

Title: Correlation of leaf and community traits and various spectra in meadows along an elevation gradient

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ABSTRACT:

The capabilities of remote sensing technology are advancing rapidly, and with it, the applications of those technologies are also advancing. Indices such as Normalized Difference Vegetation Index (NDVI) have long been used to determine vegetative health, but no direct measurements have been made to quantify communities until recently. Studies into the spectral properties of plants and ways in which to expand the quantifiable measurements done with hyperspectral imaging and increasing with the sophistication of imaging technology and modeling techniques. Partial Least Squares Regression (PLSR) is a technique that can pair spectral wavelengths with measured traits. With these relationships, we can measure traits directly from the spectral signature without the need of pervasive and destructive sampling techniques. The PLSR developed with this methodology is built using data from montane and subalpine meadows in the area of Gothic, Colorado, but can be tested on other vegetative communities to assess the model's resilience in more varied conditions. We used hyperspectral imaging with wavelengths from 400-1000 nanometers and collected leaf and community traits to construct the model. **describe your results and conclusions.**

INTRODUCTION:

Remote sensing is a field that has seen expansion from imagery attained using imagery flyovers to drone level and ground level imaging used to assess vegetative health. With an increased variety in available sensors and reduced cost of imaging apparatuses, the uses of remote sensing have expanded and with that expansion has come an increase in research as to what properties correspond with what wavelengths, a pursuit that is in need of further investigation.

This being the case, recent research has shown that wavelengths beyond this range are relevant to certain traits exhibited by plants. Gates et al. (1964) began to investigate the spectral properties of plants using a spectrophotometer, and he analyzed the impacts that age and plant type have on the spectral signature of a leaf. Carter & Knapp (2001) gave a review of the spectra at which plant stress can be observed, mainly in the area of 700 nm. It has been shown that peaks in 400-500 nm, 670-680 nm, and near-infrared (NIR) are typically lower in stressed leaves than in healthy leaves due to the absorbance by pigments at these wavelengths (carotenoids at 400-500 nm, chlorophyll at 670-680 nm, and water stress and anatomy changes in the NIR).

Since the advent of more sophisticated remote sensing technology, researchers have begun to develop indices, the most common and basic of which is NDVI. These indices can take many factors into account (i.e. L-ATSAVI which accounts for litter, vegetation, and soil in its equation) (He et al. 2006). These indices can be strong determinations of the relative health and stress of a community, but they are unable to develop direct measurements of traits on the community, plant, and leaf levels.

As direct measurement has become possible, there has been research into the wavelengths that are important to a variety of community-level and plant-level traits. Chavana-Bryant et al. (2016) found that leaf water content (LWC), leaf mass per unit area (LMA), and phosphorous were linked to tree age in a community of Amazonian trees by the spectra of the leaves measured using an ASD on a leaf to leaf basis. A similar method was used by Serbin et al. (2014) to pair LMA, nitrogen content, carbon content, fiber %, lignin %, and cellulose % with parts of the electromagnetic spectrum, namely Short-wave

Infrared (SWIR), visible (VIS), and near infrared (NIR). Both studies used a partial least squares regression (PLSR) approach (developed by Wold et al., 2001), which allowed the authors to calibrate variation in these chemical and physiological leaf traits to their spectral signatures. To directly measure these leaf traits can be time consuming, expensive, and difficult; many require the removal of leaves and other destructive measures, but with remote sensing and PLSR modelling techniques, pairing of spectral signatures and quantification of leaf traits and community traits, not indices, is now possible. In this study, we took a similar approach but rather than just focusing on leaf traits, we attempted to correlate community traits with the spectra as well, namely carbon flux and in turn, net primary productivity (NPP).

We predicted that using PLSR we will be able to determine what wavelengths are most indicative of the measured traits being collected on the leaf level and the community level (LMA, LWC, CO₂ Flux, NPP) and will be able to measure them remotely in future study. As these models are developed and rectified, they can be used at a variety of spatial and temporal scales, allowing researchers to better understand communities remotely, without even having to touch the ground.

METHODS:

Site Selection:

We selected sites for this study to include the elevation gradient previously established by researchers in the Enquist laboratory group and two of the Department of Energy (DOE) intensive sites. The sites exist along an elevation gradient of 2400 meters to 3600 meters (Figure 1). All sites are within 45 minutes of Gothic, CO and include the Washington Gulch, Slate River Valley, Almont, and Paradise Divide Areas. Each of the Enquist sites have five 1.5x1.5 meter plots along a microclimate and small elevational gradient and are placed haphazardly to encourage randomness. DOE plots have 6 large plots (15mx10m) on which a variety of research is occurring. Within these larger plots, we created two paired plots that are toward the middle of the plots' slope gradient. Most sites are dominated by meadow species such as *Veratrum californicum*, *Potentilla fruticosa*, *Festuca sp.*, *Chrysanthemum leucanthemum*, and *Taraxacum sp.* and peak photosynthetic conditions were sought in taking measurements.

Leaf Sampling:

Five leaves will be taken from the image frame and identified in the spectral image using a digital camera. In order to obtain LMA (eq. 1), leaves were weighed and scanned. After scanning, leaves were dried for 72 hours and weighed again to calculate LWC (eq. 2).

$$eq. 1: LMA = \frac{mass_{leaf}}{area_{leaf}}$$

$$eq. 2: LWC = \frac{fresh\ weight - dry\ weight}{dry\ weight}$$

CO₂ Flux Sampling:

The entire Enquist Lab is measuring net ecosystem productivity and net primary productivity using a soil flux chamber on a LiCOR 6400 and a flux tent and a LiCOR 7500. More detailed methods of these measurements can be read in Sloat et al (2015).

Hyperspectral Sampling:

Using an SOC 710 hyperspectral camera (Surface Optics Corporation, San Diego, CA), which measures 128 wavelengths from 400-1000 nm, images will be taken of the plots from a height of approximately 1.8 meters. A dark image was taken in order to control for noise in the image caused by temperature at the time of the measurement. All images are taken between 11AM and 1PM to attempt to attain peak light conditions. A 50% reflective standard will be used to standardize images based on ambient light conditions. Images were screened for static and waves which could be caused by wind or shaking of the camera.

Statistical Analysis:

Using the reference images and ENVI (Environment for Visualizing Images, Harris Geospatial Solutions, Boulder, CO) the sampled leaves were selected in the hyperspectral image as an ROI (region of interest). These were then loaded alongside the image and dark image into MATLAB code written by Jin Wu of the Brookhaven National Lab and edited by Alex Brummer of University of Arizona and myself (MATLAB, MathWorks, Natick, MA). This code outputs CSV files of the spectral information for each identified leaf and this data was then inputted into a PLSR model which will correlated measured traits to spectral wavelengths measured on the plants. The data was split 70% to build the model and 30% to test it.

RESULTS: Pending

As of the writing of this paper, we have used the leaf weights, both fresh and dry to calculate LWC and LMA (fig. 3 & 4). These traits saw slight variation between species, but many were of the same functional groups, and therefore the differences were not large. Community trait data has yet to be compiled and therefore was not included in the analysis addressed in this paper, but will be included in a future draft.

Spectral signatures for each leaf were graphed for visualization, (ex. Fig. 5) and also put into a *.csv file for analysis in the PLSR.

The PLSR produced best fit models using the optimal number of components, resulting in a correlation between the predicted values and the actual values (Fig. 6 & 7). These correlations were weak, but the eventual analysis will include two more sites and many more traits.

Revised Timeline:

6/24- begin taking images, collect leaves

7/6- begin analysis of early data points

7/18- take last measurements

7/25- submit final paper draft

8/1- do final analyses (PLSR)

7/29-8/2- revise and complete write-up

8/4- submit final copy of write-up

8/4- student symposium talk

DISCUSSION:

This paper is the beginning of a much larger project. In this paper, leaf water content and leaf mass per unit area were correlated with hyperspectral reflectance, but the correlation was not strong. Further analysis will include leaf carbon content, leaf nutrient content, and community traits as well as 2 more sites of leaf trait data. Although many studies have quantified leaf traits using this method (Serbin et al, 2014, Chavana Bryant et al, 2016), few have attempted to correlate spectra with community traits as we plan to. With this limited data and at this stage in the project, it is of great merit to discuss the larger goals associated with this research and to explain the future of this project.

Given the expected results of this product, we will have a model with which we can predict certain plant traits in the meadows around Gothic. This study is foundational to work being done by the DOE in the valley and will be used in possible aerial remote sensing in the valley. Once the model is built, it can be ground-truthed at more sites to test its applicability for a variety of environments. Expanding the use of PLSR in ecology will allow for similar studies to develop models for a variety of environments and then compared to each other. If they are rectified against each other, an ecotype specific or even eventual universal model could be constructed preventing the need for direct, destructive measurements and allowing for speedier and more varied measurements without large field crews.

As the field of remote sensing expands along with the field of landscape ecology, models such as those created in this project will become a crucial link between the observed world and measurable traits used to quantify that world.

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TABLES AND FIGURES:

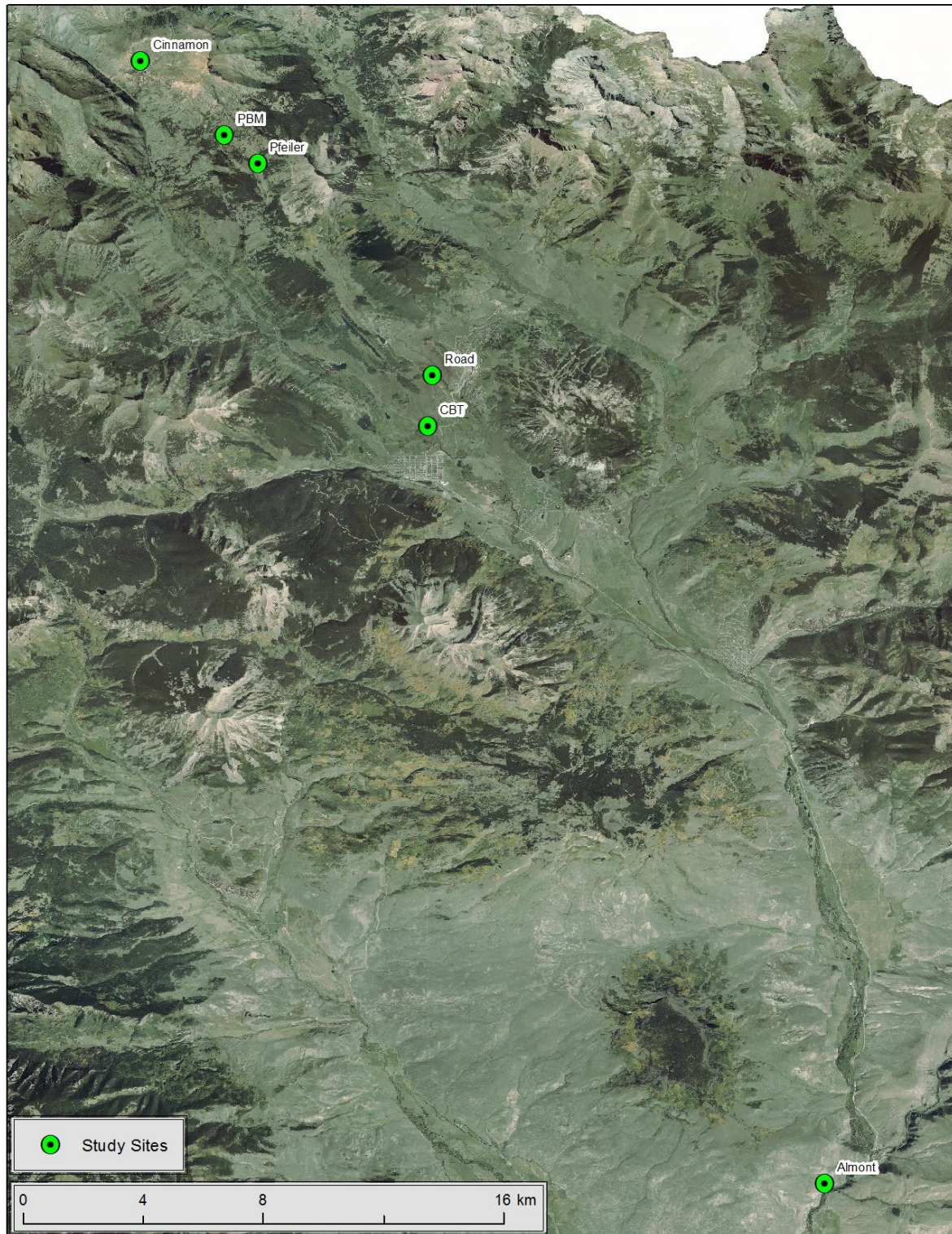


Figure 1: A locator map to show the location of study sites established by the Enquist Lab in relation to the greater Crested Butte, CO area. Aerial photo provided by RMBL.

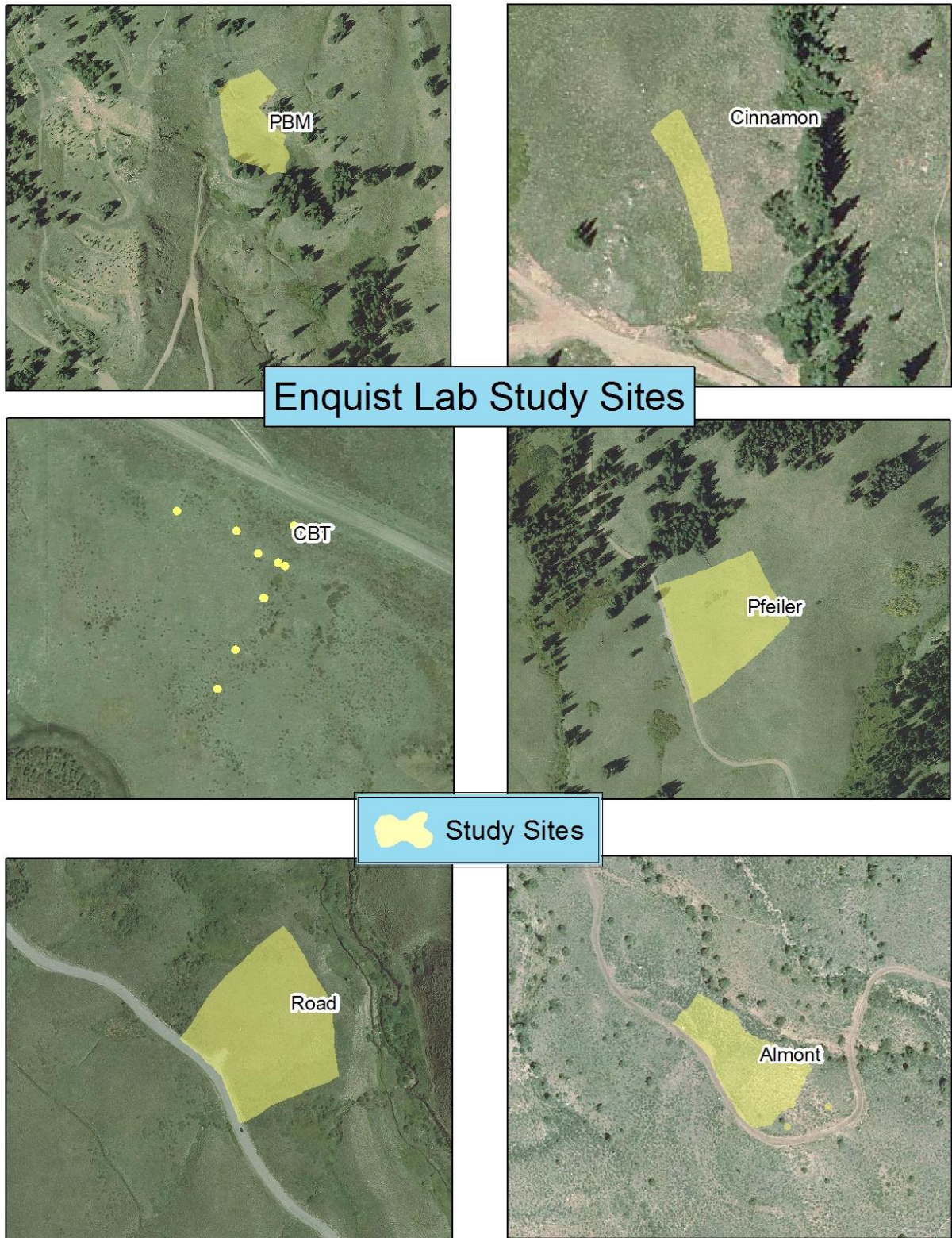


Figure 2: Location of study sites in relation to local roads. Polygons represent the general shape of sites used.

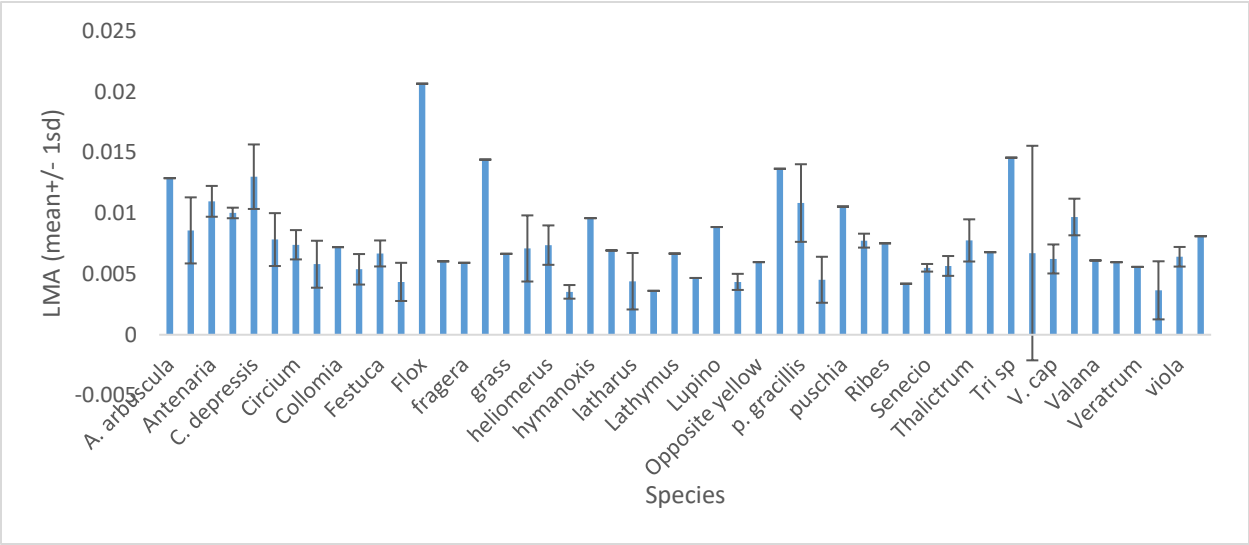


Figure 3: The average LMA per species sampled. Error bars represent one SD from the mean.

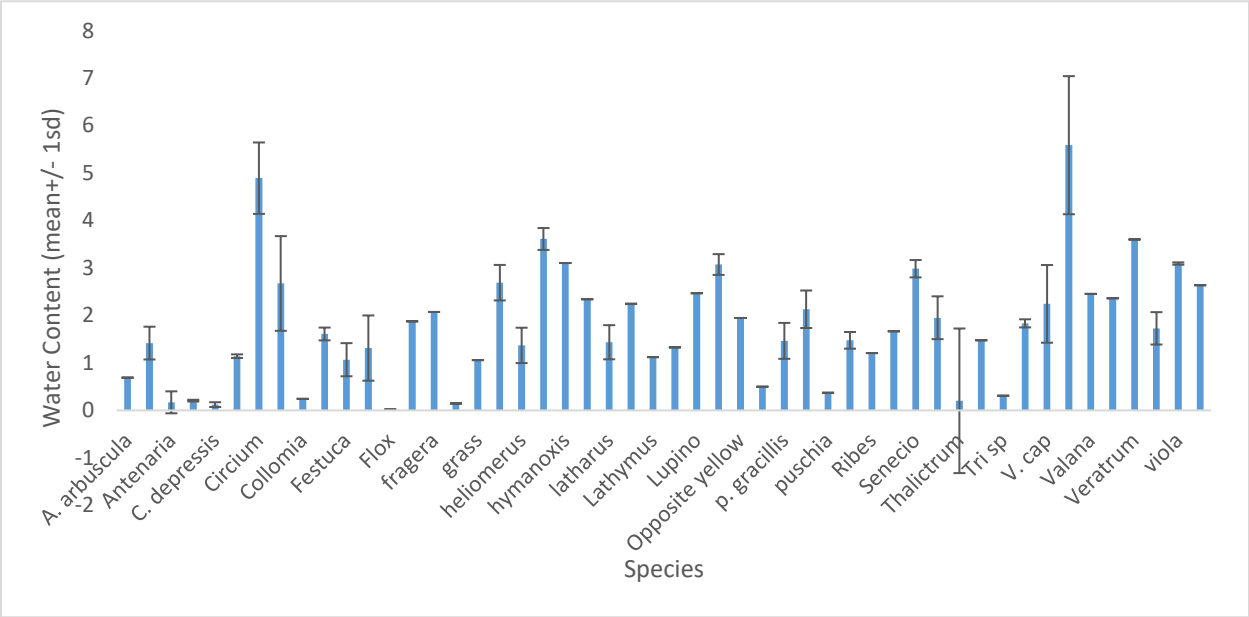


Figure 4: The average LWC per species sampled. Error bars represent one SD from the mean.

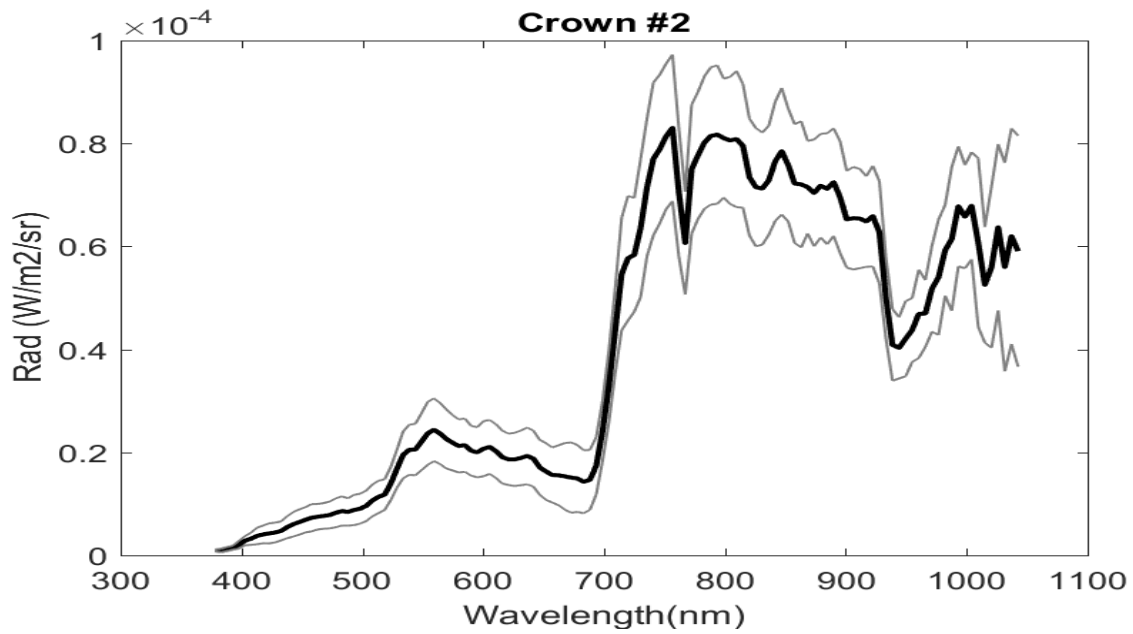


Figure 5: Example spectral reading of a leaf. Graph displays mean reflectance as a measure of radiance for the leaf and one standard deviation on the fainter line.

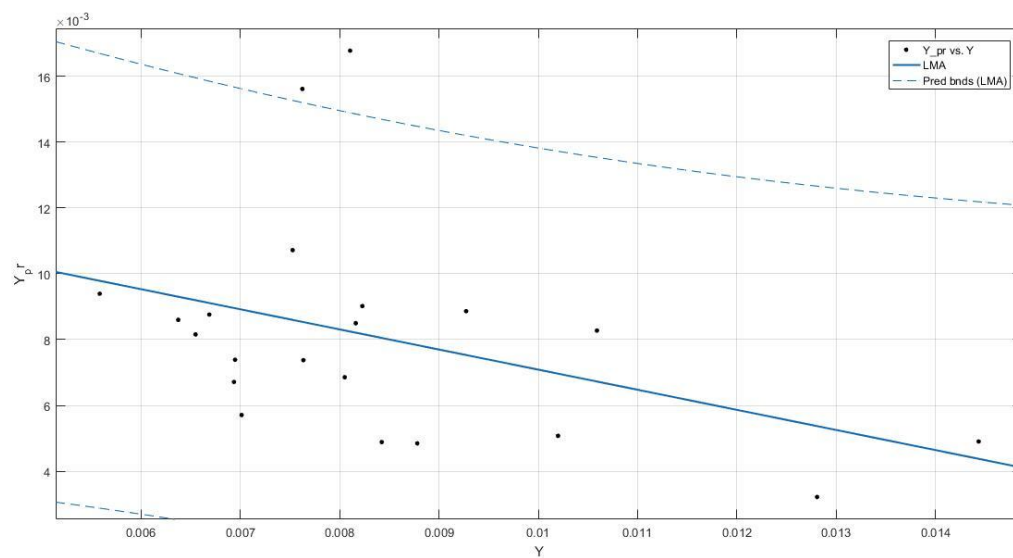


Figure 6: Graph displaying the fitness of the LMA model. Blue solid line represents a first degree polynomial that shows the line of best fit. The dashed lines show the 95% confidence intervals of that fit.

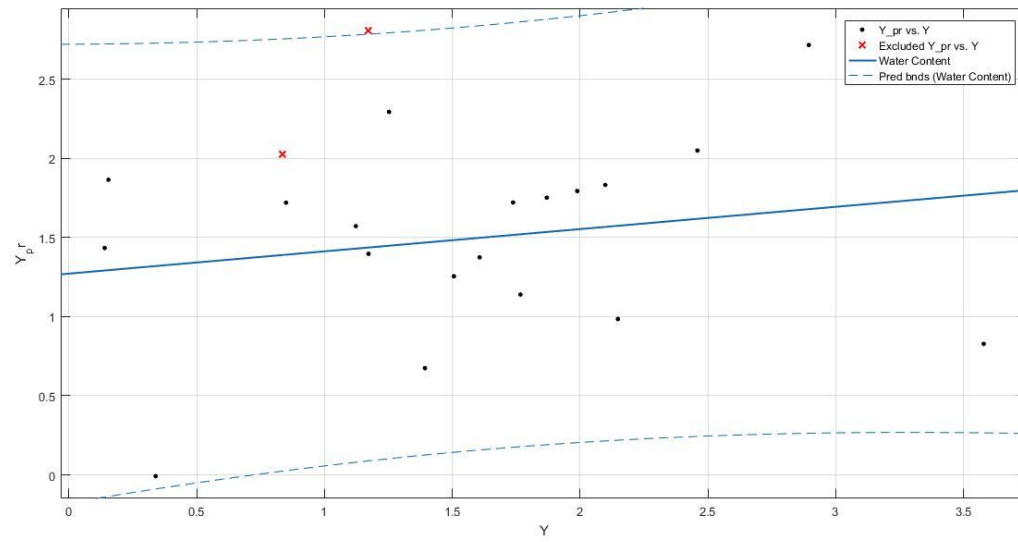


Figure 7: Graph displaying the fitness of the LWC model. Blue solid line represents a first degree polynomial that shows the line of best fit. The dashed lines show the 95% confidence intervals of that fit. Red X's are outliers in the data that were excluded from the fit.