USING AN UNMANNED AERIAL SYSTEM TO COLLECT MID-SEASON MULTISPECTRAL DATA FOR ESTIMATION OF PLANT NITROGEN STATUS IN COTTON David W. Daughtry II Wesley M. Porter Clondon H. Harris

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Abstract

Managing nitrogen (N) in cotton is critical for optimizing the ratio of vegetative and reproductive growth throughout the growing season, which maximizes the subsequent yield. At the field level, spatial variability of soil texture can lead to varying levels of nutrient uptake by the cotton crop. Unmanned Aerial Systems (UASs) have the ability to provide quick and efficient ways to detect spatial variability of N status to aid making in-season management decisions. This study was conducted during 2017 and 2018 on a research site in Tifton, GA. The main objective of the study was to correlate varying levels of plant tissue nitrogen (N) obtained from cotton tissue samples with vegetative indices (VIs) generated from multispectral imagery acquired with an UAS. Six N treatments consisting of 0, 34, 67, 101, 135 and 168 kg/ha rates were applied to attain varied levels of N in the tissue samples. Tissue samples were collected during the first, third, fifth, and seventh weeks of bloom to quantify N tissue levels temporally as a response to the applied N rate. Leaf blade and petiole tissue samples were collected and separated such that analyses provided leaf blade N (%) and petiole N (ppm). Multispectral imagery in the wavelengths of 550 nm (green), 660 nm (red), 735 nm (red-edge) and 790 nm (near infrared) was acquired during the cotton growth stages at the same time the tissue samples were collected by utilizing a commercially available quadcopter equipped with a high-resolution multispectral camera. Two vegetative indices (NDVI and NDRE) were analyzed for correlation with leaf blade and petiole tissue N levels at each sampling date and tracked temporally. Regression equations correlating the VIs to actual N levels were generated to evaluate the use of different VIs for accurately measuring N levels in the crop at the selected growth stages.

Introduction

The United States is the third largest cotton (Gossypium hirsutum L.) producer globally (Meyer, 2018) with over 4.3 million hectares of upland cotton harvested in 2017 (NCC 2017). Georgia is the second largest cotton producer in the U.S., with over 513,000 hectares harvested in 2017 (USDA 2018). Cotton tends to be grown in fields that have high variability in nitrogen (N) and other plant available nutrients (Torbett et al., 2008). The indeterminate growth habit of cotton causes it to be affected by varying levels of N (Reddy et al., 1997; Zhao et al., 2010). Insufficient N can reduce yield and quality, while an excess supply of N can negatively affect plant growth and development (Ballester et al., 2017; Porter et al., 2010). Reduced leaf size, number of fruiting nodes, boll retention, and reduction in yield can all be results of low N levels (Arnall et al., 2016; Bajwa et al., 2004). Whereas, excessive N can lead to excessive vegetative growth, increased fruit abscission, and reduced fiber quality and yield (Arnall et al., 2016; Zhao et al., 2010). The combination of spatial variability of N, cotton sensitivity to non-optimal N levels, and the challenges of detecting N levels in a timely fashion, make suppling the correct amount of N to the correct areas of a field at the correct time very challenging. Tissue testing can be used to measure leaf-blade N and petiole NO3-N which has aided in N status monitoring (Wood et al., 1992) that can be used to make application decisions. However, collecting tissue samples is time consuming and expensive and are subjective point samples used to interpolate cotton N status in the areas from which they were collected. Ground-based sensors have been used to assess the N status of plants without the requirement of physical tissue samples. The Soil-Plant Development (SPAD) Section of Minolta Camera developed a hand-held meter to measure leaf chlorophyll content by measuring the transmittance in two bands, (400 - 500 nm)and 600 – 700 nm) which provides a quick, non-destructive in situ assessment of leaf chlorophyll content (Feibo et al., 1997). Assessing the N status of cotton by taking leaf or canopy reflectance measurements has been well documented as a non-destructive sampling method in research (Arnall et al., 2016; Ballester et al., 2017; Raper and Varco 2014; Tarpley et al., 2000). Chlorophyll absorbs light in the visible region of the light spectrum (380 – 750 nm) for photosynthesis; therefore, leaf reflectance in that region is correlated to the concentration of chlorophyll (Buscaglia and Varco, 2002). The center of a chlorophyll molecule has four nitrogen surrounding a magnesium and therefore nitrogen content is directly related to chlorophyll concentration (Tarpley et al., 2000). Cotton leaf tissue N

concentration during the first square, first bloom, and mid bloom growth stages have shown high correlation to chlorophyll meter readings per the findings of Wood et al. (1992). Spectral data recorded from plants have been used to create different ratios or vegetative indices. A popular vegetative index is the normalized difference vegetation index (NDVI) which has been used to predict mid-season N requirements for cotton (Arnall et al., 2016; Porter et al., 2010). NDVI uses the near-infrared (NIR) and red spectra, combined into the equation (Arnall et al., 2016; Huang et al., 2013; Porter et al., 2010; Zhao et al., 2010):

$$NDVI = \frac{(NIR - Red)}{(NIR + Red)}$$

The strongest wavelength correlations with leaf N concentration, lint yield, and plant total N content were noted near 700 nm by Raper and Varco, (2014). N deficiency causes a decrease in leaf chlorophyll content, resulting in an increase in spectral reflectance between 550 nm and 710 nm (Zhao et al., 2010). Trimble's_® GreenSeeker[®] optical sensor has been developed to collect reflectance measurements in the red and near infrared bands (Arnall et al., 2016; Porter et al., 2010). The GreenSeeker[®] is one type of many sensors that can be used in remote sensing technology. Remote sensing technology can be used on many different platforms including ground-based platforms, satellites, planes, and unmanned aerial systems (UASs). N detection via ground-based methods can be time consuming. The use of manned aircraft is expensive and must be scheduled in advance; whereas, satellites often provide images with coarse spatial resolution and cannot collect crop imagery when fields are covered by clouds. UASs are much less expensive when compared to manned aircraft and can collect high resolution images under ubiquitous cloud cover. Recently, UAS research has become more popular and has the capability to identify field variability in a timely manner (Huang et al., 2013).

Objectives

The main objective of this study was to determine if controlled application rates of N fertilizer would provide detectable varied levels of leaf and petiole tissue nitrogen content which could be correlated with UAS based multispectral data. The secondary objectives of this study were to:

- 1. Track the changes of plant tissue N temporally during the season by N rate treatment
- 2. Determine the correlations between NDVI and NDRE and the N content of leaf and petiole tissue during critical N uptake growth stages
- 3. Determine the optimal timing for collecting UAS multispectral data for making in-season fertility decisions

Materials and Methods

This study was conducted during the 2017 and 2018 growing seasons utilizing Deltapine DPL 1646 B2XF planted in 4 row plots that were 3.7 m x 12.2 m under conventional tillage in Tifton, GA. Six N rate treatments of 0, 34, 67, 101, 135, and 168 kg N/ha were implemented. For each N treatment, ammonium nitrate (34-0-0) was used with one-third of the total rate applied one week after planting and the remaining two-thirds of each treatment applied during the 1st Square. Leaf blade and petiole samples were collected during the 1st square, 1st week of bloom (WOB), 3rd WOB, 5th WOB, and 7th WOB physiological stages. The most recently matured leaf and respective petiole for that leaf were collected for the samples. During 2017, only leaf blade samples were removed during the 1st square sampling date; however, all other sample dates during 2017 and 2018 had both leaf blade and petioles samples. The leaf and petiole samples were separated within one hour of collection and placed in paper bags in a forced air dryer for a minimum of 48 hours. The leaf blade and petiole samples were then sent to a private lab for nutrient analyses. Lint yield was obtained by harvesting the center two rows of each plot. The seed cotton was ginned at the University of Georgia MicroGin, lint turnout was calculated and utilized to calculate lint yield of each respective plot. Multispectral data were collected using a Sequoia multispectral sensor (Sequoia, Micasense Inc., Seattle, WA, USA) mounted on a 3DR Solo UAV (3D Robotics, Inc, Berkeley, CA). The four discrete spectral bands collected by the Sequoia were Green (550 nm, 40 nm wide), Red (660 nm, 40 nm wide), Red Edge (735 nm, 10 nm wide), and Near Infrared (790 nm, 40 nm wide). An autonomous flight plan was created and utilized for every flight for the entirety of each season by using the Tower app on an Android tablet. A radiometric calibration panel was used to collect calibration images with the Sequoia before and after each flight. The calibration images were used in Pix4Dmapper Pro (Pix4D SA, Switzerland) software along with all of the flight images to create a normalized reflectance orthomosaic image for each band. The orthomosaic image for each band was imported into ArcGIS Version 10.4.1 (ESRI, Redlands, CA) and then used to create NDVI and NDRE maps. A shape file was created around each plot that placed a polygon over all four rows. The VI data was clipped to each plot, averaged by plot, and exported in tabular format. Both of the VI data sets were

averaged by treatment and plotted separately with leaf blade and petiole N averages by treatment to obtain R² values using a linear regression model in Excel. The R² values for each sampling date were recorded and graphed over time.

Results and Discussion

The leaf blade N results from 2017 (Figure 1) show the changes in average leaf blade N % for each treatment over time. During the 1st square sampling date, the average leaf blade N % for all treatments was higher than the rest of the sampling dates and there was little difference in leaf blade N% individual treatments during this sample date. During the 1st WOB, the 0 kg N/ha rate provided the lowest leaf blade N % value and as N rate treatment increased, average leaf blade N% increased sequentially. Additionally, there was a clear separation between average leaf blade N% for each of the treatments for the 1st WOB sampling date. The 3rd, 5th, and 7th WOB samples followed a similar trend as the 1st WOB samples; however, the difference between treatments was not as pronounced as several treatments produced similar leaf blade N% averages on those sampling dates.



Figure 1. Average leaf blade nitrogen results from nitrogen treatments collected during critical physiological stages for the 2017 season.

The 2017 petiole N results (Figure 2) show changes in petiole N ppm for each treatment over time. Due to a lack of available tissue for collection for a representative sample, petiole samples were not collected during the 1st square growth stage of 2017. Similar to the leaf blade N results, during the 1st WOB, petiole N ppm was the lowest for the 0 kg N/ha treatment and increased sequentially by treatment with clear separation of treatments. A similar trend was observed at the 3rd WOB and the 5th WOB with the exception of the 0 kg N/ha and 34 kg N/ha treatments having no separation in the average petiole N ppm values during the 5th WOB. The 7th WOB provided little to no separation of the average petiole N ppm values for most of the treatments.



Figure 2. Average petiole nitrogen results from nitrogen treatments collected during critical physiological stages for the 2017 season.

The 2017 average lint yield by treatment graph (Figure 3), shows an increase in lint yield as N rate treatment increased with the exception of the 168 kg N/ha treatment that yielded slightly less than the 135 kg N/ha treatment. The 0 kg N/ha rate produced an average lint yield of 1170 kg/ha which can be attributed to residual N from peanuts being grown in this field in the 2016 season. Following peanut provides a N credit that reduces the N rate for cotton by up to 25% per the University of Georgia recommendations (Whitaker et al., 2018).



Figure 3. Average lint yield from nitrogen treatments for the 2017 season.

The coefficient of determination (Figure 4) for both NDVI and NDRE was low during the 1st square sampling date; however, a significant increase in correlation was observed for the subsequent sampling dates. During the 1st WOB sampling, NDVI had a stronger coefficient of determination than NDRE, which suggests that it is a better predictor of leaf blade N during the early bloom physiological stage. However, as the season progressed NDRE had a stronger coefficient of determination for the 3rd, 5th, and 7th WOB physiological stages. It should be noted that cotton biomass is correlated with N rate; thus, NDVI being a biomass sensitive VI, has a stronger correlation prior to canopy closure.

After canopy closure, NDRE has been shown to have a stronger correlation than NDVI, which was observed from the data of this study. A similar trend was observed in the petiole N data (Figure 5); albeit, the coefficient of determination was significantly weaker for NDVI at the 5th WOB and 7th WOB physiological stages. Petiole N can be susceptible to environmental conditions and boll load which may potentially weaken the coefficient of determination in these data.



Figure 4. Coefficient of determination over time for leaf blade nitrogen and VI averages for the 2017 season.



Figure 5. Coefficient of determination over time for petiole nitrogen and VI averages for the 2017 season.

The leaf blade tissue results from 2018 (Figure 6) show the changes in average leaf blade N % for each treatment over time. During the 1st square sampling date, the leaf blade N % averages were similar to the 2017 leaf blade N % averages increasing sequentially from the lowest N treatment to the highest N treatment. However, the 1st, 3rd, 5th, and 7th WOB leaf blade N % averages did not follow the same trends that were observed during 2017. During these crop stages, the leaf blade N % averages had slight separation, but did not have the significant separation in treatment that was observed during the 2017 season. Similarly, the 2018 petiole N results (Figure 7) had increasing petiole N averages as N rate increased during the 1st square sampling date with the exception of the 67 kg N/ha treatment, which produced a higher petiole N average than the 101 kg N/ha treatment. Similar to the 2018 leaf blade N % averages, the petiole ppm averages for the 1st, 3rd, 5th, and 7th WOB had little to no separation and did not follow the pattern that was observed in the 2017 data.



Figure 6. Average leaf blade nitrogen results from nitrogen treatments collected during critical physiological stages for the 2018 season.



Figure 7. Average petiole nitrogen results from nitrogen treatments collected during critical physiological stages for the 2018 season.

The average lint yield by treatment from the 2018 season (Figure 8) showed no significant response based on N rate treatment. The 0 kg N/ha rate treatment had a lower yield than the rest of the treatments, but overall there was little difference between treatment yields when compared to 2017. The 2018 treatment lint yield averages were higher than the 2017 treatment lint yield averages, suggesting that N was not a limiting factor for lint yield.



Figure 8. Average lint yield from nitrogen treatments for the 2018 season.

Summary

The results from this study showed that UASs have the potential to estimate in-season plant N status in cotton when varying levels of N are detectable and confirmed with tissue sampling. Data collected during 2017 showed a response to N treatments in both leaf blade and petiole tissue N averages with the highest level of separation occurring during the 1st WOB sampling date. The correlation during this physiological stage to multi-spectral imagery was strongest with NDVI. After the 1st WOB stage, NDRE produced a stronger correlation to the measured leaf blade and petiole tissue N averages. The correlations observed from this study suggest that VIs as predictors of N status in cotton should be selected based the physiological stage. The University of Georgia recommends that the optimal timing for sidedress applications of N is between the 1st square and 1st WOB stages; however, sidedress applications of soil applied N can be successfully applied until the 3rd WOB (Whitaker et al., 2018). Studies have shown that applications of soil applied N after the 3rd WOB have very little effect on lint yield (Wright et al., 2003). The 2017 data suggest that there is a relation between leaf blade N, petiole N, NDVI, NDRE, and subsequent lint yield. Therefore, utilizing UASs to collect mid-season multispectral data can potentially aid in making decisions for the application of soil applied N during the 1st and 3rd WOB when it can be beneficial in terms of added yield. Multispectral data and tissue data were not collected during the 2^{nd} WOB in this study; therefore, it is unknown at which point the coefficient of determination of NDRE surpasses the coefficient of determination of NDVI. The 2018 data did not follow the same trends as the 2017 in response to N treatments. Yield data from 2018 showed little to no difference between treatments supporting the finding that there were little to no difference in the tissue results by treatment. The data from both years will be further analyzed, which may produce causation for the observed variation between the two years' data. There are plans to conduct this study again during the 2019 season.

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