deviation or the plant's rate of reaction to atmospheric temperature change as dependent variables. Bar plots were then used to summarize the proportional effect of each individual leaf trait (ie. The estimated slope of the scaled predictor variables).

Results

Due to time constraints, three of the six plots were analyzed for a total of 147 leaf samples from

26 species were analyzed out of 257 leaf samples from 40 species. After analyzing the thermal data in RStudio (version 3.3.1) for each leaf sample and comparing the linear regression models of each leaf trait against leaf-air temperature deviation, the data was scaled. The principal components analysis along with the scree and bi plot showed that the variables were required to explain 90% of the variance in data, suggesting that although there was slight correlation between certain leaf traits such as leaf angle and leaf height, the significance was low enough to conclude that all independent variables tested would be included in the multiple regression analyses.

The regression analysis for the five leaf traits plotted against the leaf-air temperature deviation resulted an R-squared value of 0.4819 and a P-value of 2.15×10^{-11} . The regression analysis for the leaf traits plotted against the plant's rate of reaction to variable air temperature resulted in an R-squared value of 0.2513 and a P-value of 9.33×10^{-5} . The bar Plots for both the leaf-air temperature deviation and the plant's rate of reaction yielded similar results. Both plots show that there is negative effect on these dependent variables by stomatal conductance, fresh to dry mass ratio, and leaf height. On the other hand, Leaf area and leaf angle have positive effects on the leaf-air temperature deviation as well as the plant's rate of reaction.

Discussion

The results for the five leaf traits plotted against leaf-air temperature deviation explains 48% of the total deviation and our low p-value states that there is a strong significant relationship between these five traits and the leaf-air temperature deviation. As leaf height above the ground has the largest proportional effect bar it can be concluded that leaf height has the greatest effect on the temperature deviation. As the leaf height above the ground increases, the total deviation

from atmospheric temperature for a leaf is lessened. This is potentially due to the lessened boundary layers, and therefore the increased environmental exposure.

When the leaf traits were plotted against the rate of reaction, 25% of the plant's ability to react to variance in atmospheric temperature was explained by these five leaf traits. A p-value of 9.33×10^{-5} expresses again a strong significance of relationship between these traits and the plant's rate of reaction. Leaf height again has the strongest proportional effect of the plant's rate of reaction. The negative relationship states that as the leaf's height above the ground increases, it's ability to react to atmospheric temperatures decreases. This can again be explained by decreased boundary layers and the leaf's overall exposure to harsher environmental conditions.

Although much of leaf-air temperature deviation and some of a plant's rate of reaction can be explained due to a combination of these five leaf traits, and in particular leaf height above the ground, there are still other factors that are affecting the deviation and the rate of reaction. Some of these factors may include wind velocity, stomatal ratio or relative humidity. By allocating more resources and time studying and analyzing these alternative traits, a plant's ability to thermoregulation can be better explained. Further studies of leaf traits at the microclimate level can yield a better understanding of how leaf traits are affecting a plant's ability to survive in a given community across variable environmental conditions.

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Figures

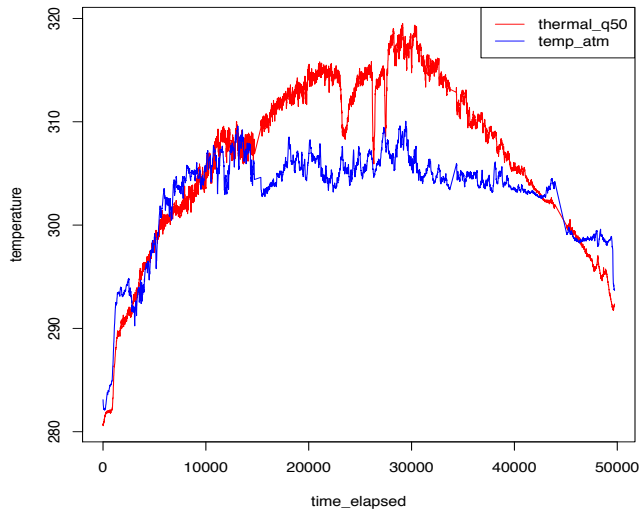


Figure 1. Time series graph that shows the air temperature (Kelvin) plotted next to the median temperature (Kelvin) of a VAL-2-3 as a function of time (seconds)

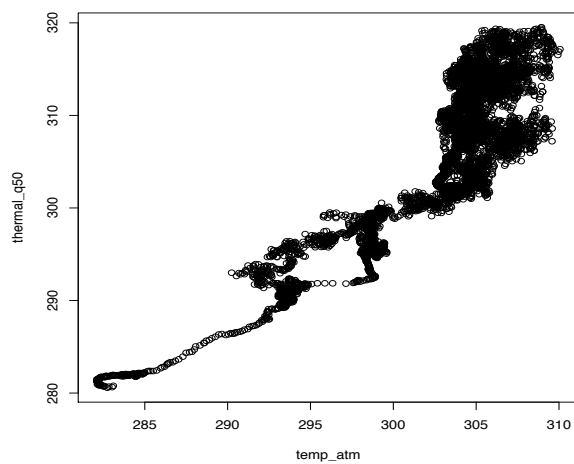


Figure 2. Atmospheric temperature (Kelvin) plotted against median leaf temperature (Kelvin)

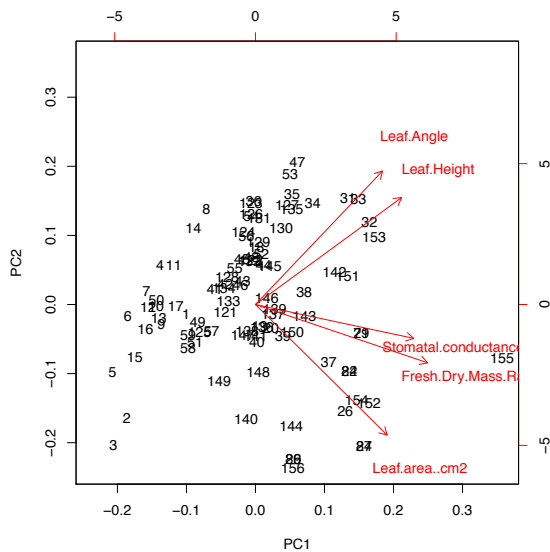


Figure 3. Bi-plot with scaled data to determine if any independent variables are too correlated to be used in the Multiple Regression Analysis.

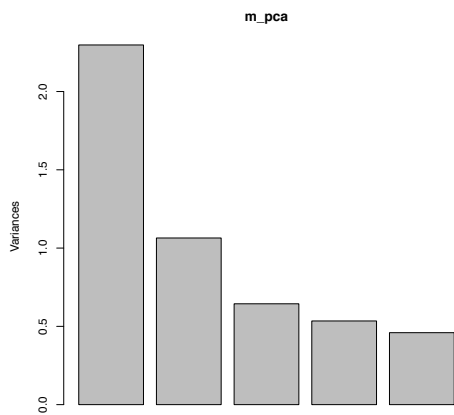


Figure 4. Scree plot comparing component variance against

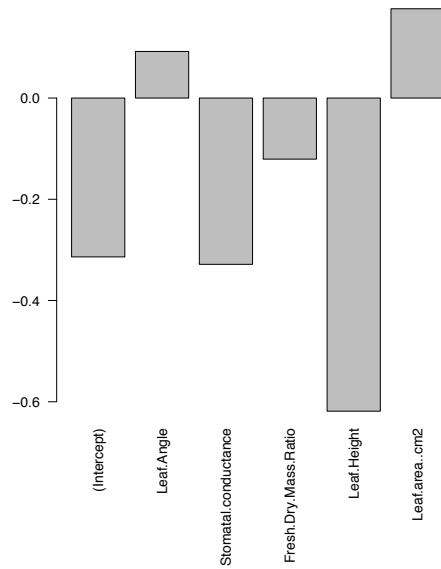


Figure 5. Bar graph comparing leaf traits and their proportional effects on leaf-air temperature deviation.

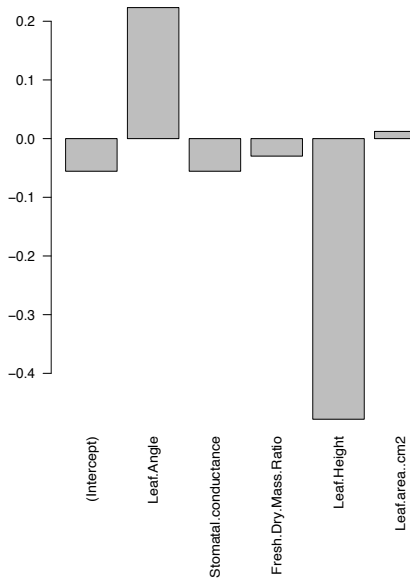


Figure 6. Bar graph comparing leaf traits and their proportional effect on how quickly a plant can react to changing atmospheric temperatures.

Site	Elevation (m)
Almont	2581
Crested Butte	2705
Road	2820
Pheiler	3175
Painter Boy Mine	3357
Baldy	3537

Figure 8. Site Elevations

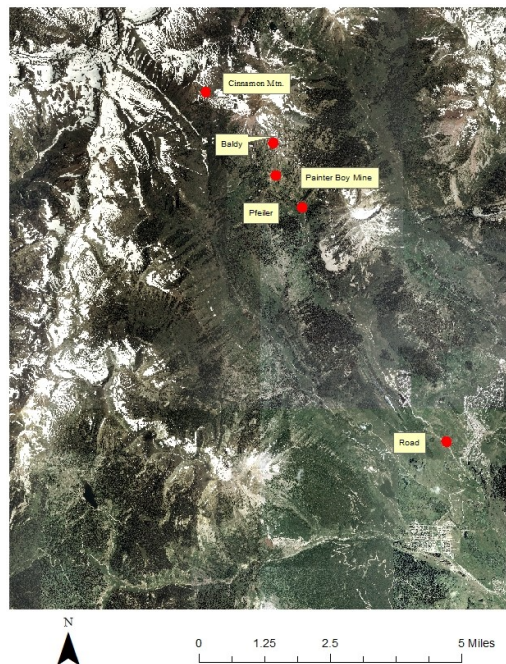


Figure 8. Long term Washington Gulch Sites (Crawford 2015)